Batch Fecundity in Multiple Spawning Fishes

J. ROE HUNTER, NANCY C. H. LO, and RODERICK J. H. LEONG
Southwest Fisheries Center, National Marine Fisheries Service, NOAA, P.O. Box 271, La Jolla, CA 92038

ABSTRACT

Methods for estimating batch fecundity are described, including identification and sampling the eggs that constitute one spawning batch within the ovary. Using northern anchovy, *Engaulis mordax*, as an example, methods are developed for evaluating the accuracy and precision of batch fecundity estimates. Included in this analysis are the number and location of ovarian tissue samples, optimal numbers of females, and selection of the appropriate function to express the relation between batch fecundity and female weight. We found for anchovy that the optimum number of ovarian tissue samples was 2-3 per ovary and that to keep the coefficient of variation for the mean fecundity of population under 10% required a sample of 50 or more females. Analysis of covariance indicated that the batch fecundity of northern anchovy varied significantly among years (1951-84) indicating that the relation between female weight and fecundity must be newly established each year.

INTRODUCTION

A key issue in the estimation of fecundity of oviparous fishes is whether or not the annual fecundity can be estimated from the standing stock of advanced oocytes in the ovary prior to the onset of the reproductive season. In some boreal species, frequently called total or isochronal spawners, all the eggs to be released in a season develop synchronously prior to spawning (hence the term isochronal) and spawning typically takes place over a short period (Holden and Raitt 1974). In such species, the standing stock of oocytes within a certain range of maturity classes is considered to represent the annual fecundity of the spawner. The groups of oocytes to be spawned in the season are usually identifiable because a distinct hiatus in oocyte maturity classes exists between the small, immature, unyolked oocytes that occur the year around and the synchronously maturing annual batch (Hickling and Rutenberg 1936; Yamamoto 1956). Although some of these fishes may spawn repeatedly during the season, for example, whiting and haddock, the standing stock of yolked eggs is considered representative of the annual fecundity (Hislop 1975; Hislop et al. 1978; Hislop, pers. commun.[1]). An exception to this occurs when unfavorable conditions result in resorption of some of the advanced eggs in the ovary at the end of the season. The extent of this potential bias (overestimation of annual fecundity) is unknown.

In many temperate and tropical fishes (frequently called multiple, partial, serial, or heterochronal spawners), annual fecundity is seasonally indeterminate and batch fecundity is the only useful measurement. In such fishes the standing stock of yolked eggs, regardless of maturity state, give no indication of annual fecundity because these fishes continuously mature new spawning batches throughout a typically protracted spawning season. In the active ovaries of fishes with indeterminate annual fecundity, the oocytes usually occur in nearly all maturity stages; they range in size continuously from small unyolked oocytes <0.1 mm diam. to yolked oocytes 0.4-0.7 mm diam., and no large hiatus exists between maturity classes of oocytes except for one between hydrated oocytes and advanced yolked oocytes which is of a temporary nature. Such fishes usually spawn many times during a season. The northern anchovy spawns at 7-10 d intervals for 2 or 3 mo and averages 20 spawnings per yr (Hunter and Leong 1981), and the scianid, *Seriphus politus*, has a similar reproductive output (DeMartini and Fountain 1981). Thus, for these fishes, identification of a predetermined annual spawning batch is a hopeless exercise, and the only useful fecundity measurement is the number of eggs produced in a single spawning batch (batch fecundity); annual fecundity is a function of both the batch fecundity and the number of spawnings per year. Spawnings are so numerous in these fishes that small unyolked oocytes <0.1 mm diam. would have to mature in a season to account for the number of spawnings (Hunter and Leong 1981).

The standing stock of oocytes is occasionally used to estimate annual fecundity in such common fishes as *Scomber*, *Trachurus*, and *Merluccius*, which by the standard criteria have indeterminate fecundity. That annual fecundity is predetermined in such fishes is an assumption with little or no supporting evidence. The criteria and approaches for distinguishing between determinate and indeterminate fecundity are discussed in greater detail (Hunter and Macewicz 1985).

The objective of this paper is to describe the methodologies for estimating batch fecundity in fishes with indeterminate seasonal fecundity. We do not consider the well documented methodology

[1]John R. G. Hislop, DAFS Marine Lab., P.O. Box 101, Victoria Road, Aberdeen AB9 8DB, Scotland, pers. commun. Oct. 25, 1983.

for fecundity estimation of fishes with seasonally determinate fecundity (see, for example, Holden and Raitt 1974). In anchovy and other fishes with indeterminate annual fecundity, the oocytes in active ovaries are typically distributed in 1-2 modes (Fig. 1), each mode representing a single spawning batch. Maturation of oocytes and vitellogenesis are a continuous cycle: when one spawning batch is spawned, another spawning batch is ready for the last stages of maturation and spawning (Fig. 2). Vitellogenesis proceeds rapidly after a spawning, with the ovary doubling in dry weight during the interval between spawnings (Hunter and Leong 1981). The final stage of maturation, hydration, is characterized by a rapid secretion of fluid of low specific gravity into the advanced eggs by the granulosa cells of the follicle (Fulton 1898). This fluid causes more or less complete fusion or solution of the yolk granules producing the translucent appearance of hydrated eggs. The volume of the egg or wet weight increases three- or four-fold (Fulton 1898), but the increase in dry weight is negligible (LeClus 1979a). In northern anchovy, hydration begins about 12 h before spawning when the eggs are between 0.6 and 0.8 mm (major egg axis) and causes a four-fold increase in wet weight of the ovary as the egg increases to 1.3 mm (major axis) (Hunter and Macewicz 1980; Hunter and Leong 1981) (Fig. 2). Ovulation and spawning soon follow completion of hydration in most clupeoids (anchovy, pilchard, sardines, and others) but in herring, a total spawner, ovulated eggs may be retained in the ovary for an extended period.

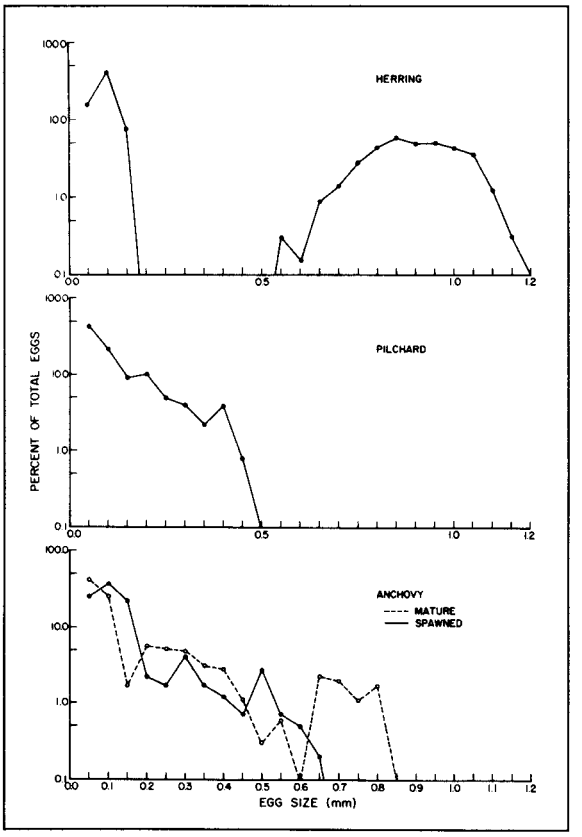

Figure 1.—Frequency distribution of oocyte diameter in the ovaries of herring, pilchard (Hickling and Rutenberg 1936), and northern anchovy (Hunter and Leong 1981). Herring spawns a single batch each year; other species are multiple-batch spawners. In the anchovy, the solid line shows a recently spawned female, the broken line a female about to spawn (just before hydration of the oocytes). (From Blaxter and Hunter 1982.)

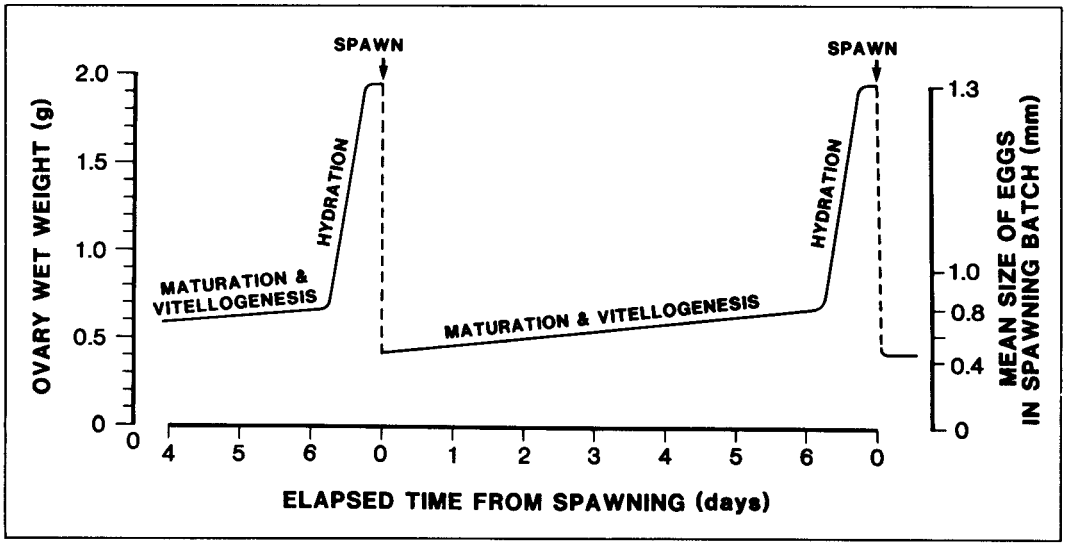

Figure 2.—Maturation cycle of a 12.5 g northern anchovy female during peak spawning months where the average interval between spawnings is 7 d. The change in mean wet weight of the ovary is indicated on the left axis and the mean diameter of the oocytes in the most advanced spawning batch on the right axis. Data from Hunter and Goldberg (1980) and Hunter and Macewicz (1980).

LABORATORY PROCEDURES

Identifying and Counting Oocytes in a Batch

A number of criteria have been used to identify the oocytes to be included in a spawning batch. These include 1) counts of all yolked oocytes; 2) estimation of the number of oocytes in the most advanced spawning batch by measuring the size distribution of oocytes in the ovary and identifying the most advanced (largest) modal group of oocytes; and 3) estimation of the number of oocytes in a spawning batch by counting the number of hydrated oocytes.

The first method can be rejected because the standing stock of yolked oocytes gives indication neither of total fecundity nor of batch size in fishes with indeterminate fecundity and is appropriate only for fishes with determinate seasonal fecundity. The second method (oocyte size frequency) usually gives results similar to those based on counts of hydrated oocytes if females with highly advanced oocytes are used (Hunter and Goldberg 1980; Laroche and Richardson 1980). We believe the third method, counting the hydrated oocytes, is preferable because it requires less time and avoids the problem of partitioning oocytes between the most advanced mode and the adjacent group of smaller oocytes. Before describing methods 2 and 3, it is important to consider methods of sampling the ovary.

Methods of Sampling an Ovary

It is impractical to count and measure all advanced oocytes, or to count all hydrated oocytes in an ovary, owing to the great fecundity of most marine fishes. Thus, regardless of the method used for identifying the spawning batch, ovarian subsamples are required, and these are related to either the ovarian weight (gravimetric method) or the total volume of an aqueous suspension of all oocytes in the ovary (volumetric method). The gravimetric method is based on counting oocytes in weighed samples of ovarian tissue and relating the tissue samples to the total ovary weight. In the volumetric method, the ovary is preserved in Gilson's fluid which frees the oocytes from the ovarian tissue by breaking down the connective tissue (see Baganel 1967 for the recipe for Gilson's fluid). The released oocytes are cleaned, put in a volumetric cylinder filled to a known volume with water, shaken to provide thorough mixing, subsamples of known volume are withdrawn using a Stempel pipette, and the oocytes staged and counted (Holden and Raitt 1974). Automatic oocyte counters may also be employed.

The volumetric method may be used for batch fecundity estimation if the eggs constituting the batch are identified using the egg size-frequency method; however, the volumetric technique is inappropriate if the hydrated oocyte method is used because Gilson's fluid destroys hydrated eggs. Substantial shrinkage of oocytes occurs when ovaries are preserved in Gilson's fluid; an average shrinkage of 24% (compared to formalin-preserved ovaries) occurs when skipjack and yellowfin tuna ovaries are preserved in Gilson's fluid, but no differential shrinkage occurs among oocyte size classes (Joseph 1963). Thus, to make oocyte size classes comparable to live, or formalin-preserved, or histological sections, the extent of shrinkage must be measured and the data corrected. Treatment with Gilson's fluid also destroys the ovary, making histological analysis impossible.

We use the gravimetric method of MacGregor (1957), which is somewhat similar to the gravimetric method "B" of LeClus (1977) who evaluated two gravimetric and one volumetric techniques. Although LeClus obtained very low coefficients of variation for all techniques in her 1977 methods paper, the relative fecundity estimated for the South African anchovy in a subsequent paper (LeClus 1979b) was as variable as those employing less complicated procedures (MacGregor 1957, 1968; Hunter and Goldberg 1980). Her use of vacuum dry weight of the ovary, instead of formalin wet weight, seems an unnecessary refinement. Natural variability in batch fecundity appears to be much greater than the variation caused by differences in the technique of sampling the ovary.

The step-by-step procedure for the hydrated oocyte method is outlined below. It will be elementary to many biologists, but it is intended as a guide for inexperienced staff. Except for some details outlined in a subsequent section, the same procedure can be used to estimate batch fecundity using the oocyte size-frequency method.

The Hydrated Oocyte Method

1. The basic method is as follows: Numbers of hydrated oocytes in weighed tissue samples of formalin-preserved ovary are counted and the counts are then projected to estimate numbers of hydrated oocytes in the entire ovary which is assumed to be equivalent to batch fecundity.

2. Prior to batch fecundity estimation, females are accurately weighed, and the ovary removed, weighed, and stored in an individual vial of buffered 10% formalin. Ovaries which appear hydrated are noted along with an estimate of the numbers of free eggs in a collection jar (Hunter 1985). Only hydrated ovaries which have not lost oocytes are used for fecundity estimation; ovaries that have lost oocytes in the jar are rejected after histological examination because they contain postovulatory follicles.

3. Needed supplies and equipment include a balance sensitive to 0.1 mg (which should be checked with standard weights), a dissection microscope with a 10× objective, hand counter, forceps, scalpel, bottle of glycerin (33% glycerol solution by volume) with eyedropper, glass slides (25×75 mm), cover slips (22×50 mm), paper towels for blotting, and weighing paper.

4. Remove ovary from the formalin fixative and blot dry with paper towel. Break the ovarian membrane and remove three tissue samples of the ovary. The ovary is soft and the sample can be removed easily with the tip of the forceps or scalpel. Remove samples from positions about one-third of the distance from each end of the ovary to insure that no two samples come from the same portion of the ovary (only one ovary, left or right, need be used). Try to obtain a tissue weight of 30-50 mg, as this will contain an adequate number of hydrated oocytes (100-200). Place sample on a preweighed piece of weighing paper and record weight to nearest 0.1 mg. Pieces of ovary can be added or removed to vary sample weight.

5. Place the sample on a slide and cover with 3-4 drops of glycerin. After 10-15 min, loosen the oocytes by gently tapping the piece of ovary with the blunt tip of the forceps. After the oocytes are loosened, add 3 or 4 more drops of glycerin, spread the sample over the slide, and cover with a cover slip so that it floats on the fluid. (We found that this concentration of glycerin had no effect on the diameters of oocytes taken from formalin-preserved ovaries, even after 24 h.)

6. Place the slide under the microscope, and with a hand counter tally the number of hydrated oocytes in the sample. Hydrated oocytes can be distinguished easily from other oocytes by their large size

(usually ≥0.8 mm in the major axis in northern anchovy), wrinkled appearance when formalin preserved (yolked but nonhydrated oocytes usually retain their smooth surface contour), and by their translucence (nonhydrated eggs are relatively opaque, Fig. 3). Some damage to the hydrated oocytes may occur during slide preparation. In some cases, the chorion may be ruptured and the yolk extruded. Do not count empty chorions, and count only those fragments judged to be major portions of the oocytes.

7. Batch fecundity (Y) for each female is calculated from the product of the number of hydrated oocytes (eggs) per unit weight in the tissue sample and the ovary weight (left and right sides combined) (Z).

8. The egg production method requires that batch fecundity be expressed as a function of female weight and not length, i.e., $EY = f(w)$ where female weight (w) is the formalin wet weight of the female without the ovary (formalin wet weight can be converted to live weight using coefficients given in Hunter 1985). We use ovary-free wet weight of females since females with hydrated ovaries temporarily have a higher weight than the average female because of the increased weight of the hydrated ovary. The ratio of female body weight without ovary to female weight with ovary (excluding females with hydrated or immature ovaries) can be used to convert ovary-free wet weight to total body weight. This ratio in northern anchovy was 0.95 for female anchovy taken 1978-79 (Hunter and Macewicz 1980).

Oocyte Size-Frequency Method

If the number of females with hydrated oocytes is insufficient for a batch fecundity estimate, the more time-consuming oocyte size-frequency distribution method can be employed (MacGregor 1957). This method takes 1-3 h per fish (3 tissue samples per fish) as compared with 1-1.5 h for the hydrated oocyte method. In this method, a size-frequency distribution of oocytes is constructed and the most advanced modal group of oocyte size classes (the mode composed of the largest oocytes) is determined by inspection. The total number of oocytes within the oocyte size classes that constitute the advanced modal group is considered to be the spawning batch. This method usually gives results similar to those based on counts of hydrated oocytes if females with highly advanced oocytes are used (Hunter and Goldberg 1980; Laroche and Richardson 1980). The mean size of the group of oocytes that constitutes the most advanced spawning batch should be ≥0.5 mm, as estimates of batch fecundity are somewhat inflated if a less mature ovary is used (Hunter and Goldberg 1980).

In northern anchovy, a tissue sample weight of ≤10 mg will insure that about 100 oocytes are included in the most advanced modal group of oocytes. All oocytes ≥0.3 mm in a tissue sample are counted using a set of hand counters and measured to the nearest 0.05 mm. We use an optical comparator at 50× magnification and measure the oocytes with a rule on the viewing screen of the comparator. A starting oocyte size ≤0.3 mm is recommended to insure that a

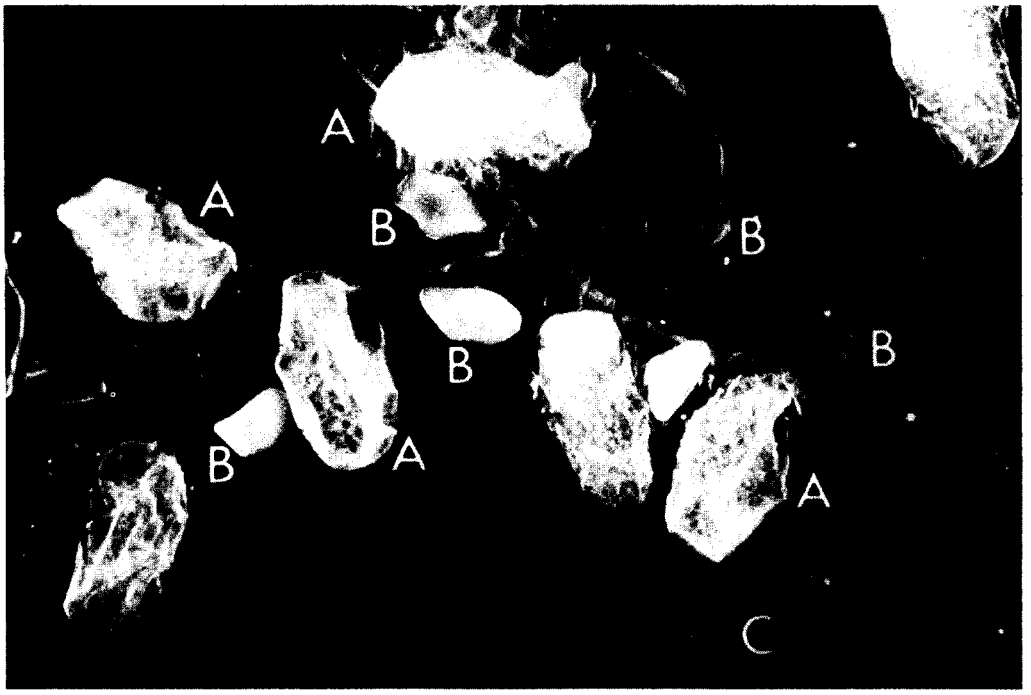

Figure 3.—Hydrated oocytes in a tissue sample taken from a northern anchovy ovary preserved in formaldehyde solution. A. Hydrated oocytes; B. Unyolked and yolked oocytes (before hydration); C. Empty chorion of a hydrated oocyte (the chorion, or a major fragment of it, would be included in the count of hydrated oocytes for the batch fecundity estimate). These hydrated oocytes were 1.2 mm long in the major axis.

sufficient number of 0.05-mm oocyte size classes exist below the most advanced modal group. Inclusion of these small oocyte size classes insures an accurate separation of the tail of the advanced mode of oocytes from the smaller oocytes adjacent to it. To separate the tail of the advanced modal group from the adjacent group of smaller oocytes, we use probability paper analysis (Harding 1949; Cassie 1954). In other respects, the oocyte size-frequency method is the same as the hydrated oocyte method, and the previous section can be used as a guide.

ACCURACY AND PRECISION

In this section we develop methods to evaluate the accuracy and precision of estimating batch fecundity using the hydrated egg methodology, and apply these procedures to northern anchovy fecundity data. Accurate estimation of batch fecundity depends upon selection of an unbiased location for the samples of ovarian tissue and the selection of an appropriate regression model to express the relation between female weight and batch fecundity. The precision of the estimate depends upon the number of ovarian tissue samples taken per female and the total number of females. The weight of the individual tissue sample also affects precision, but we have not considered this element. We have instead kept the tissue samples within a weight range that yields about 100-200 hydrated oocytes per sample.

In the first subsection we use analysis of variance to detect the possible effects of location of tissue samples within the ovary. In this analysis we use a sample of 12 northern anchovy ovaries in which 6 tissue samples were taken per ovary at specified locations. In the next subsection we determine how the number of tissue samples affect the precision of the fecundity estimate; consider various fecundity-weight models; and determine the optimum numbers of fish and tissue samples for a given cost. For this analysis we add an additional 12 fish to the sample used in the first section, all of which had 6 tissue samples per fish. In the third subsection we use seven data sets on anchovy batch fecundity taken 1951-60 and 1978-84 (n ranges, 19-127) to validate selection of the fecundity-female weight model and to assess the precision of regression estimates of batch fecundity. In the final subsections we consider how the number of fish in the sample affects precision of the fecundity estimate and how the batch fecundity varies among years.

Location of Tissue Samples Within the Ovary

To determine if location of the tissue samples affects estimates of batch fecundity in anchovy, we took 6 ovarian tissue samples from each of the 12 ovaries; three samples were taken from the left ovary and three from the right. In each set of three, one sample was taken in the center and the other two were about one-third of the distance from each end of the ovary. The number of eggs per unit weight of the ovary (x) was calculated for each tissue sample. We tested effects of right or left ovarian side, and position of tissue samples within a side, using the two-way analysis of variance. The natural logarithm of x was used in the analysis because there is a positive correlation between the same sample mean and its standard deviation. The assumption of homogeneity within sample variance is violated when the means differ.

No difference existed either in the location of the tissue sample within right or left sides of the anchovy ovary or between right and left sides (Table 1). Thus in northern anchovy, tissue samples can be taken from any location or from either the right or left sides. It should be noted, however, that all of our samples were of females taken at a time of day (1900-0100 h) when hydration was nearly complete. If females are taken earlier in the day, position effects may be likely because hydration does not proceed at a uniform rate throughout the ovary; rather it begins at the periphery and spreads to the central section of the ovary, producing larger hydrated oocytes (more mature) at the periphery than in the center. Thus the number of hydrated oocytes per gram of ovary may be higher in the central section and lower on the periphery at early stages of hydration. Variation in extent of hydration does not appear to be a major source of error, but it should be evaluated prior to making a fecundity estimate in a new species or when samples are taken at a new time of day.

Table 1.—Effect of location of ovarian tissue samples from northern anchovy on the number of hydrated eggs per unit sample weight (g). Effects evaluated by taking tissue samples (n = sample size) from three positions (two ends, I and III, and middle, II) from both the right and left ovary. Analysis of variance indicates insignificance of either side or position within a side (SS = sum of squares; MS = mean square).

Positions of sample in ovary	Mean no. of eggs/g of ovary tissue								
	Right ovary			Left ovary			Both ovaries		
	$\bar{x}$	s	n	$\bar{x}$	s	n	$\bar{x}$	s	n
I	5,237	740	12	5,143	1,158	12	5,190	950	24
II	4,919	790	12	5,243	831	12	5,081	810	24
III	5,296	1,005	12	5,540	2,019	12	5,418	1,564	24
All	5,151	845	36	5,309	1,396	36			

Two-way analysis of variance of eggs/g of ovary tissue.

SS due to	df	SS	MS	F
Right vs. left ovary	1	0.0053	0.0053	0.14
Position within ovary	2	0.0268	0.0134	0.34
Interaction	2	0.0269	0.0134	0.34
Error	66	2.5854	0.0392	—
Total	71	2.6444	—	—

Optimum Number of Tissue Samples and Numbers of Fish for Fecundity Estimation

In this section we develop equations to estimate the optimum number of fish and tissue samples to be used for fecundity estimation using data on the northern anchovy. The precision of the sample variance (or mean square error) around the regression of batch fecundity on female weight (σ_A^2) (a measure of the goodness of fit of the fecundity-weight model) was used to determine the optimum number of tissue and fish samples. This procedure required a definition of a general fecundity model and development of functions to express the error terms.

A. The general fecundity model—The true batch fecundity (Y) where all eggs are counted, and the fish weight relationship (w) is defined as:

$$Y_w = f(w) + A \qquad (1)$$

where the error term (A) has a mean = 0, and variance = σ_A^2. Y and Y_w are interchangeable in later sections. Since all the hydrated eggs in a batch (Y) are not counted, $f(w)$ are fitted to the estimated batch fecundities ($\hat{Y}$) calculated from m ovarian tissue samples per fish.

Let's denote for the ith fish, $i=1,\ldots,n$:

w_i = gonad-free fish weight = fish weight minus gonad weight
Y_i = total number of hydrated eggs in the ovary
y_{ij} = hydrated egg count in the jth tissue sample $j=1,\ldots,m$
z_{ij} = weight of jth tissue sample
Z_i = formalin wet weight of gonad
m = number of tissue samples from an ovary
M_i = maximum number of tissue samples in an ovary
$\hat{Y}_{ij}$ = estimated total number hydrated eggs in the ovary from the jth tissue sample = $(y_{ij}/z_{ij})Z_i$
$\hat{Y}_i$ = estimated total number of hydrated eggs in the ovary $= \sum_{j=1}^{m} \hat{Y}_{ij}/m$
$\bar{Y}$ = sample mean number of hydrated eggs = $\sum_i \hat{Y}_i/n$
$\hat{Y}_i$ = estimate of batch fecundity from the regression model.

Suppose $\hat{Y}_{ij}$ is an unbiased estimate of Y_i, then

$$\hat{Y}_{ij} = Y_i + \hat{Y}_{ij} - Y_i = f(w_i) + A_i + e_{ij} \tag{2}$$

where $e_{ij} = \hat{Y}_{ij} - Y_i$ is the within-ovary error term and is assumed normally distributed with mean = 0 and variance = σ_e^2. Using Equation (2) and the fact that

$$\begin{aligned} \hat{Y}_i &= \sum_{j=1}^{m} \hat{Y}_{ij}/m, \text{ we have} \\ \hat{Y}_i &= f(w_i) + A_i + e_{i\cdot} \\ &= f(w_i) + \xi_i \end{aligned} \tag{3}$$

where $\xi_i = A_i + e_{i\cdot}$ and $\sigma_{\xi_i}^2 = \sigma_A^2 + \frac{M_i - m}{M_i}\frac{\sigma_e^2}{m}$.

Thus the variance around the regression line based upon data set $(\hat{Y}_i, w_i)$ is composed of two variance components: One is σ_A^2 and the other is σ_e^2, the within-ovary variance. The unbiased estimates of $\sigma_{\xi_i}^2$ and σ_e^2 are

$$s_{\xi_i}^2 = \frac{\sum_{i=1}^{n} [\hat{Y}_i - f(w_i)]^2}{n-q} \tag{4}$$

and

$$s_e^2 = \frac{\sum_i \sum_j (\hat{Y}_{ij} - \hat{Y}_i)^2}{n(m-1)}. \tag{5}$$

The parameter q is the number of regression coefficients in the model and n is the number of fish sampled. Most if not all fecundity regressions use only two coefficients (Bagenal 1967) and consequently we use $q = 2$ in this article, and subsequent computations should be redone if $q>2$. For simplicity, we assume that $M_i = M_j = M$ for $j \neq i$, and $\sigma_{\xi_i}^2 = \sigma_{\xi_j}^2 = \sigma_\xi^2$.

Thus

$$\sigma_A^2 = \sigma_\xi^2 - \frac{M-m}{M}\frac{\sigma_e^2}{m}$$

and

$$s_A^2 = s_\xi^2 - \frac{M-m}{M}\frac{s_e^2}{m}. \tag{6}$$

The variance of the sample variance (s_A^2) is used in the next section for computing the optimum number of ovarian tissue samples for fecundity estimation.

B. Optimum number of tissue samples—Since the goodness of fit of any model is measured by s_A^2, we chose to minimize the variance of s_A^2 with respect to the number of tissue samples (m) for a fixed total cost function (Scheffé 1959).

$$c = c_1 n + c_2 mn \tag{7}$$

where c is the total funds available,
c_1 is the cost of processing a fish, and
c_2 is the cost of processing a tissue sample.

Under the assumption of normality of the error terms, one obtains the variance of s_A^2 as:

$$var(s_A^2) = 2\sigma_A^4 \left[\frac{\left\{1 + \left(\frac{1}{m} - \frac{1}{M}\right)\theta\right\}^2}{(n-q)} + \left(\frac{1}{m} - \frac{1}{M}\right)^2 \frac{\theta^2}{n(m-1)} \right] \tag{8}$$

where $\theta = \frac{\sigma_e^2}{\sigma_A^2}$ and $\hat{\theta} = \frac{s_e^2}{s_A^2}$.

The optimum sample sizes for selected parameter values are listed in Table 2. The optimum tissue sample size (m) depends on the values of each of the five parameters, i.e., q, $\theta = \sigma_e^2/\sigma_A^2$, c, c_1, and c_2.

Table 2.—Optimum number of ovarian tissue samples required for batch fecundity estimation of northern anchovy with two regression coefficients (q=2) and maximum tissue samples >30 within an ovary (M>30) for various degrees of data variability ($\theta=\sigma_e^2/\sigma_A^2$) and cost constraints ($c/c_2$ and c_1/c_2).

Ratio of within-ovary variance to variance about the line ($\theta=\sigma_e^2/\sigma_A^2$)	Total funds available for estimated cost per tissue sample (c/c_2)	Relative index of processing costs (costs of fish/cost of sample (c_1/c_2)*		
		0*	4	8
θ = 0.5	50	2	2	2
	200	2	3	3
	10,000	2	3	3
θ = 1	50	2	3	3
	200	2	4	4
	10,000	2	4	5
θ = 2	50	3	4	4
	200	3	5	6
	10,000	3	6	7
θ = 3	50	3	5	4
	200	4	7	8
	10,000	4	7	9
θ = 4	50	4	6	4
	200	5	8	10
	10,000	5	9	11
θ = 5	50	5	6	4
	200	6	9	11
	10,000	7	11	13

*0 = case where cost of capturing fish is negligible relative to cost of fecundity estimate.

The finite population correction factor $(1-m/M)$ can be ignored for $M \geq 30$. This means that it is unlikely that the finite population correction will ever be used because it is very unlikely that a tissue sample as large as 1/30 of the ovary weight would ever be used in a fecundity estimation. Hence, an infinitely large M ($M \geq 30$) can be used for all calculations.

To evaluate the adequacy of Equation (3), we compute a ratio of σ_ξ^2 and σ_A^2, i.e., $K = \sigma_\xi^2/\sigma_A^2$; $K \geq 1$. A large K implies the variation in Equation (3) is too high and a larger sample size is desirable. When K is close to 1, the precision of Equation (3) is nearly as good as that of Equation (1) which uses the true fecundity where all eggs in a batch are counted. K is computed from the following equation:

$$K = \frac{\sigma_\xi^2}{\sigma_A^2} = \left(\frac{1}{m} - \frac{1}{M}\right)\theta + 1 \qquad (9)$$

and

$$\hat{K} = \frac{s_\xi^2}{s_A^2} = \left(\frac{1}{m} - \frac{1}{M}\right)\hat{\theta} + 1 \cdot$$

Table 3 gives K values for number of tissue samples ranging from 1 to 6 for $\theta = 0.5$-0.6. K can be computed easily for other values of θ and number of possible tissue samples within an ovary. The precision of $\hat{K}$ depends on the variance of s_ξ^2 and s_A^2 and the cost function for processing the fish and making the fecundity estimate [Equation (7)]. Costs dictate the number of fish used and the number of tissue samples taken per fish.

To evaluate K for different cost functions (hence sample sizes), it is necessary to select a proper function to express the relationship between fecundity and female weight, $f(w)$. We used four models and the data set of 24 fish (Table 3). The four models were:

$$\hat{Y} = \begin{cases} f_1(w) = aw^b + e \\ f_2(w) = ae^{bw} + e \\ f_3(w) = a + b\ln(w) + e \\ f_4(w) = a + bw + e \cdot \end{cases}$$

The linear model had the greatest precision because it had the lowest s_ξ^2 and s_A^2 (Table 3). The linear model also seems a preferable method of expressing the fecundity-weight relationship in northern anchovy when much larger data sets are considered (see next section).

The parameter $\hat{\theta} = s_e^2/s_A^2$ (a measure of the relative variability within tissue samples) ranged from 0.5 to 0.6, depending on the regression model selected for the 24-fish sample. In our case, the cost (c_1) of catching and curating a single fish is negligible relative to the cost (c_2) of counting the hydrated eggs in a tissue sample because many fish must be taken for the spawning frequency estimation. Thus for northern anchovy, $c_1/c_2 \sim 0$. Using Table 2, the optimum number of tissue samples (m) is 2. For m=2, and $\hat{\theta}$=0.5, $\hat{K}$=1.3 [Equation (9)]. This means that the variance around Equation (3) is about 1.3 times that of the model based on counts of all hydrated eggs in the ovary [Equation (1)]. To reduce the $\hat{K}$ value, more tissue samples are needed which increases the total cost (c). For northern anchovy, there is no reason to increase the number of tissue samples beyond three, because the reduction in $\hat{K}$ becomes negligible at larger sample sizes (Table 3).

Validation of the regression model

In the previous section we concluded that the linear model for expressing the relation of female batch fecundity to female weight was preferable, but we used a small data set (n = 24) in which 3-6 tissue samples were taken per fish. In this section we test and evaluate this conclusion by fitting a linear model ($\bar{Y}_w = \bar{Y} + b(w-\bar{w})$) and two nonlinear models ($\bar{Y} = ae^{bw}$ and $\bar{Y} = aw^b$) to all existing data sets on the fecundity of northern anchovy (1950-84) (Tables 4 and 5). In these sets the number of tissue samples per fish varied from 1 to 3. Mean square error (MSE) = $\Sigma(\hat{Y}-\bar{Y})^2/(n-2)$ was computed for all three models for each data set. Although no apparent difference existed among MSE values for the three models, and no pattern existed in the residuals (Fig. 4), the simple linear model is preferable because: 1) it explains as much variation as the cur-

Table 3.—Effect of ovarian tissue samples taken per ovary (m) on the ratio K for various models that express the relation between female weight, w (without ovary) and batch fecundity, $\hat{Y}_w$.

Regression model $f(w)$	s_ξ^2 ($\times 10^3$)	Maximum no. of tissue samples (M)	$^*s_A^2$ ($\times 10^3$)	$^\dagger s_e^2/s_A^2$	$^\ddagger K$ for 1-6 tissue samples/ovary					
					1	2	3	4	5	6
(1) aw^b	6,302	∞	5,729	0.60	1.60	1.30	1.20	1.15	1.12	1.10
a = 325.07		80	5,772	0.60	1.59	1.29	1.19	1.14	1.11	1.09
b = 1.2423		30	5,844	0.59	1.57	1.28	1.18	1.13	1.10	1.08
(2) ae^{bw}	7,112	∞	6,539	0.53	1.53	1.27	1.18	1.13	1.11	1.09
a = 3,281.28		80	6,582	0.52	1.51	1.25	1.17	1.12	1.10	1.08
b = 0.0612		30	6,654	0.50	1.50	1.24	1.16	1.11	1.09	1.07
(3) $a+b\ln(w)$	6,274	∞	5,702	0.60	1.60	1.30	1.20	1.15	1.12	1.10
a = −31,831.73		80	5,744	0.60	1.59	1.29	1.19	1.14	1.11	1.09
b = 15,371.04		30	5,816	0.59	1.57	1.28	1.18	1.13	1.10	1.08
(4) $a+bw$	6,220	∞	5,648	0.61	1.61	1.31	1.20	1.15	1.12	1.10
a = 3,190.38		80	5,690	0.60	1.59	1.29	1.19	1.14	1.11	1.09
b = 838.11		30	5,762	0.60	1.58	1.28	1.18	1.13	1.10	1.08

*From equation (6).

†Within-ovary variance (s_e^2) = 3,436×10³ from Equation (5); ratios computed using 7 digits.

‡K is a measure of the variance around regression line relative to the "free" variance around the curve (Equation (1)). When K = 1, the variance from subsampling ovarian tissue is 0 since all eggs are counted.

Table 4.—Sample statistics for batch fecundity estimation of the central subpopulation of northern anchovy for various years. (See Addendum for 1985 data.)

Statistic	1951-60*	1978	1979	1980	1981	1982	1983	1984
n[†]	19	23	44	33	127	109	83	87
Linear regression coefficient (b)	532	279	693	563	752	617	588	532
Linear regression intercept (a)	1,122	2,023	−4,410	−1,891	−1,979	−180	−1,002	−554
$s_{y \cdot x}$[‡]	2,752	3,103	2,935	1,276	2,522	2,583	1,282	1,281
Sample mean batch fecundity ($\bar{Y}$)[†]	10,270	7,546	8,506	7,745	9,083	10,031	5,828	5,862
Sample mean ovary-free fish weight ($\bar{w}$)[†]	17.18	19.76	18.64	17.10	14.70	16.54	11.63	12.06
Sample standard deviation of fish weight (s_w)	6.57	6.62	5.32	4.77	5.81	4.41	4.75	5.54
$\Sigma(w-\bar{w})^2$	777	964	1,208	728	4,253	2,100	1,850	2,639
Mean fish weight from survey ($\bar{\bar{w}}$)	—	—	—	17.50	16.20[¶] 13.40	18.60	12.90[¶] 11.20 9.63	12.02
Standard error of fish weight ($s_{\bar{\bar{w}}}$)	—	—	—	0.96	0.47[¶] 0.52	0.37	1.56[¶] 0.79 0.37	0.46
Adjusted mean batch fecundity for ovary-free fish weight = 14.95 g[§]	8,900	4,597	6,241	6,423	9,237	9,055	7,870	7,640
Standard error of batch fecundity for 14.95 g fish[§]	536	494	358	408	207	225	264	256

*From MacGregor (1968).
[†]Sample mean batch fecundity ($\bar{y}$) and mean fish weight ($\bar{w}$) are computed from data set with n fish.
[‡]Standard deviation about the line.
[§]Computed from analysis of covariance.
[¶]Computed from individual cruises (1981 values for two cruises and 1983 for three cruises).

vilinear model; 2) its regression coefficients have a simple biological meaning (b = batch fecundity/gram female weight, $\bar{Y}$ = mean batch fecundity); and 3) for the egg production estimate, the fecundities of the largest and smallest fish are not as critical as for the fish in the middle range, which is well explained by the simple linear model.

Analysis of these eight data sets also indicated that the standard deviation of the batch fecundity is a linear function of the female weight (R^2 ranges from 0.2 to 0.6). The minimum variance and unbiased estimates of the regression coefficients can be obtained through a weighted least squares regression with the inverse of the variance as the weight (Draper and Smith 1981). In most cases the standard errors of regression coefficients from the weighted least squares were smaller than those from the regular least squares.

Table 5.—Mean square error (MSE)* of linear (unweighted and weighted) and nonlinear (exponential and power function) models for relationship between batch fecundity and fish weight of northern anchovy, based on data sets by years.

	Regression models				
	Linear		Nonlinear		
Year	Unweighted ($\times 10^{-6}$)	Weighted	Exponential ($\times 10^{-6}$)	Power function	Sample size (n)
1951-60	7.6	9.0	7.3	7.5	19
1978	9.6	8.3	10.2	8.1	23
1979	6.5	7.4	7.8	8.3	44
1980	1.6	1.5	1.5	1.5	33
1981	6.4	5.6	7.1	6.4	127
1982	6.8	4.9	6.7	6.6	109
1983	1.7	1.9	1.9	1.6	83
1984	2.2	1.3	3.5	2.3	87

*MSE = $\Sigma(\hat{Y}-\hat{\bar{Y}})^2/(n-2)$ where $\hat{Y}$ is the estimated batch fecundity from subsamples. MSE, rather than R^2, is used to measure the fitness of the model because R^2 is inappropriate for nonlinear equations.

Precision of Batch Fecundity Estimation and the Numbers of Females

In the previous sections, we have considered the optimum number of ovarian tissue samples of northern anchovy relative to costs and have examined various regression models for biases in expressing the fecundity-fish weight relationship. This analysis indicated that if two or three tissue samples per fish are taken, further precision can be obtained only through increasing the numbers of fish in the sample, and we also showed that for anchovy the simple linear model is the preferred regression model. The objective of this section is to determine the effect of the number of females on the precision of the regression estimates of batch fecundity ($\hat{Y}$) using the linear model. All the historical data for batch fecundity in the northern anchovy (1951-84) were used in this analysis.

The average batch fecundity for the spawning population can be estimated ($\hat{\bar{Y}}_{\bar{\bar{w}}}$) from a regression model where $\bar{\bar{w}}$ is an average of ovary-free female fish weight for the survey. Batch fecundity is based

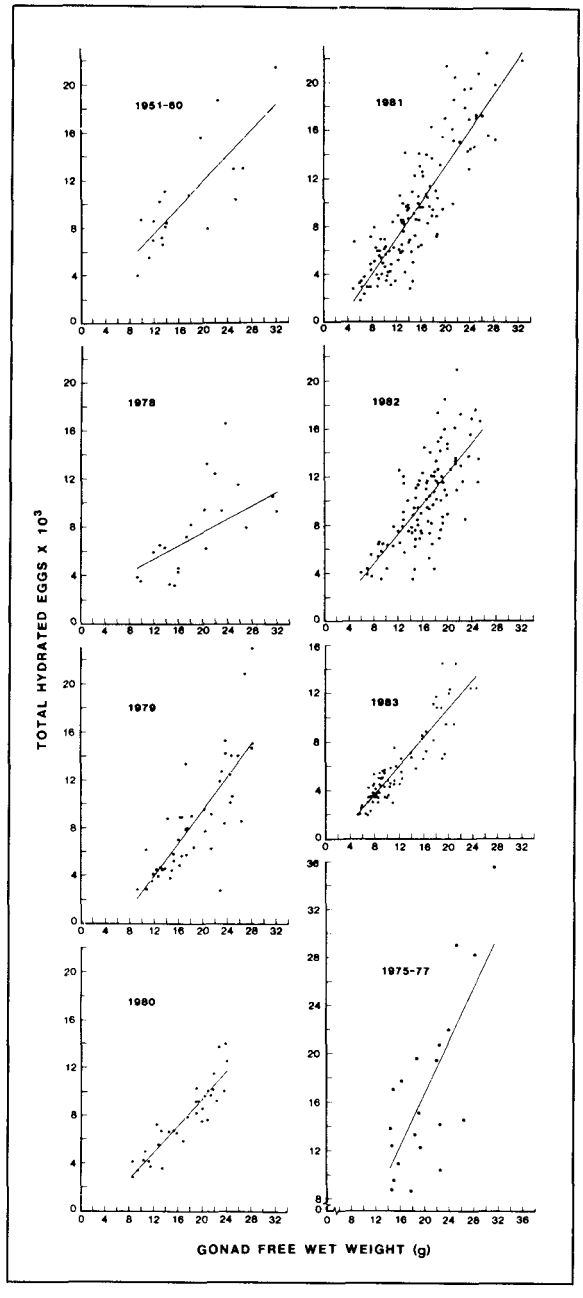

Figure 4.—Relation between batch fecundity of northern anchovy and their weight in formaldehyde solution (less ovary) for various years and subpopulations. All panels except lower right are for central subpopulation; lower right is for northern subpopulation, 1975-77, from Laroche and Richardson (1980). Fecundity estimates for the central population for 1951-60 are from MacGregor (1968) and are estimated using the oocyte frequency distribution method. Equations for lines are given in Table 4 for central subpopulation. For northern subpopulation $\hat{Y} = -5287 + 1098\ w$, $R^2 = 0.55$ (our calculation from Laroche and Richardson 1980).

on ovary-free weight whereas the weight of each female in the survey includes the ovary weight. Thus the mean weight of females ($\hat{w}$) in the survey must be adjusted to ovary-free weight to estimate the batch fecundity for the average female in the population. This is done by multiplying female weight by 0.95, i.e., $\bar{\bar{w}} = 0.95 \cdot \hat{w}$ (Hunter and Macewicz 1980) (Table 4). The average female weight in the fecundity samples ($\bar{w}$) may be different from $\bar{\bar{w}}$ because the fish used in the fecundity estimate are a small subsample of the thousands of mature fish taken in a survey. Hence, $\bar{\bar{w}}$ is preferable for biomass estimation. Generally speaking, $\bar{w}$ and $\bar{\bar{w}}$ are similar although occasionally the difference has been as great as 2 g.

The precision of the regression estimate of batch fecundity is measured by its coefficient of variation, $cv(\hat{Y}_w)$. The variance of $\hat{Y}_{w^*}$ for a particular fish weight w^* is equal to

$$\sigma^2 \left[\frac{1}{n} + \frac{(w^* - \bar{w})^2}{\sum (w - \bar{w})^2} \right] \tag{10}$$

where σ^2 is the variance around the regression line (estimated by s_ξ^2) and w^* is measured without error. To evaluate the effect of numbers of fish on the precision of the regression estimate, we calculated the cv of $\hat{Y}_{\bar{w}}$ and $\hat{Y}_{\bar{w}-2}$ for each of the data sets. Four elements affecting the variance of $\hat{Y}_{w^*}$ are σ^2, n, w^*, and the sums of squares ($\Sigma(w-\bar{w})^2$). Although the variances of fish weight (s_w^2) are quite similar among years, the sum of squares ($\Sigma(w-\bar{w})^2 = (n-1)s_w^2$) are different (Table 4). A small value of $\sigma^2_{\hat{Y}_{w^*}}$ results from either a large n and/or a wide range of fish weights. Increasing sample size n and/or increasing $\Sigma(w-\bar{w})^2$ reduces $\sigma^2_{\hat{Y}_{w^*}}$.

The coefficient of variance of $\hat{Y}_{\bar{\bar{w}}}$ was expressed as a function of the number of fish in the fecundity sample. For this analysis we used two versions of the power function ($cv(\hat{Y}_w|n) = \alpha \cdot n^\beta$): One in which $\bar{\bar{w}}$ is assumed to be the same as $\bar{w}$ and the other in which $\bar{\bar{w}}$ is equal to $\bar{w} - 2$ [Equations (11) and (12)]. In the latter equation, the cv of $\hat{Y}_{\bar{w}-2}$ sets the upper bound for the cv of $\hat{Y}_{\bar{\bar{w}}}$.

$$cv\left(\hat{Y}_{\bar{w}-2} \,|n\right) = 0.644\, n^{-0.735} \tag{11}$$

$$cv\left(\hat{Y}_{\bar{\bar{w}}} \,|n\right) = 0.438\ n^{-0.561} \tag{12}$$

For a sample size of 20 fish, the cv of $\hat{Y}_{\bar{\bar{w}}}$ falls between 0.07 and 0.08, and for a sample size of 60 it ranges between 0.03 and 0.04 [depending upon whether Equation (11) or (12) was used] (Fig. 5). Assuming that the coefficient of variation for the fecundity estimation should be less than 0.10, one should select a sample size that yields a cv of $\hat{Y}_{\bar{\bar{w}}} = 0.05$ because Equations (11) and (12) assume the fish weight (w) is measured without error; thus, for northern anchovy, a sample size of 50-60 females is adequate. In the biomass estimation, the cv for the fecundity estimation is a function of variances of both $\hat{Y}_{\bar{\bar{w}}}$ and $\bar{\bar{w}}$. The variance formula for the fecundity estimate including variance of $\bar{\bar{w}}$ is given below.

$$\sigma^2_{\hat{Y}_w} = \sigma^2_{\left[\bar{Y} + b(\bar{\bar{w}} - \bar{w})\right]} = \sigma^2 \left\{ \frac{1}{n} + \frac{(\bar{\bar{w}} - \bar{w})^2}{\sum (w - \bar{w})^2} \right\} + b^2 \sigma^2_{\bar{\bar{w}}} \tag{13}$$

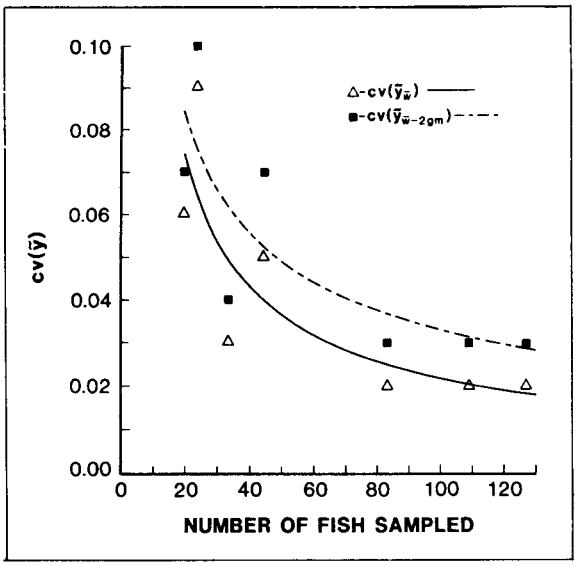

Figure 5.—Coefficient of variation (*cv*) of the regression estimates of batch fecundity for the average female anchovy weight as a function of the number of fish used to estimate fecundity in various years (see Table 4). Solid line is the *cv* for the average weight of females in the fecundity sample, and dashed line is the upper bound of the *cv* for the average female weight in the population.

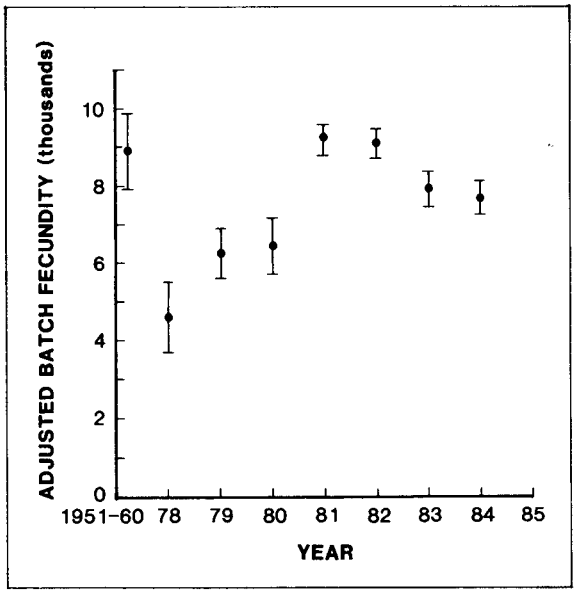

Figure 6.—Adjusted batch fecundity estimates for average female weight equal to 14.95 g, and the 95% confidence interval (represented by vertical bars) by years (see Table 4).

Variation of Batch Fecundity Among Years

The relation between batch fecundity and fish weight has been estimated annually for the egg production biomass estimation of northern anchovy because we believed that the relationship could change from year to year. Otherwise one equation would suffice for all years. To test this assumption, we performed an analysis of covariance to compare batch fecundity per female weight (regression coefficients, *b*) among years. The results show that not all slopes are the same, and have ranged from 279 eggs/g in 1978 to 752 eggs/g in 1981 (Table 4). Moreover, the average batch fecundity for a female of 14.95 g (the mean weight of all females in the fecundity samples when all samples are combined) also differs significantly among years (Fig. 6). The average number of eggs produced per spawning by a standard female of 14.95 g has varied by a factor of 2 over the last 7 yr (1978-84). This does not take into account the variation in average weight of females in the spawning population which also shows significant interannual variation. No doubt exists that batch fecundity of the northern anchovy population varies interannually and that it is necessary to estimate batch fecundity for each biomass estimation. On the other hand, if an average fecundity for all years (1951-84) is necessary, the following equation can be applied: $\bar{Y}_w = -1104 + 614\ w$.

SUMMARY

1. Laboratory methods are described for estimating the batch fecundity of fishes with indeterminate annual fecundity using counts of the number of hydrated eggs within weighed tissue samples of the ovary.

2. In northern anchovy, location of the tissue sample within the ovary has no effect on counts of hydrated eggs. Location of tissue samples may be important in fishes with larger ovaries or possibly in anchovy in the earliest stages of hydration. Thus, position effects must be evaluated for each species and sampling time.

3. The optimum number of tissue samples was 2 or 3 per ovary in northern anchovy, but a higher number may be necessary if position effects exist.

4. To maintain the coefficient of variation for the average fecundity of the population at less than 10% requires a sample of 50 or more females.

5. In northern anchovy, a simple linear regression model was preferable to nonlinear models for expressing the fecundity-female weight relation but this must be evaluated for each population.

6. For the egg production estimates, the average batch fecundity of the population can be calculated from the average weight of the females in the population (based on a weighted sample mean) using batch fecundity-female weight regression model.

7. The batch fecundity of northern anchovy has varied significantly among years (1951-84) indicating the batch fecundity-fish weight relation must be estimated for each egg production estimate.

LITERATURE CITED

BAGENAL, T. G.
1967. A short review of fish fecundity. *In* S. D. Gerking (editor), The biological basis of freshwater fish production, p. 89-111. John Wiley, N.Y.

BLAXTER, J. H. S., and J. R. HUNTER.
1982. The biology of the clupeoid fishes. Adv. Mar. Biol. 20:1-223.

CASSIE, R. M.
1954. Some uses of probability paper in the analysis of size frequency distribution. Aust. J. Mar. Freshwater Res. 5:513-522.

DeMARTINI, E. E., and R. K. FOUNTAIN.
1981. Ovarian cycling frequency and batch fecundity in the queenfish, *Seriphus politus*: Attributes representative of serial spawning fishes. Fish. Bull., U.S. 79:547-560.

DRAPER, N. R., and H. SMITH.
1981. Applied regression analysis, 2d ed. John Wiley, N.Y., 709 p.

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.

HARDING, J. P.
1949. The use of probability paper for the graphical analysis of polymodal frequency distributions. J. Mar. Biol. Assoc. U.K. 28:141-153.

HICKLING, C. F., and E. RUTENBERG.
1936. The ovary as an indicator of the spawning period in fishes. J. Mar. Biol. Assoc. U.K. 21:311-317.

HISLOP, J. R. G.
1975. The breeding and growth of whiting, *Merlangius merlangus* in captivity. J. Cons. int. Explor. Mer 36:119-127.

HISLOP, J. R. G., A. P. ROBB, and J. A. GAULD.
1978. Observations on effects of feeding level on growth and reproduction in haddock, *Melanogrammus aeglefinus* (L.) in captivity. J. Fish. Biol. 13:85-98.

HOLDEN, M. J., and D. F. S. RAITT.
1974. Manual of fisheries science. Part 2—Methods of resource investigation and their application. FAO Fish. Tech. Pap. 115 (Rev. 1), 214 p.

HUNTER, J. R.
1985. Preservation of northern anchovy in formaldehyde solution. *In* R. Lasker (editor), An egg production method for estimating spawning biomass of pelagic fish: Application to the northern anchovy, *Engraulis mordax*, p. 63-65. U.S. Dep. Commer., NOAA Tech. Rep. NMFS 36.

HUNTER, J. R., and S. R. GOLDBERG.
1980. Spawning incidence and batch fecundity in northern anchovy, *Engraulis mordax*. Fish. Bull., U.S. 77:641-652.

HUNTER, J. R., and R. LEONG.
1981. The spawning energetics of female northern anchovy, *Engraulis mordax*. Fish. Bull., U.S. 79:215-230.

HUNTER, J. R., and B. J. MACEWICZ.
1980. Sexual maturity, batch fecundity, spawning frequency, and temporal pattern of spawning for the northern anchovy, *Engraulis mordax*, during the 1979 spawning season. Calif. Coop. Oceanic Fish. Invest. Rep. 21:139-149.
1985. Measurement of spawning frequency in multiple spawning fishes. *In* R. Lasker (editor), An egg production method for estimating spawning biomass of pelagic fish: Application to the northern anchovy, *Engraulis mordax*, p. 79-94. U.S. Dep. Commer., NOAA Tech. Rep. NMFS 36.

JOSEPH, J.
1963. Fecundity of yellowfin tuna (*Thunnus albacares*) and skipjack (*Katsuwonus pelamis*) from the eastern Pacific Ocean. Inter-Am. Trop. Tuna Comm., Bull. 7:255-292.

LAROCHE, J. L., and S. L. RICHARDSON.
1980. Reproduction of northern anchovy, *Engraulis mordax*, off Oregon and Washington. Fish. Bull., U.S. 78:603-618.

LeCLUS, F.
1977. A comparison of four methods used in fecundity determination of the pilchard, *Sardinops ocellata*. Fish. Bull., S. Afr. 9:11-15.
1979a. Dry mass yolked oocytes of the South West African pilchard *Sardinops ocellata* in relation to maturity stages and spawning cycles, 1972-1974. Investl. Rep. Sea Fish. Brch. S. Afr. 119, 29 p.
1979b. Fecundity and maturity of anchovy *Engraulis capensis* off South West Africa. Fish. Bull., S. Afr. 11:26-38.

MacGREGOR, J. S.
1957. Fecundity of the Pacific sardine (*Sardinops caerulea*). Fish. Bull., U.S. 57:427-449.
1968. Fecundity of the northern anchovy, *Engraulis mordax* Girard. Calif. Fish Game 54:281-288.

SCHEFFÉ, H.
1959. The analysis of variance. John Wiley, N.Y., 477 p.

YAMAMOTO, K.
1956. Studies on the formation of fish eggs. I. Annual cycle in the development of ovarian eggs in the flounder, *Liopsetta obscura*. J. Fac. Sci. Hokkaido Univ., Ser. VI, Zool. 12.

ADDENDUM

Since this volume went to press an additional estimate of the biomass of the central subpopulation of northern anchovy was made in 1985. Batch fecundity parameters (listed in Table 4) for 1985 were as follows: n, 85; b, 682; a, −2,036; $s_{y \cdot x}$, 1,936; $\bar{Y}$, 8,490; $\bar{w}$, 15.43; s_w, 4.26; $(w-\bar{w})^2$, 1,524; $\bar{\bar{w}}$, 14.50; $s_{\bar{w}}$, 0.32; adjusted mean batch fecundity for 14.95 g fish, 8,156; and standard error of batch fecundity for 14.95 g fish, 211.