REPRODUCTIVE MODALITY AND BATCH FECUNDITY OF THE EUROPEAN HAKE (*MERLUCCIUS MERLUCCIUS* L.) IN THE BAY OF BISCAY

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ABSTRACT

Appraisal of reproductive modality is necessary to quantify the reproductive potential of a fish species. We believe that European hake, *Merluccius merluccius* (L. 1758), may have an indeterminate fecundity modality because of (1) a continuum of oocyte sizes in the ovary of mature female hake during the whole reproductive season, i.e., no gap in the oocyte frequency distribution between immature and vitellogenic oocytes; (2) the maintenance of a similar diameter of vitellogenic oocytes throughout the spawning season; (3) a steady level of remnant total fecundity during the spawning cycle; and (4) the prevalence of atresia at the end of the spawning period.

Batch fecundity of hake in 1994 averaged 165 eggs g^{-1} (SE = 9.89) gutted female weight. Spawning frequency, based on the duration of different ovarian developmental stages, ranged from 0.052 to 0.189, which is equivalent to a batch interval of 19 to 5.3 days. Finally, a quantification of the per batch and annual reproductive output was made.

INTRODUCTION

The literature on European hake indicates that this species spawns several times in the reproductive season, and therefore is a fractional spawner (Andreu 1955; Pérez and Pereiro 1985; Sarano 1986).

Past reproductive studies on hake do not indicate if annual fecundity is determined at the onset of spawning or is variable, with the standing stock of yolked oocytes at the beginning of the spawning season being unrelated to annual fecundity. Potential annual fecundity is defined as the total number of advanced-yolked oocytes matured per year, uncorrected for atretic losses (Hunter et al. 1992). After correcting for atretic losses, the total number of eggs spawned per female in a year is called the realized annual fecundity. In fishes with determinate fecundity, the standing stock of yolked oocytes prior to the onset of spawning is considered to be equivalent to the potential annual fecundity. This decreases with each spawning because the standing stock of yolked oocytes is not replaced during the spawning season.

The term *indeterminate* refers to species in which potential annual fecundity is not fixed before the onset of spawning (Hunter et al. 1992). In such species, previtellogenic oocytes can develop and be recruited into the yolked oocyte stock at any time during the season (*de novo* vitellogenesis; Hunter and Goldberg 1980). Estimation of total fecundity in the ovary prior to the onset of spawning is meaningless if, during the spawning season, oocytes are recruited to that stock. In such species, the annual fecundity should be estimated from the number of oocytes released per spawning (batch fecundity), the percentage of females spawning per day (spawning frequency), and the duration of the spawning season (Hunter et al. 1985).

The objective of this study was to identify the fecundity modality of the European hake and to estimate its potential annual reproductive output. Four lines of evidence (Hunter et al. 1989; Greer Walker et al. 1994) were investigated to assess the fecundity modality of European hake:

1. Stage-specific variation of oocyte size-frequency distribution: A distinct hiatus separating the yolked-oocyte stock from the unyolked stock indicates that annual fecundity is determinate, whereas the lack of a hiatus may indicate that annual fecundity is indeterminate.
2. Seasonal decline in total fecundity: A decrease in the stock of vitellogenic oocytes during the spawning season supports evidence for determinate fecundity.
3. Seasonal increase in the mean diameter of the advanced vitellogenic oocytes: Fishes with determinate fecundity will show an increase in the average diameter of yolked oocytes because no new yolked oocytes are recruited to replace those that have been spawned during the season.
4. Incidence of atresia during the spawning season: Fishes with indeterminate fecundity show a generalized prevalence of atresia and resorption of mature oocytes at the end of the spawning season (West 1990).

MATERIALS AND METHODS

Mature hake were sampled aboard commercial fishing vessels working in the Bay of Biscay (fig. 1) in 1994 and 1995, during the main reproductive season of hake in the area (January to May; Martin 1991). No samples

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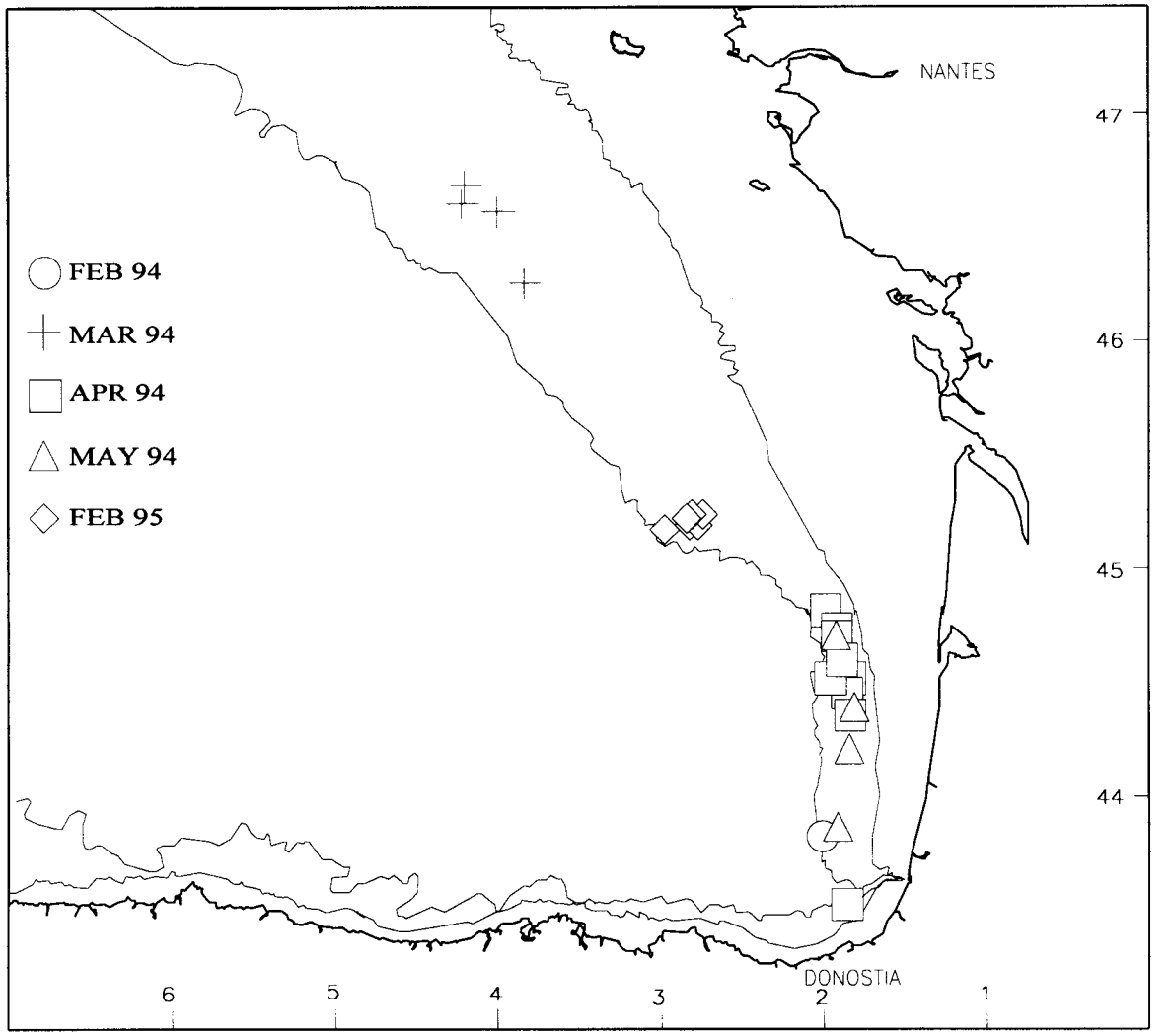

Figure 1. Location of fishing stations for gonad sampling in the Bay of Biscay. Samples were collected by longline (February 1994) or by pelagic trawl (March–May 1994 and February 1995).

were collected at the beginning (January) or at the end of the spawning season (June–July).

Mature females were selected at random from each trawl catch. All individuals were sexed, and total length was measured to the nearest 0.1 cm. Gonads were extracted and preserved in 4% buffered formaldehyde (Hunter 1985). The preserved ovaries provided material for histological descriptions (Hunter et al. 1985), oocyte size-frequency distributions, and estimates of fecundities. A total of 272 gonads of mature female hake were collected in 1994, and 79 gonads in 1995. Females ranged in total length from 40 to 102 cm in 1994 and from 41 to 82 cm in 1995.

We calculated total weight and gutted weight for each fish by using the length–total weight and total weight–gutted weight relationship given by Martin (1991).

Each ovary was classified histologically according to the most advanced oocyte stage present in the ovary. We followed Wallace and Selman's (1981) criteria for oocyte staging, with some modifications for hake ovarian structure. The diameter sizes for the different oocyte stages were measured only from oocytes that had been sectioned through the nucleus. The presence or absence of all the oocyte and postovulatory follicle stages was recorded.

Total fecundity (F_t) is defined as the total number of vitellogenic oocytes (>150 μm) in the ovary. In all of the 27 ovaries collected for total fecundity assessment, histological analysis identified postovulatory follicles, indicating that these females had already spawned. Consequently, our estimates of total fecundity should be regarded as estimates of the total remaining fecundity or remnant fecundity. These estimates were carried out with the gravimetric method (Hunter et al. 1989).

Batch fecundity (F_b) was determined by the gravimetric, hydrated-oocyte method (Hunter and Goldberg 1980; Hunter et al. 1985). When applying the gravimetric

method, we did not evaluate the possible effect of spatial differences of oocyte density on the fecundity estimates.

The fraction of females spawning daily was assessed from the prevalence of the different spawning stages (Hunter and Goldberg 1980; Hunter and Macewicz 1985).

RESULTS

Stages of Oocyte Development

According to the classification by Wallace and Selman (1981), hake show "asynchronous ovaries" where oocytes of all stages are present without dominant populations. The different stages are a consequence of a continuous development process, since the cellular events of oocyte growth do not sequentially replace one another, but rather are initiated sequentially and remain active throughout oocyte development (Selman and Wallace 1989).

The criteria for spawning stages are described below. We found that oocytes smaller than 150 μm contained no yolk and may constitute a reserve fund for next year. Vitellogenesis starts at 150 μm and develops into the nuclear migratory stage (750 μm). The last stage of maturation—hydration (Fulton 1898)—is easily recognized by the translucent appearance of the oocytes (table 1).

Ovulation follows hydration and results in ruptured, empty postovulatory follicles and the release of the oocyte, which is now considered an egg. New postovulatory follicles are readily identifiable, but they degenerate afterwards (fig. 2). Four stages of postovulatory follicle (POF) deterioration were recorded: (1) The structure of the follicle is very well maintained with no signs of deterioration; the granulose and thecal nuclei are clearly distinguishable. (2) The first signs of vacuolation and nuclear pycnosis appear, but the follicle is still well recognizable. (3) The follicle is being resorbed; the granulose and thecal nuclei cannot be differentiated; the structure is disorganized; and vacuoles are abundant. (4) The follicle has shrunk substantially and become a very small structure, which will eventually disappear.

Reproductive Modality

Oocyte size distribution. The pattern of development of mature hake ovaries is shown in figure 3. Hake ovaries show a continuous distribution of oocyte sizes throughout the stages of mature gonad development. Several modes representing spawning batches appear. No hiatus can be observed between the unyolked (<150 μm diameter) and the vitellogenic oocytes. The only observable hiatus is between yolked oocytes and the mode of mature hydrated oocytes just before ovulation and is typical of partial or fractional spawners (Hunter and Goldberg 1980; West 1990).

The sequence of events in ovaries within the (partial) spawning cycle is the same from February until May.

TABLE 1
Development Stages of Oocytes

Development stage	Characteristics	Diameter (mm) Oocyte	Diameter (mm) Nucleus
Chromatin nuclear	A large nucleus surrounded by a thin layer of cytoplasm. The nucleus contains a large nucleolus, and also a series of very small peripheral nucleoli. The oocyte is surrounded by a few squamous follicle cells.		<0.045
Perinucleolar	Bigger nucleus with several big peripheral nucleoli. Some vacuoles appear in the cytoplasm. The chorion precursor material begins to appear in patches.		0.045–0.07
Vitellogenic (yolked)			
Cortical alveoli formation	Small yolk vesicles start to appear in the cytoplasm. Oil droplets begin to accumulate in the cytoplasm. The chorion and follicle layers are apparent.	0.15–0.25	0.07–0.10
VIT 1	Oil droplets occupy more cytoplasmic area than yolk granules; yolk granule size of 0.003–0.075 mm.	0.25–0.45	0.10–0.12
VIT 2	Oil droplets occupy a similar cytoplasmic area to yolk granules; yolk granule size 0.075–0.125 mm.	0.45–0.55	0.12–0.15
VIT 3	Oil droplets occupy less cytoplasmic area than yolk granules; yolk granule size 0.125–0.20 mm.	0.55–0.65	0.15–0.20
Maturation			
Early migration	Oil droplets fuse into a unique oil globule, and the nucleus starts to migrate peripherally. Size of yolk granules: 0.20–0.25 mm.	0.65–0.75	0.20–0.25
Late migration	Nuclear migration continues; yolk granules fuse into plates starting in the center and extending centrifugally. Size of yolk granules: >0.25 mm.	0.75–0.95	
Hydration	Yolk has fused into a homogeneous mass. The nucleus has disintegrated. The cytoplasm and the cortical alveoli are restricted to a thin peripheral layer.	0.95–1.15	

Summary of oocyte developmental stages in European hake ovaries. The histological characteristics and the size ranges are given for each stage. Measures are made from histological sections.

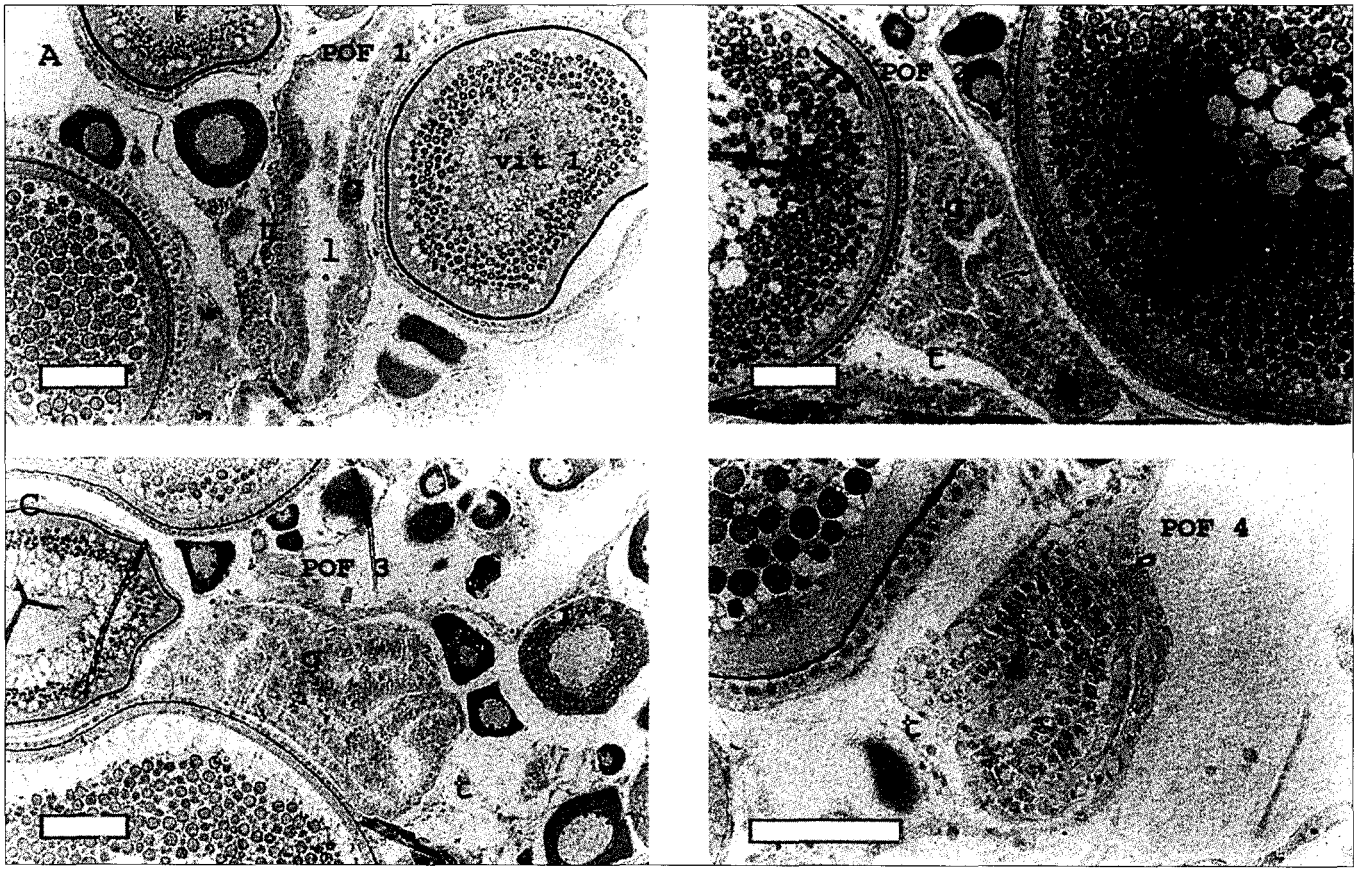

Figure 2. Postovulatory follicle stages: upper left, POF 1 (200×); upper right, POF 2 (200×); lower left, POF 3 (200×); lower right, POF 4 (400×). *t* = thecal connective cell layer; *g* = granulosa epithelial cell layer; *l* = lumen of follicle; bar = 0.1 mm.

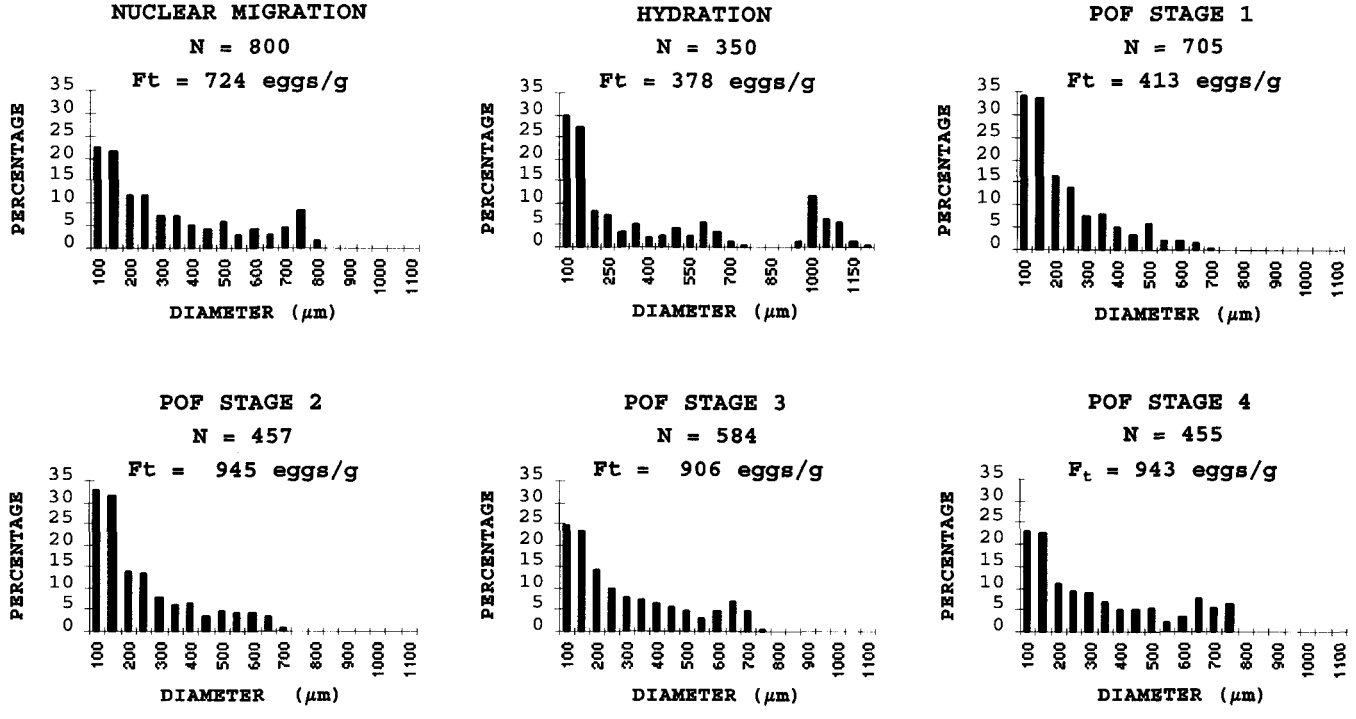

Figure 3. Evolution of oocyte size-frequency distribution (% abundance per 0.05 mm size class) through different spawning stages.

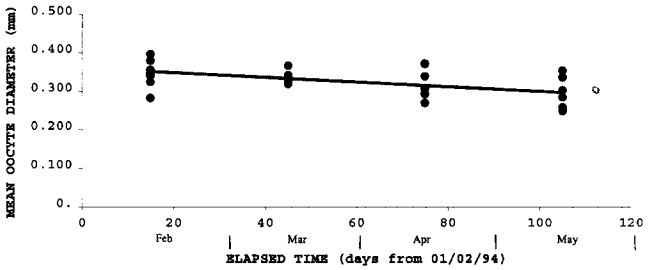

Figure 4. Relation between mean diameter (MD) of the advanced vitellogenic oocytes (excluding hydrated oocytes) and elapsed time, expressed as MD (mm) = 0.3609 − 0.0006 T (days); R^2 = 0.2683; p = 0.004; N = 29.

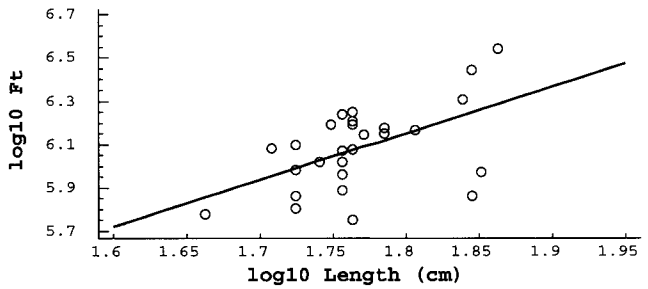

Figure 5. Relation between total (remnant) fecundity and total length for 27 active females collected in 1994.

Mean diameter. The mean diameter of the stock of yolked oocytes greater than 150 μm, but not including hydrated oocytes, decreased steadily from February to May (fig. 4). The points fit to a linear regression had a significant negative slope ($\alpha < 0.05$, $r^2 = 0.2683$). Certainly the mean diameter of the yolked oocytes did not increase during the reproductive season, one of the criteria used by Hunter et al. (1992) for determinate fecundity.

Total (remnant) fecundity. Twenty-seven females from 1994 were analyzed (fig. 5) for total fecundity. The total number of yolked oocytes (>150 μm) in the ovaries (F_t) of European hake in the main spawning season increased linearly with female length (L).

$$\log 10\ F_t = 2.292 + 2.144 \log 10\ L,\ n = 26,\ R^2 = 28.1\%$$

Total relative fecundity (number of vitellogenic oocytes per g gutted weight) ranged from 298 to 1,606 eggs g^{-1}, for an average of 957 eggs g^{-1} (CV = 34%).

A linear regression of fecundity on female weight for the elapsed time did not have a significant slope, indicating that total fecundity did not decrease substantially during the spawning season (fig. 6). An ANOVA on total fecundity using elapsed time as a main factor did not show any significant difference ($\alpha < 0.05$). Length was used as a covariant in this analysis to allow for the increasing relation between total fecundity and months ($p = 0.2517$).

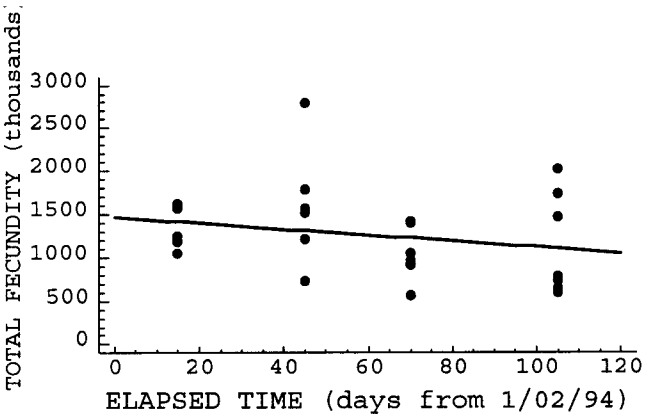

Figure 6. Relation between total fecundity and elapsed time expressed as F_t = 1465.3 − 3.43 T (days); R^2 = 5.23; N = 27.

Atresia. The incidence of atresia was low in the hake ovaries that we analyzed, perhaps because the females were sampled in peak spawning months. No ovaries were sampled at the end of the spawning period (June and July), when postspawning females with potentially highly atretic ovaries would be expected to be present. Sarano (1986) reported that the prevalence of atresia was low in ovaries with signs of recent spawning, whereas extensive atresia of yolked oocytes was prevalent in postspawning females taken at the end of April and May.

Batch Fecundity

Batch fecundity (F_b) relationships were established for 66 hydrated ovaries collected. The relation of batch fecundity to fish length (L) was best described by

$$F_b = -0.196L^{3.404} (N = 66,\ R^2 = 61.8\%)$$

A one-way ANOVA on batch fecundity did not show any significant difference between either years or months ($p = 0.344$, $p = 0.689$). We used fish weight in this analysis as a covariant to take into account the increasing relation between batch fecundity and fish weight.

We analyzed the relation between batch fecundity and fish gutted weight (W_g), and the resulting linear regressions presented better fits to the data. Batch fecundity increased with female gutted weight. The intercept of the regression line was not significantly different from 0 ($\alpha < 0.05$) and consequently the regressions were forced through the origin (fig. 7). The slope of this relationship provided a direct estimate of relative batch fecundity (number of eggs spawned per gram of female gutted weight).

The overall batch fecundity for 1994 was 165 eggs g^{-1} (gutted weight) (SE = 9.89), ranging from 67 to 379 eggs g^{-1}. When analyzing the evolution of batch fecundity during the season, we found small variations between months. Batch fecundity varied from 155 eggs

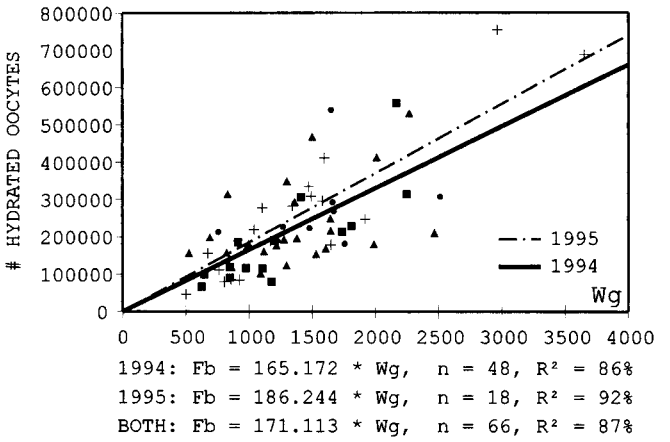

Figure 7. Batch fecundity (F_b) and gutted female weight (W_g) relationship for hydrated female hake sampled in 1994 and 1995. The linear regression fits for 1994 (heavy line) and 1995 (broken line) are shown. Symbols indicate month + year female was collected: diamond = Feb. 94; plus = Feb. 95; circle = March 94; triangle = April 94; and square = May 94.

g^{-1} in February, to 176 (SE = 26.79) in March, 170 (SE = 13.95) in April, and 145 (SE = 16.26) in May. In 1995 the mean batch fecundity reached 186 eggs g^{-1} (SE = 13.3).

Spawning Frequency

Because the duration of the spawning stages of hake ovaries are unknown, spawning frequency cannot be estimated. But spawning stages can be used to compare the relative rates of spawning during the season. The proportions of females with ovaries in different stages of development are shown in table 2.

If daily spawning were synchronous, oocyte stages would be observed at predictable times of day, but the diel timing of hake spawning is not known and the times of day that our samples were caught were too irregular for us to reach any conclusions. Thus we have neither adequate field sampling data by time of day nor laboratory data needed to assign the ages and duration to reproductive stages.

Hydrated females were much more prevalent than those in the migratory-nucleus stage, indicating that hydrated females may have been oversampled or that our catches were concentrated at times of day when hydrated females are more numerous and the migratory-nucleus stage is rare. The use of the migratory-nucleus stage to estimate spawning fraction requires that this stage would last less than one day.

Assuming a duration of less than 24 hours for the nuclear-migration stage implies a spawning fraction of 0.0525 (CV = 30%) and a batch interval of about 19 days in 1994. If hydrated females were not oversampled and the hydration stage lasted about 24 hours, then the spawning fraction would be 0.189 (CV = 17%) and the batch interval would be 5.25 days for 1994. As a reference point, we use the mean spawning frequency of these two stages—0.12 (batch interval = 8.3 days) in 1994.

Annual Fecundity

We computed different levels of realized annual fecundity, assuming that the spawning frequency was equivalent to the frequencies of migratory-nucleus stages (5.25%), hydrated stage (19%), and the mean of these stages (12%). The key assumptions here are that the stages last about 24 hours and that they are unbiased samples of the population (table 3).

To calculate annual fecundity one needs to know the duration of the spawning season. For an individual hake this is not known, but Sarano (1986) gave a duration for the stock of 3 months. Martin (1991) said that the period of reproduction of female hake at the population level lasted from January to April–May, but he stated that the limits of the reproductive period at the individual level are difficult to define.

By assuming a 3-month period from mid-January to mid-May and using the values of batch fecundity and of spawning fraction as stated above, we estimate that the realized annual fecundity of hake would be within the

TABLE 2
Percentage of Mature Females at Each Spawning Stage

			Spawning stages					
					POF classes[c]			
Month	Year	*N* mature females	NM[a]	H[b]	1	2	3+4	Other[d]
Feb.	94	21	4.8	4.8	19.1	19.1	52.2	0.0
Mar.	94	58	5.2	19.0	22.4	10.3	37.9	5.2
Apr.	94	141	7.1	26.8	9.2	8.5	38.0	10.4
May	94	52	3.9	25.0	9.6	7.7	38.4	15.4
Feb.	95	79	9.1	20.8	15.6	12.5	35.1	6.0

Collection of samples was by longline (Feb. 1994) or by pelagic trawl (March, April, May 1994 and Feb. 1995).
[a]Nuclear-migration-stage oocytes present.
[b]Hydrated oocytes present.
[c]See Results, Stages of Oocyte Development.
[d]Active ovaries, no signs of spawning, badly fixed ovaries, bad histological cuts, etc.

TABLE 3
Hypothetical Level of Realized Fecundity for an Individual Female Hake

			Nuclear-migration stage			Hydrated stage			Average spawning fraction		
Months	Duration (days)	F_h eggs/g	S^a %	Number of spawnings	Number of eggs spawned/g	S^a %	Number of spawnings	Number of eggs spawned/g	S^a %	Number of spawnings	Number of eggs spawned/g
February	15	165	5	0.79	130	19	2.85	470	12	1.8	297
March	30	165	5	1.58	260	19	5.7	940	12	3.6	594
April	30	165	5	1.58	260	19	5.7	940	12	3.6	594
May	15	165	5	0.79	130	19	2.85	470	12	1.8	297
Total	90			4.74	780		17.1	2820		10.8	1782

Based on the findings of this work for 1994, assuming that the migratory-nuclear and hydrated stage lasts less than 1 day and that both stages are independent estimates of frequency.
[a]Spawning fraction values (S) are given according to the prevalence of the nuclear-migratory stage, hydrated stage, and average or mean spawning fraction value (0.12).

range from 780 eggs g^{-1} (female gutted weight) to 2,820 eggs g^{-1} (1,782 eggs g^{-1} for a spawning fraction of 0.12) for 1994. Thus a 60 cm hake with a corresponding gutted weight of about 1,332 g would spawn a total number of eggs ranging from 1,038,960 to 3,756,240 (2,373,624 eggs for a spawning frequency of 0.12). The total (remnant) fecundity we measured for a 60 cm fish averaged 1,265,774 eggs, which lies in the lower part of the annual fecundity estimate range.

DISCUSSION

The four lines of evidence pursued in this study do not support the conclusion that European hake have determinate fecundity or fixed annual fecundity. None of the evidence supports this hypothesis for hake, but negative evidence is not nearly as strong as positive. For example, failure of fecundity to decline with season could be due to not enough samples analyzed. Lack of a break in frequency distribution of oocytes is not strong evidence for indeterminate fecundity, whereas the presence of a substantial break is strong evidence for determinate fecundity. In addition, the negative slope in diameter could be considered either as evidence for determinate fecundity by the loss of advanced oocytes due to spawning without complete replacement, or as evidence for indeterminate fecundity by recruitment of new yolked oocytes into the yolk class. Even the lower end of the estimate of annual fecundity based on S, spawning frequency, and F_h, batch fecundity, are within the range for total fecundity.

Since the assumption of determinate fecundity, when it does not exist, can lead to a very large bias, we believe it is preferable to assume fecundity in European hake is indeterminate until there is strong evidence to the contrary. There is no risk in assuming indeterminate fecundity, because estimates of spawning biomass or annual fecundity will not be biased if the standard approaches for indeterminate fish are used (batch fecundity and spawning fraction).

Sampling in February 1994 was done from a longline boat. This can explain the unusually low prevalence of hydrated females in the samples. Hydrated females are unusual in longline catches, possibly because females may not feed in that condition. On the contrary, the unusually high incidence of postovulatory follicle stage 1 females sampled by longline might be a consequence of their rapid switch from spawning to feeding because of their lack of feeding during hydration. On the other hand, the longline may be an unbiased sampler indicating a population spawning ratio of 4.8 per day.

Estimates of total fecundity of European hake are available in the literature (table 4). Sarano (1986) and Pérez and Pereiro (1985) provided overall relations between total number of vitellogenic oocytes and fish length. Sarano's results differ substantially from ours. Sarano's results are half of the levels given by Pérez and Pereiro (1985), although the latter use a higher threshold for vitellogenic oocytes (250 vs. 150 μm). But when a similar oocyte size threshold is used, Pérez and Pereiro's results are comparable to our results. According to Pérez and Pereiro, a 60 cm fish would have around 800,000–900,000 vitellogenic oocytes, which would correspond to the total fecundity.

In our case, with a threshold of 150 μm for vitellogenic oocytes, we found that a 60 cm fish has a total (remnant) fecundity of around 1,266,000. This figure is more than an order of magnitude higher than the value given by Sarano (1986) but of the same magnitude as the figure provided by Pérez and Pereiro (1.39 times higher). Clearly, determining the size threshold of vitellogenic oocytes is a critical stage in the estimation of total fecundity. Since the numbers of oocytes present in these size classes are very high, the final counts will greatly depend on the location of this threshold.

At the same time, the level of batch fecundity for the same fish would be 216,471 hydrated oocytes, on average. This means that a mature ovary contains about six potential spawning batches of eggs greater than 150 μm.

TABLE 4
Total Fecundity (F_T)-Length (L) Relationships, where $F_T = AL^B$

Source	Size range[a]	B	A	Length (cm)	F_t number oocytes	F_t/g	F_t/cm
Sarano (1986)[b]	>160 μm	3.03	0.25	60	61,057	45	1,018
Pérez and Pereiro (1985)	>250 μm	3.27	0.144	60	908,803	682	15,147
This work	>150 μm	2.144	2.29	60	1,265,774	950	21,096
This work	>250 μm	2.23	1.93	60	785,737	590	13,096

Note: All equations based on regression of $\log_{10}$ = transformed data.
[a]The minimum diameter threshold for the standing stock of vitellogenic oocytes counted in the ovaries.
[b]The coefficients are taken from a potential relationship ($y = a*x^b$) given by Sarano (1986).

The number of batches spawned per year probably depends on the energy available for spawning, which could produce more than the six-batch standing stock, or less.

The relative batch fecundity (165 eggs g^{-1}) is very close to the estimates of 173 eggs g^{-1} gonad-free weight given for the Peruvian hake, *Merluccius gayi gayi* (Alheit 1986).

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LITERATURE CITED

Alheit, J. 1986. A new method for determining batch fecundity of hake (genus: *Merluccidae*). ICES. CM. 1986/G:62.

Andreu, B. 1955. Observaciones sobre el ovario de merluza, *Merluccius merluccius*, y caracteristicas de la puesta. Inv. Pesq. Tomo 4:49–56.

Fulton, T. W. 1898. On the growth and maturation of the ovarian eggs of the teleostean fishes. Annu. Rep. Fish. Board Scotl. 16:88–124.

Greer Walker, M., P. R. Witthames, and J. I. Bautista de los Santos. 1994. Is the fecundity of the Atlantic mackerel (*Scomber scombrus*) determinate? Sarsia 79:13–26. Bergen.

Hunter, J. R. 1985. Preservation of northern anchovy in formaldehyde solution. *In* An egg production method for estimating spawning biomass of pelagic fish: application to the northern anchovy, *Engraulis mordax*, R. Lasker, ed. NOAA Tech. Rep. NMFS 36:63–65.

Hunter, J. R., and S. R. Goldberg. 1980. Spawning incidence and batch fecundity in northern anchovy, *Engraulis mordax*. Fish. Bull. 77 no.3.

Hunter, J. R., and B. J. Macewicz. 1985. Measurement of spawning frequency in multiple spawning fishes. *In* An egg production method for estimating spawning biomass of pelagic fish: application to the northern anchovy, *Engraulis mordax*, R. Lasker, ed. NOAA Tech. Rep. NMFS 36:79–93.

Hunter, J. R., N. C. H. Lo, and R. J. H. Leong. 1985. Batch fecundity in multiple spawning fishes. *In* An egg production method for estimating spawning biomass of pelagic fish: application to the northern anchovy, *Engraulis mordax*, R. Lasker, ed. NOAA Tech. Rep. NMFS 36:67–78.

Hunter, J. R., B. J. Macewicz, and C. A. Kimbrell. 1989. Fecundity and other aspects of the reproduction of sablefish, *Anaplopoma fimbria*, in central California waters. Calif. Coop. Oceanic Fish. Invest. Rep. 30:61–72.

Hunter, J. R., B. J. Macewicz, N. C.-H. Lo, and C. A. Kimbrell. 1992. Fecundity, spawning, and maturity of female Dover sole, *Microstomus pacificus*, with an evaluation of assumptions and precision. Fish. Bull., U.S. 90:101–128.

Martin, I. 1991. A preliminary analysis of some biological aspects of hake (*M. merluccius* L. 1758) in the Bay of Biscay. ICES C.M. 1991/G:54. Demersal Fish Committee.

Pérez, N., and F. J. Pereiro. 1985. Aspectos de la reproducción de la merluza, *M. merluccius*, de la plataforma gallega y cantábrica. Bol. Ins. Esp. Ocean. 2(3)1985:39–47.

Sarano, F. 1986. Cycle ovarien du merlu, *M. merluccius*, poisson à ponte fractionée. Rev. Trav. Inst. Péches. Marit. 48(1 et 2):65–76.

Selman, K., R. Wallace. 1989. Cellular aspects of oocyte growth in teleosts. Zool. Sci. 6:211–231.

Wallace, R., and K. Selman. 1981. Cellular and dynamic aspects of oocyte growth in teleosts. Am. Zool. 21:325–43.

West, G. 1990. Methods of assessing ovarian development in fishes: a review. Aust. J. Mar. Freshwater Res. 41:199–222.