

The spawning frequency of sardine, *Sardina pilchardus* (Walb.), off the Atlantic Iberian coast

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ABSTRACT

Oocyte development for *Sardina pilchardus* was established, and a histological classification of postovulatory follicle aging was developed from the examination of laboratory-spawned sardine. The established criteria were compared with those used for field collections taken during the Portuguese-Spanish joint Egg Production Method Survey (March-May 1988).

The fraction of mature females with postovulatory follicles, < 48 hours old, for the Iberian Peninsula was 0.14 (standard deviation = 0.0122), indicating that the mean interval between spawnings was approximately 7 days.

Key words: Spawning fraction, sardine, Iberian Peninsula.

RESUMEN

Frecuencia de puesta de sardina, *Sardina pilchardus* (Walb.), en las costas de la península Ibérica.

Se determina para la especie *Sardina pilchardus* el proceso del desarrollo ovocitario, y se clasifica histológicamente las diferentes edades de degeneración de los folículos postovulatorios por medio del estudio de gónadas de sardinas inducidas artificialmente a la puesta. Los criterios establecidos se comparan con los utilizados en muestras procedentes de las campañas de investigación para el desarrollo del Método de Producción Diaria de Huevos, realizadas por Portugal y España conjuntamente (marzo a mayo de 1988).

Se determina la fracción de hembras maduras que ponen por día mediante el estudio de los folículos postovulatorios de edad menor de 48 horas, que en el caso de la sardina de la península Ibérica es de 0.14 (desviación típica 0.0122), indicando que el intervalo medio entre dos puestas parciales es de 7 días aproximadamente.

Palabras clave: Fracción de hembras reproductoras, sardina, península Ibérica.

INTRODUCTION

Sardine (*Sardina pilchardus*) spawn more than once within a spawning season, and present more than one mode of advanced oocytes in their active ovaries (Andreu and Pinto, 1957). Estimation of spawning frequency is essential to understand the reproductive biology of a species with an indeterminate annual fecundity. The standing stock of more advanced oocytes gives no

indication of annual fecundity, because new batches are regularly recruited from small unyolked oocytes during the spawning period (Hunter and Macewicz, 1985).

Spawning frequency and the number of eggs released per spawning are required to estimate annual reproductive effort or fecundity, and are also needed to find out how these variables are related to population size and age structure (Hunter, Macewicz and Sibert, 1986).

The first histological work on *Sardina pilchardus* was done by Pinto and Andreu (1956). By histological examination, they found that the fish length at which the sexual glands of sardine first become outlined within the coelom was 37 mm. They also found that sexual differentiation took place in larvae 45 mm long, and ovaries were completely organized in those 100 mm long.

This paper aims to describe the postovulatory follicle reabsorption process and to establish a criteria for determining follicle age in order to estimate the spawning frequency of *Sardina pilchardus*.

MATERIAL AND METHODS

The material used in this paper is based on sardine samples collected by pelagic and bottom trawls during the Daily Egg Production Method (DEPM) survey held March to May 1988 by both Spain (R/V Cornide de Saavedra) and Portugal (R/V Noruega) (see tables I to V), and on sardine spawned in captivity (12 °C, February 1989).

Adult sardines captured by commercial purse seine during February 1989 were induced to spawn in the laboratory using the technique developed by Leong (1971). Three sardines spawned: one, 215 mm total length (L_t) was sacrificed 19 h after spawning; the second, 175 mm L_t , died 34 h after spawning; and the third, 180 mm L_t , died 61 h after spawning. Their ovaries were used as a first approach for the establishment of the histological criteria for determining the age of sardine postovulatory follicles. The validated classification system obtained was compared with that developed for the sea-caught females sampled during the DEPM survey.

Ovaries were removed and preserved in two different fixatives: Spain, 10 % buffered formalin (NBF) according to Hunter (1985); and Portugal, acid-alcohol-formalin (AAF) after Bodian (1937) in Lillie and Fullmer (1976), modified. The second fixative presented a high penetration coefficient; even large ovaries were well and homogeneously fixed. Harris' hematoxylin and eosin (H & E) staining procedure was used.

Histological characteristics of oocytes

Oocyte development and maturation in teleosts has been frequently subdivided into many stages. Hunter and Macewicz (1985) proposed a simpler histological classification system in which they combined the stages defined by former authors into four oocyte stages (unyielded, partially yielded, yielded and hydrated). We adopted this classification system for our work.

Unyielded Oocytes

In this stage, smaller oocytes are distributed in an orderly manner along the ovarian septa and have no cytoplasmic inclusions. They generally exhibit a polygonal section, sometimes rectangular and rarely spheric or oval. They present a basophilic cytoplasm, very densely stained with hematoxylin, and a voluminous central nucleus containing several nucleoli. Later on, oocytes present two different chromatic affinities (H & E): the inward zone (perinuclear cytoplasm) lightly stained, and the outward zone, which presents the same staining as the smaller oocytes. These two groups of oocytes were classified by Andreu (1951) as α phase (< 100 μm). Macroscopically, they present a transparent appearance which corresponds to the immature or inactive stage (stage I in Andreu and Pinto, 1957).

Subsequently, the oocytes become more spherical, and the perinuclear zone less defined than in the previous groups. In oocytes with a larger diameter, the zona radiata appears between the oocyte and the growing follicle as a thin, faint eosinophilic membrane. The follicle begins to be visible in oocytes with diameters between 100 and 150 μm (Andreu, 1951) as a thin layer of elongated cells (the beginning of the granulosa cells).

Partially Yielded Oocytes

This stage involves oocytes in which vitellogenesis (yolk deposition) begins, determining a different staining reaction; the cytoplasm basophilic appearance progressively decreases. Oocytes become more spherical, and oil vacuoles migrate to the neighbor-

hood of the nucleus and become more voluminous. Delicate striations appear in the zona radiata. The follicle layer is thicker, and the differentiation between thecal and granulosa cells is clear.

Yolk deposits begin to form near the central cytoplasmic area as small spherules, while the remaining cytoplasm has a basophilic appearance. As vitellogenesis proceeds, larger globules spread into the oocyte limits when yolk granules reach the perinuclear zone.

Yolked Oocytes

Oocytes are more voluminous, and they all contain yolk granules throughout the region between their periphery and the perinuclear zone. Vitellogenesis continues, and yolk granules vary to large globules. Oil vacuoles fuse at the proximity of the nucleus to form a single oil droplet. The zona radiata stretches and the follicle now corresponds to a thin membrane that surrounds the oocyte. Yolked oocytes macroscopically present a yellowish appearance.

Hydrated Oocytes

The final stage of vitellogenesis corresponds to hydration, a rapid uptake of the fluid by the follicle (Fulton, 1898, in Hunter and Macewicz, 1985). Hydration seems to begin when the nucleus has completed its migration to the animal pole and, simultaneously, yolk globules begin to fuse, forming yolk plates. The enlargement of the oocyte, which reaches about 1 mm diameter, causes the flattening of the thecal and granulosa cells as well as the thinning of the zona radiata.

Spawning Frequency

Two methods have been developed for measuring the spawning rate of multiple spawning fish (Hunter and Macewicz, 1985). Sardine spawning frequency was determined using the postovulatory follicle method. This method requires a division of the processes of deterioration and resorption of

the follicles into a series of distinct histological stages, each one with a specific age assigned (Hunter and Macewicz (1985)).

Since in 1988 no validation system existed to classify sardine postovulatory follicles by age, the histological characteristics of the deteriorating follicles, the time of sampling, and the average population peak spawning time was used to assign ages to the postovulatory follicles. In February 1989 an artificially induced spawning technique was used to obtain the time after spawning, i.e. age, of sardine postovulatory follicles. These ages were then used to confirm the histological criteria applied to the postovulatory follicles of sea-caught females sampled in the DEPM survey during 1988 (Olmedo *et al.*, 1990).

The classification system for *Sardina pilchardus* postovulatory follicle ages is described below. Results for Portugal and Spain are shown in tables VI to X. Despite the relatively few numbers of immature females present in the samples in two western Cantabrian stations, immature females represented almost 72 % of total females sampled (table VII), which may be related to a differential distribution of immature and mature shoals during the spawning period.

The incidence of alpha-atresia increases in more degenerated postovulatory follicles.

Age 0-h postovulatory follicles

Since no laboratory female was sacrificed at the moment of spawning, the description of age 0-h follicles corresponds to ovaries with both new postovulatory follicles and hydrated eggs from sea-caught females. Cord-like cell layers of collapsing follicles form loose folds or loops at ovulation (fig. 1).

Age 19-h postovulatory follicles

After spawning, fully collapsed postovulatory follicles form a much more tightly folded or looped structure. It is relatively large, irregular in shape, and has an irregular lumen. The thecal cell layer is very thin

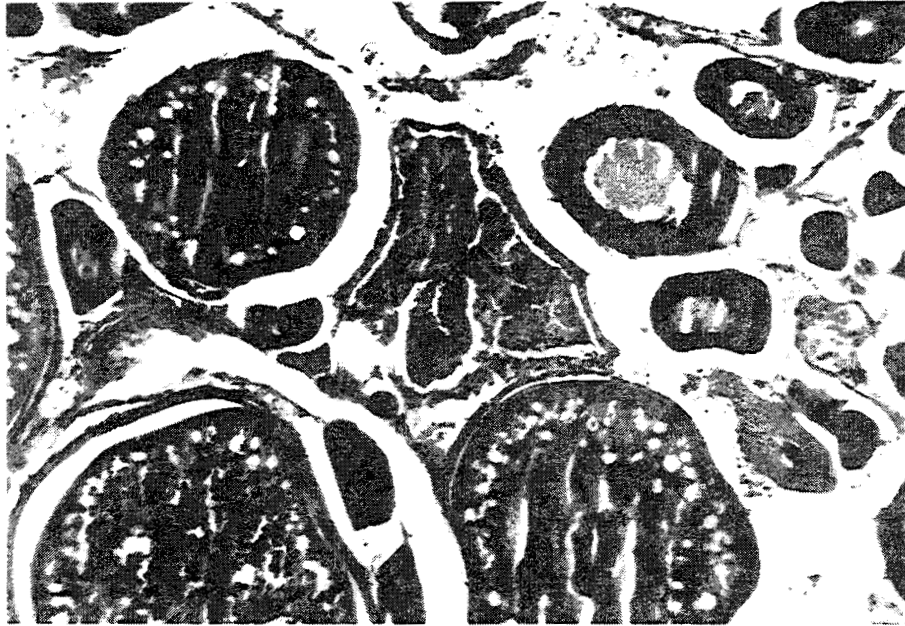


Fig. 1.—Postovulatory follicle, age Day-0. Elapsed time from spawning, about 0-5 h.

Fig. 1.—Folículo postovulatorio de 0 días de edad. Tiempo transcurrido desde la puesta de 0 a 5 horas.

and clearly distinguishable from the granulosa layer. The granulosa cells are polygonal and arranged in an orderly manner

along the lumen edge. The granulosa nuclei are evident, healthy, and linearly arranged (fig. 2).

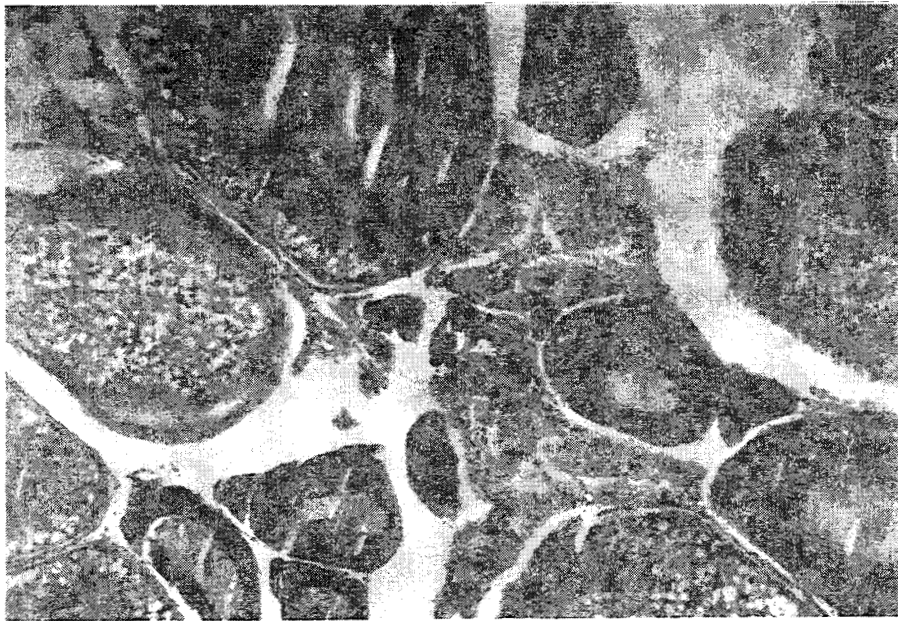


Fig. 2.—Postovulatory follicle, age 19 h.

Fig. 2.—Folículo postovulatorio de 19 horas de edad.

Age 34-h postovulatory follicles

The follicular structure (fig. 3) tends to become more compact and shows a less irregular shape than the follicle previously described. The follicle layer presents fewer folds and loops. The thecal layer is still no-

derly manner, and present pycnotic nuclei. The lumen is very reduced. The number of postovulatory follicles per section appears to diminish. According to Hunter and Macewicz (1985), this reduction may be related to resorption or, alternatively, to the growth of the larger oocytes in the ovary.

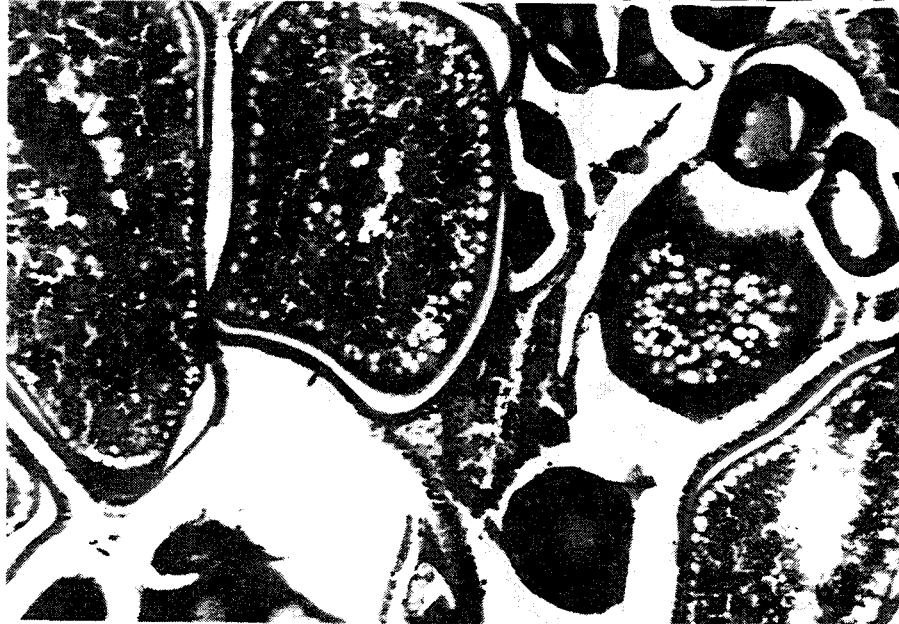


Fig. 3.—Postovulatory follicle, age 34 h.

Fig. 3.—Folículo postovulatorio de 34 horas de edad.

ticeable and well differentiated from the granulosa layer, and contains groups of blood cells. The lumen is much reduced compared to the previous stage. The arrangement of granulosa cells is less orderly and, in a small number of them, pycnotic nuclei are present.

Age 61-h postovulatory follicles

The regressing follicle is more shrunken and has fewer folds than the 34-h follicle (fig. 4). Despite this, identification and classification is still possible. The theca layer is still present but, in some cases, it is indistinguishable from the granulosa layer. The granulosa cells are not arranged in an or-

Older than 61-h postovulatory follicles

The comparative analysis of postovulatory follicles from laboratory-spawned females and sea-caught females confirms that the degenerative processes of sardine follicles last beyond 61 hours after spawning. Thus, it was possible to differentiate another distinct stage of postovulatory follicle, with an age greater than 61 hours. This group can be characterized by a major reduction in their structure and occurrence. Generally, the follicle presents a triangular shape and the theca layer is totally indistinguishable from the granulosa layer. The lumen is very reduced or absent.

The persistence of postovulatory follicles in the sardine ovary longer than in the



Fig. 4.—Postovulatory follicle, age 61 h.

Fig. 4.—Folículo postovulatorio de 61 horas de edad.

northern anchovy (*Engraulis mordax*) or the Peruvian anchovy (*Engraulis ringens*) but similar to duration in the Pacific sardine, may be related to differences in water temperature.

RESULTS AND DISCUSSION

The application of the Egg Production Method to the Atlantic Iberian sardine, *Sardina pilchardus*, required the estimation of the spawning fraction. Since spawning fraction is a daily estimate, the postovulatory follicle ages analyzed were grouped into four Day classes: Day-0 is ages from 0 to 6 hours after spawning; Day-1 is ages 18 to 30 hours; Day-2 is ages 42 to 54 hours; and Day-3 is ages older than 60 hours (sampling was not on a 24-hour basis; therefore, the "Days" reflect only ages actually encountered). It was decided that the estimation of the proportion of females spawning per day and per trawl should be based on the number of mature females whose postovulatory follicles were classified as Day-1 or Day-2,

i.e., $(\text{Day-1} + \text{Day-2})/2$ Pérez *et al.*, 1992). Day-3 follicles were not used since they presented ageing difficulties.

In Galicia (Spain), an oversampling of females with Day-0 follicles was observed, biasing the spawning fraction estimate (Pérez *et al.*, 1992). Notwithstanding, in Portugal the fraction of Day-0, Day-1 and Day-2 spawners did not reveal significant differences among them, indicating no bias in the Day-0 proportion (Cunha *et al.*, 1992). As a consequence, since Day-0 females can be overrepresented in the trawl stations, these females were not included in the spawning fraction estimate. The adjusted spawning fraction estimates by region along the Atlantic Iberian coast are summarized in table XI.

The low spawning fraction value for Galicia compared to the other regions was in agreement with sardine adult and egg distributions along the Atlantic Iberian coast. Along the Atlantic Iberian coast, the fraction of mature female sardine with less than 48-hour-old postovulatory follicles averaged 0.14 (standard deviation = 0.0122). This in-

licated that the mean interval between each spawning was approximately 7 days.

and Technological Cooperation and from the Junta Nacional de Investigação Científica e Tecnológica of Portugal.

ACKNOWLEDGEMENTS

This work was supported by funds from the U.S.-Spain Joint Committee for Scientific

Table I.—Characteristics of the 12 collections of female sardine taken along the Galician coast during the DEPM Cruise (April-May 1988). (BT): bottom trawl.

Tabla I.—Características de las 12 colecciones de hembras de sardinas capturadas a lo largo de las costas de Galicia en la campaña del DEMP (abril-mayo 1988). (BT): arrastre demersal.

Collection number	Date (yyymmdd)	Time of day (hh.mm)	Average ovary-free weight (g)	Gear	Latitude N	Longitude W
1	880401	17.52	60.713	BT	42° 33.2'	08° 55.7'
2	880401	19.55	60.136	BT	42° 31.4'	08° 57.5'
3	880402	17.22	66.910	BT	42° 45.3'	09° 01.1'
6	880406	20.44	25.208	BT	42° 21.9'	08° 50.7'
8	880408	16.59	62.500	BT	43° 18.9'	08° 56.1'
12	880409	20.45	79.400	BT	43° 24.7'	08° 31.1'
13	880409	22.06	59.711	BT	43° 23.9'	08° 34.2'
14	880410	01.12	47.956	BT	43° 24.7'	08° 31.7'
19	880412	19.28	81.509	BT	43° 49.9'	07° 55.1'
20	880412	20.45	83.280	BT	43° 49.1'	07° 55.0'
21	880413	18.20	77.452	BT	43° 41.8'	07° 15.7'
22	880413	20.47	56.050	BT	43° 35.1'	07° 02.3'

Table II.—Characteristics of the 11 collections of female sardine taken along the western Cantabrian coast during the DEPM Cruise (April-May 1988). (BT): bottom trawl.

Tabla II.—Características de las 11 colecciones de hembras de sardinas capturadas a lo largo de las costas del Cantábrico occidental en la campaña del DEMP (abril-mayo 1988). (BT): arrastre demersal.

Collection number	Date (yyymmdd)	Time of day (hh.mm)	Average ovary-free weight (g)	Gear	Latitude N	Longitude W
24	880415	19.34	63.708	BT	43° 35.7'	06° 42.8'
25	880416	13.53	81.326	BT	43° 41.8'	06° 19.9'
26	880419	17.52	63.400	BT	43° 40.0'	06° 15.7'
28	880421	21.02	78.450	BT	43° 46.0'	05° 42.0'
30	880422	20.44	90.347	BT	43° 38.6'	05° 34.6'
31	880423	20.13	84.280	BT	43° 36.8'	05° 23.7'
32	880424	22.57	39.400	BT	43° 30.1'	04° 59.8'
33	880424	19.18	99.324	BT	43° 32.1'	04° 55.8'
35	880426	21.38	84.624	BT	43° 28.7'	04° 35.3'
36	880427	18.00	93.640	BT	43° 26.7'	04° 23.5'
37	880428	20.05	88.300	BT	43° 27.6'	04° 09.5'

Table III.—Characteristics of the 7 collections of female sardine taken along the eastern Cantabrian coast during the DEPM Cruise (April-May 1988). (BT): bottom trawl.

Tabla III.—Características de las 7 colecciones de hembras de sardinas capturadas a lo largo de las costas del Cantábrico oriental en la campaña del DEMP (abril-mayo 1988). (BT): arrastre demersal.

Collection number	Date (yyymmdd)	Time of day (hh.mm)	Average ovary-free weight (g)	Gear	Latitude N	Longitude W
38	880430	08.54	76.200	BT	43° 28.8'	03° 21.4'
39	880501	01.26	78.764	BT	43° 31.9'	03° 43.6'
40	880501	18.38	89.912	BT	43° 31.2'	03° 23.4'
41	880501	16.21	90.482	BT	43° 25.6'	03° 06.7'
42	880502	19.22	83.946	BT	43° 28.2'	02° 55.2'
43	880504	14.55	87.644	BT	43° 21.4'	02° 17.7'
44	880504	18.42	95.386	BT	43° 22.0'	02° 15.7'

Table IV.—Characteristics of the 7 collections of female sardine taken in the northern region of Portugal (north of Nazaré) during the DEPM Cruise (March 1988). (BT): bottom trawl and (PT): pelagic trawl.

Tabla IV.—Características de las 7 colecciones de hembras de sardinas capturadas a lo largo de la zona norte de Portugal (norte de Nazaré) en la campaña del DEMP (marzo 1988). (BT): arrastre demersal y (PT): arrastre pelágico.

Collection number	Date (yyymmdd)	Time of day (hh.mm)	Average ovary-free weight (g)	Gear	Latitude N	Longitude W
4	880301	21.30	40.24	BT	41° 35.2'	09° 01.1'
6	880303	03.32	17.55	BT	41° 13.6'	08° 56.2'
8	880303	23.37	32.42	BT	41° 09.0'	08° 56.3'
9	880306	10.16	46.58	BT	41° 02.6'	08° 45.5'
10	880306	13.42	48.47	PT	40° 57.3'	08° 54.0'
12	880307	16.30	19.94	PT	40° 21.1'	09° 03.5'
13	880307	20.24	26.14	BT	40° 35.1'	09° 02.1'

Table V.—Characteristics of the 2 collections of female sardine taken in the southern region of Portugal (south of Nazaré) during the DEPM Cruise (March 1988). (BT): bottom trawl and (PT): pelagic trawl.

Tabla V.—Características de las 2 colecciones de hembras de sardinas capturadas a lo largo de la zona sur de Portugal (sur de Nazaré) en la campaña del DEMP (marzo 1988). (BT): arrastre demersal y (PT): arrastre pelágico.

Collection number	Date (yyymmdd)	Time of day (hh.mm)	Average ovary-free weight (g)	Gear	Latitude N	Longitude W
1	880227	12.45	22.87	PT	38° 38.0'	09° 30.0'
18	880325	21.58	48.75	BT	38° 22.5'	08° 58.9'

Table VI.—Number of female sardine in different spawning and atresia states along the Galician coast during the DEPM Survey (April-May 1988). Legend: atresia states. (0): no alpha- α atresia of yolked oocytes, (1): alpha- α atresia in < 50 % of yolked oocytes and (2): alpha- α atresia in > 50 % of yolked oocytes.

Tabla VI.—Número de hembras de sardina en los diferentes estados de puesta y atresia obtenidas en aguas de las costas de Galicia durante la campaña del DEMP (abril-mayo 1988). Leyenda: estados de atresia. (0): no aparece atresia tipo α en los ovocitos vitelados, (1): atresia α aparece en menos del 50 % de los ovocitos vitelados y (2): atresia α aparece en más del 50 % de los ovocitos vitelados.

Collection number	Atresia state	Day-0	Day-1	Day-2	Nonspawning females	Tot. mature females	Tot. immature females
1	0	0	2	0	6	25	0
	1	0	0	0	0	0	
	2	0	0	0	0	0	
Total		0	2	0	6	25	
2	0	16	1	2	6	25	0
	1	0	0	0	0	0	
	2	0	0	0	0	0	
Total		16	1	2	6	25	
3	0	0	0	0	2	25	0
	1	0	0	0	0	0	
	2	0	0	0	0	0	
Total		0	0	0	2	25	
6	0	0	0	0	3	3	0
	1	0	0	0	0	0	
	2	0	0	0	2	2	
Total		0	0	0	5	5	
8	0	0	0	1	0	1	0
	1	0	0	0	0	0	
	2	0	0	0	0	0	
Total		0	0	1	0	1	
12	0	0	0	0	0	0	0
	1	0	0	0	1	1	
	2	0	0	0	0	0	
Total		0	0	0	1	1	
13	0	0	0	1	14	15	1
	1	0	0	0	1	1	
	2	0	0	0	2	2	
Total		0	0	1	17	18	
14	0	0	0	2	10	12	1
	1	0	1	0	1	2	
	2	0	0	0	1	1	
Total		0	1	2	12	15	

Table VI.—(Cont.)

Tabla VI.—(Cont.)

Collection number	Atresia state	Day-0	Day-1	Day-2	Nonspawning females	Tot. mature females	Tot. immature females
19	0	20	0	0	0	24	0
	1	1	0	0	0	1	
	2	0	0	0	0	0	
	Total	21	0	0	0	25	
20	0	24	1	0	0	25	0
	1	0	0	0	0	0	
	2	0	0	0	0	0	
	Total	24	1	0	0	25	
21	0	0	0	4	13	17	0
	1	0	0	0	3	3	
	2	0	0	1	4	5	
	Total	0	0	5	20	25	
22	0	1	0	2	10	13	0
	1	0	0	0	2	2	
	2	0	0	0	9	9	
	Total	1	0	2	21	24	

Table VII.—Number of female sardine in different spawning and atresia states along the western Cantabrian coast during the DEPM Survey (April-May 1988). Legend: atresia states. (0): no alpha- α atresia of yolked oocytes. (1): alpha- α atresia in < 50 % of yolked oocytes and (2): alpha- α atresia in > 50 % of yolked oocytes.

Tabla VII.—Número de hembras de sardina en los diferentes estados de puesta y atresia obtenidas en aguas de la costa oeste del Cantábrico durante la campaña del DEPM (abril-mayo 1988). Leyenda: estados de atresia. (0): no aparece atresia tipo α en los ovocitos vitelados, (1): atresia α aparece en menos del 50 % de los ovocitos vitelados y (2): atresia α aparece en más del 50 % de los ovocitos vitelados.

Collection number	Atresia state	Day-0	Day-1	Day-2	Nonspawning females	Tot. mature females	Tot. immature females
24	0	0	0	0	3	3	18
	1	0	0	0	4	4	
	2	0	0	0	0	0	
	Total	0	0	0	7	7	
25	0	0	0	1	6	23	1
	1	0	0	0	1	1	
	2	0	0	0	0	0	
	Total	0	0	1	7	24	

Table VII.—(Cont.)

Tabla VII.—(Cont.)

Collection number	Atresia state	Day-0	Day-1	Day-2	Nonspawning females	Tot. mature females	Tot. immature females
26	0	0	0	0	0	0	1
	1	0	0	0	0	0	
	2	0	0	0	0	0	
Total		0	0	0	0	0	
28	0	0	3	0	11	14	1
	1	0	0	0	1	1	
	2	0	0	0	0	0	
Total		0	3	0	12	15	
30	0	2	1	7	15	25	0
	1	0	0	0	0	0	
	2	0	0	0	0	0	
Total		2	1	7	15	25	
31	0	0	2	6	13	21	0
	1	0	0	2	2	4	
	2	0	0	0	0	0	
Total		0	2	8	15	25	
32	0	0	2	0	4	6	18
	1	0	0	0	1	1	
	2	2	0	0	0	0	
Total		2	2	0	5	7	
33	0	0	2	7	12	23	0
	1	0	0	0	2	2	
	2	0	0	0	0	0	
Total		0	2	7	14	25	
35	0	5	3	1	8	17	0
	1	0	0	0	0	0	
	2	0	0	0	0	0	
Total		5	3	1	6	17	
36	0	0	1	3	15	20	0
	1	0	1	1	3	5	
	2	0	0	0	0	0	
Total		0	2	4	18	25	
37	0	1	0	0	0	1	0
	1	0	0	0	0	0	
	2	0	0	0	0	0	
Total		1	0	0	0	1	

Table VIII.—Number of female sardine in different spawning and atresia states along the eastern Cantabrian coast during the DEPM Survey (April-May 1988). Legend: atresia states. (0): no alpha- α atresia of yolked oocytes, (1): alpha- α atresia in < 50 % of yolked oocytes and (2): alpha- α atresia in > 50 % of yolked oocytes.

Tabla VIII.—Número de hembras de sardina en los diferentes estados de puesta y atresia obtenidas en aguas de la costa este del Cantábrico durante la campaña del DEPM (abril-mayo 1988). Leyenda: estados de atresia. (0): no aparece atresia tipo α en los ovocitos vitelados, (1): atresia α aparece en menos del 50 % de los ovocitos vitelados y (2): atresia α aparece en más del 50 % de los ovocitos vitelados.

Collection number	Atresia state	Day-0	Day-1	Day-2	Nonspawning females	Tot. mature females	Tot. immature females
38	0	0	0	0	1	1	0
	1	0	0	0	0	0	
	2	0	0	0	1	1	
Total		0	0	0	2	2	
39	0	5	5	4	4	18	0
	1	0	2	4	1	7	
	2	0	0	0	0	0	
Total		5	7	8	5	25	
40	0	0	5	3	12	22	0
	1	0	0	1	2	3	
	2	0	0	0	0	0	
Total		0	5	4	14	25	
41	0	0	7	2	7	24	0
	1	0	1	0	0	1	
	2	0	0	0	0	0	
Total		0	8	2	7	25	
42	0	0	5	6	11	23	0
	1	0	0	1	1	2	
	2	0	0	0	0	0	
Total		0	5	7	12	25	
43	0	0	3	2	2	7	0
	1	0	1	1	0	2	
	2	0	0	0	0	0	
Total		0	4	3	2	9	
44	0	0	3	4	14	21	0
	1	0	0	0	3	3	
	2	0	0	0	0	0	
Total		0	3	4	17	24	

Table IX.—Number of female sardine in different spawning and atresia states in the northern region of Portugal (north of Nazaré) during the DEPM Survey (March 1988). Legend: atresia states. (0): no alpha- α atresia of yolked oocytes, (1): alpha- α atresia in < 50 % of yolked oocytes and (2): alpha- α atresia in > 50 % of yolked oocytes.

Tabla IX.—Número de hembras de sardina en los diferentes estados de puesta y atresia obtenidas en aguas de Portugal (norte de Nazaré) durante la campaña del DEMP (marzo 1988). Leyenda: estados de atresia. (0): no aparece atresia tipo α en los ovocitos vitelados, (1): atresia α aparece en menos del 50 % de los ovocitos vitelados y (2): atresia α aparece en más del 50 % de los ovocitos vitelados.

Collection number	Atresia state	Day-0	Day-1	Day-2	Nonspawning females	Tot. mature females	Tot. immature females
4	0	3	11	6	9	33	0
	1	0	0	0	4	5	
	2	0	0	0	3	3	
Total		3	11	6	16	41	
6	0	0	0	0	10	10	0
	1	0	0	0	2	2	
	2	0	0	0	1	1	
Total		0	0	0	13	13	
8	0	2	1	1	19	21	1
	1	0	1	0	0	1	
	2	1	0	0	2	3	
Total		3	2	1	21	25	
9	0	0	3	0	7	10	3
	1	0	0	0	0	0	
	2	0	0	0	0	0	
Total		0	3	0	7	10	
10	0	0	1	5	16	22	1
	1	0	0	0	1	1	
	2	0	0	0	2	2	
Total		0	1	5	19	25	
12	0	0	4	1	14	19	0
	1	0	1	0	4	5	
	2	1	0	0	1	2	
Total		1	5	1	19	26	
13	0	0	3	5	10	22	1
	1	0	0	0	2	2	
	2	0	0	0	2	2	
Total		0	3	5	14	26	

Table X.—Number of female sardine in different spawning and atresia states in the southern region of Portugal (south of Nazaré) during the DEPM Survey (March 1988). Legend: atresia states. (0): no alpha- α atresia of yolked oocytes, (1): alpha- α atresia in < 50 % of yolked oocytes and (2): alpha- α atresia in > 50 % of yolked oocytes.

Tabla X.—Número de hembras de sardina en los diferentes estados de puesta y atresia obtenidas en aguas de Portugal (sur de Nazaré) durante la campaña del DEMP (marzo 1988). Leyenda: estados de atresia. (0): no aparece atresia tipo α en los ovocitos vitelados, (1): atresia α aparece en menos del 50 % de los ovocitos vitelados y (2): atresia α aparece en más del 50 % de los ovocitos vitelados.

Collection number	Atresia state	Day-0	Day-1	Day-2	Nonspawning females	Tot. mature females	Tot. immature females
1	0	0	2	1	6	15	0
	1	1	0	1	1	7	
	2	0	0	0	1	1	
Total		1	2	2	8	23	
18	0	3	2	1	4	10	0
	1	1	1	1	1	4	
	2	0	0	0	0	0	
Total		4	3	2	5	14	

Table XI.—Adjusted estimates of the spawning fraction in the different regions of the Atlantic Iberian coast. Standard deviations in parentheses.

Tabla XI.—Estimaciones de la fracción de hembras reproductoras en las diferentes regiones de la costa atlántica de la península Ibérica. Desviación típica, entre paréntesis.

SPAIN			PORTUGAL		TOTAL
E Cant.	W Cant.	Galicia	North	South	
0.21 (0.0273)	0.13 (0.0143)	0.08 (0.0160)	0.13 (0.0357)	0.12 (0.0908)	0.14 (0.0122)

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Received October, 1991. Accepted September, 1992