Baitfish Stock Assessment Using the Egg Production Method: an application on the Hawaiian anchovy or nehu (*Encrasicholina purpurea*)

D.A. Somerton*

Abstract

The biomass of nehu (*Encrasicholina purpurea*) in Pearl Harbor was estimated weekly, over a two year period, using a new stock assessment procedure known as the Egg Production Method (EPM). Although the EPM was originally developed for assessing relatively long-lived, temperate anchovies (Northern Anchovy and Anchovetta), it has proven to be a low-cost, effective way of assessing the abundance of nehu, a short-lived, tropical stolephorid anchovy. The effectiveness of the EPM is largely the result of its being based on the life history stage that is the least aggregated and easiest to sample, that is, the egg stage. Because of this, the biomass estimates obtained using the EPM are less influenced by environmental fluctuations than estimates obtained using commercial catch statistics. Over the study period, nehu spawning stock biomass varied between 0.5 and 5.0 tonnes and was clearly associated with the variation in the rate of nehu bait catch for the pole-and-line tuna fishery. Stock variation also was associated with seasonal variation in reproductive output, primarily changes in weight-specific fecundity and spawning frequency and in the survival rate from the egg stage through the first feeding larvae.

STOCK assessment of baitfishes is difficult because fishery-dependent information such as catch per unit effort is often not a good indicator of relative abundance (Wetherall 1977), and fishery-independent information such as hydroacoustic data is expensive to obtain. One promising new approach, called the Egg Production Method (EPM), was developed to assess the biomass of small schooling species from the abundance of their pelagic eggs. The basic concepts of the EPM were first developed by Saville (1964) and later elaborated by Parker (1980). The estimate of biomass can be expressed algebraically as:

$$B = \frac{P}{F\,R} \qquad \text{Equation 1}$$

where B is the biomass, P is the daily production of eggs by the population, F is the fecundity per gram body weight of spawning females, and R is the proportion, by weight, of spawning females in the population. Definition of the population, and therefore the calculation of R, can be in terms of mature fish (spawning stock) or fish larger than some minimum size (commercial stock). In practice, P is estimated by sampling pelagic eggs with a plankton net and F and R are estimated by sampling adult fish.

The EPM was initially applied to the California anchovy (*Engraulis mordax*; Lasker 1985) and anchovetta (*E. ringens*; Alheit 1985), both relatively large, long-lived, temperate species, but it is also ideally suited for stock assessment of the smaller, shorter-lived anchovies and sprats used as baitfishes by the tuna pole-and-line fisheries. One such species is the Hawaiian anchovy or nehu (*Encrasicholina purpurea*). Nehu are endemic to the Hawaiian Islands and occur exclusively within enclosed bays, especially those that receive significant freshwater input. Typical of many tropical

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* National Marine Fisheries Service, Honolulu Laboratory, Southwest Fisheries Center, NOAA, Honolulu, Hawaii, U.S.A.

stolephorid anchovies (Dalzell 1987), nehu are short-lived (Struhsaker and Uchiyama 1976) and spawn almost continuously throughout the year (Tester 1955, Clarke 1987). Unlike most other baitfishes, however, nehu occur in shallow, turbid areas during the day and migrate at night to deeper, clearer areas where they spawn and feed. The commercial fishery exploits this unusual behaviour and captures nehu with seines in their shallow daytime habitat (Uchida 1977).

In this paper an application of the EPM is applied to a population of nehu occurring within Pearl Harbor, Oahu, emphasising aspects which differentiate this from previous applications on temperate species. As nehu have a high biological turnover rate and are subjected to an intensive fishery, stock assessment must be frequent to resolve important aspects of population change. For this reason, nehu stock assessment surveys were conducted weekly rather than yearly as they have been for temperate species. In addition, the application of the EPM on nehu differed from previous studies (Picquelle and Stauffer 1985) in the methods used to estimate F and R. These differences primarily reflect the biological differences between nehu and temperate anchovies but also include measures intended to reduce the time requirements of sampling and sample processing.

Materials and Methods

Pearl Harbor was sampled weekly from 3 April 1986 to 7 April 1988. Each sampling day was separated into two periods: egg and plankton sampling and adult sampling. Eggs and plankton were sampled between 0700 and 1200 hours with a systematic sampling design (Cochran 1963) in which a single sample was taken near the geographic centre of each of 39 strata. The net used was 5 m long and 1 m in diameter at the mouth and constructed of 335 μm Nitex[1]. To allow its use from small boats, the net was not towed but instead was thrown overboard and sampled as it descended to the bottom (average depth of the sampling sites was about 12 m). Compared with a typical towed plankton net, our sampling net had two design features added to increase its effectiveness. First, to increase the sinking speed, lead weights (5 kg) were attached to the steel ring that held the mouth open. Second, to ensure closure of the net, a retrieval line was attached, not to the ring itself, but to a choke collar surrounding the net. Mesh bags of 335 μm Nitex were used as cod ends. When retrieved, these bags were sealed and placed in a 10% buffered formaldehyde solution.

In the laboratory, the plankton samples were examined without subsampling, and all nehu eggs and all large crustacean zooplankters, previously identified as preferred nehu food (Hiatt 1951), were counted. Egg and plankton densities at each station were estimated by dividing the counts by an estimate of the filtered volume computed as water depth multiplied by the mouth area of the net.

Adult nehu were sampled in a 1.5-hour period preceding sunset when the eggs to be spawned that evening were clearly hydrated and easily distinguished from unhydrated, mature eggs (Clarke 1987). Adults were captured, along with juveniles, using a beach seine of the same mesh size (9 mm) and design characteristics as a commercial nehu seine except that it was cut to approximately one-third scale (70 m long by 3 m deep) so that it could be set from a small boat. Nehu schools were located in their preferred daytime habitat, that is, shallow, turbid areas, with the same searching techniques used by commercial vessels. Schools were quickly surrounded with the seine, and a random sample of the catch was taken and immediately preserved in a 10% buffered formaldehyde solution.

In the laboratory, a subsample of nehu was drawn from each field sample: 1) An initial subsample of approximately 50 fish was randomly chosen. 2) In order of size, starting with the apparent largest individual, fish were removed from the subsample and examined microscopically for sex and stage of maturity. 3) If less than 25 mature females were obtained from the initial subsample, additional subsamples were drawn and examined completely in the same manner until at least 25 mature females were obtained. 4) To increase the speed of classifying fish, if six juveniles were drawn in succession from a subsample, all of the remaining fish in that subsample were also classified as juveniles without examination. Because individuals were chosen according to size, the remaining fish were smaller than any that had been visually classified and therefore were likely to be immature.

The criteria used to classify fish were based upon the following gross morphological features partly adapted from Clarke (1987):

- largest ova measuring ⩾0.7 mm in length indicated a mature female with hydrated eggs;
- largest ova measuring 0.5–0.7 mm in length indicated a mature female with unhydrated eggs;
- testis depth (dorsal-ventral) measuring ⩾ eye diameter indicated a mature male;

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[1] Reference to trade names does not imply endorsement by National Marine Fisheries Service, NOAA.

- largest ova measuring < 0.5 mm or testis depth measuring $<$ eye diameter indicated a juvenile.

After the subsample was chosen and classified, the following biological attributes were measured. Standard length (in millimetres) was measured for all mature fish and for the first 25 randomly chosen juveniles; any remaining juveniles were counted but not measured. Total body weight (in milligrams) was measured, after blot drying, for all fish measured for length except females with hydrated eggs; ovary-free body weight was measured for all females. Batch fecundity was estimated for females with hydrated eggs by teasing apart both of the ovaries and counting under a microscope all hydrated ova. Each count was replicated, and fecundity was estimated as the mean of the two counts. Over the 105 consecutive weeks of sampling, this process was repeated for each of 162 seine samples.

Data analysis and biomass estimation

Evaluation of Equation 1 requires estimates of *P*, *F*, and *R*. These estimates were obtained as follows. Daily egg production was estimated as

$$P = \Sigma\, D_i V_i; \qquad \text{Equation 2}$$

where D_i is the egg density at the ith station, and V_i is the volume of the ith stratum. The summation is over all 39 strata. Weight specific fecundity was estimated as

$$F = \frac{\Sigma E}{\Sigma W_{fh}} \qquad \text{Equation 3}$$

where ΣE is the sum of all hydrated ova, and ΣW_{fh} is the sum of the corrected total body weight of mature females with hydrated eggs. Spawning proportion was estimated as

$$R = \frac{\Sigma W_{fh}}{\Sigma W_m + \Sigma W_{fh} + \Sigma W_{fu}} \qquad \text{Equation 4}$$

where ΣW_m and ΣW_{fu} are the sums of the total body weights of males and of females with unhydrated eggs. The W_{fh} were corrected for the temporary weight gain due to egg hydration by regressing W_{fu} on ovary-free weight for females with unhydrated ovaries, then predicting W_{fh} from the ovary-free body weights of females with hydrated ovaries. When the commercial population rather than the spawning population was estimated, Equation 4 was modified slightly to include in the denominator the term ΣW_j or the sum of the total body weights of juveniles.

Variance and bias of the estimated biomass were approximated by using the delta method (Seber 1973). The variance estimator was

$$V(B) = B^2[CV(P)^2 + CV(F)^2 + CV(R)^2]; \qquad \text{Equation 5}$$

and the bias estimator was

$$\text{bias}(B) = B[CV(F)^2 + CV(R)^2]; \qquad \text{Equation 6}$$

where *CV* represents the coefficient of variation.

The variances needed to evaluate Equations 5 and 6 were estimated as follows. The variance of the daily egg production, *P*. was calculated as

$$V(P) = \Sigma V_i^2 V(D_i); \qquad \text{Equation 7}$$

where $V(D_i)$ was approximated as the variance in D_i among three adjacent stations, because the plankton net samples were not replicated at each station. The variances of the biological parameters were estimated as

$$V(\bar{X}) = \frac{\Sigma(Xi - \bar{X})^2}{N(N - 1)} \qquad \text{Equation 8}$$

where X_i is the ith sample mean for one of the biological parameters and $\bar{X}$ is the mean of N sample means. Since the number of samples collected each week was always small and sometimes even insufficient to estimate variance, the weekly estimates of the means and variances of the biological parameters were calculated from a pool of all samples within a 5-week moving time window centred on the particular week.

Results and Discussion

Biomass estimates

Weekly spawning and commercial biomass estimates (Figure 1) had similar patterns of variation over time, but the commercial biomass estimates were always larger with wider confidence intervals. Over the 2-year period, the population fluctuated seasonally with maximum abundance in March-April and perhaps again in late July-August and a minimum in November. This pattern of variation was similar to that observed in the abundance of nehu eggs in Kaneohe Bay, Oahu (Tester, 1955).

Egg production

Estimates of daily egg production can be influenced by two sources of bias. First, an overestimation can occur if the eggs of other fishes are misidentified as the target species. For nehu, such misidentification is unlikely because no other fishes in Pearl Harbor have eggs similar to those of nehu (Tester 1951; Clarke 1987). However, in areas where several closely related species spawn

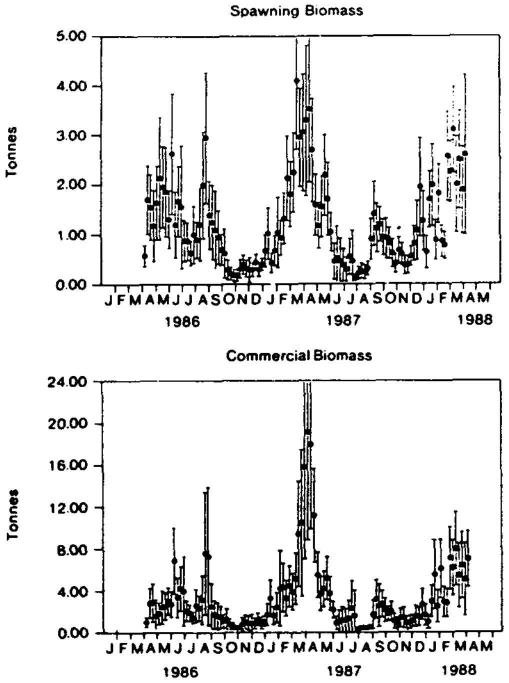

Figure 1. Weekly estimates and 95% confidence intervals of the spawning and commercial biomasses of nehu within Pearl Harbor in metric tonnes.

concurrently, egg identification may be problematical. Second, an underestimation can occur if egg mortality is high and the time between spawning and sampling is large. In northern anchovy, for example, the egg mortality rate was estimated at 53% per day (Smith and Lasker 1978), but we believe that nehu egg mortality is substantially less.

Egg mortality can be attributed to either predation or abnormal embryological development. Predation by the adults accounted for 32% of the egg mortality in northern anchovy (Hunter and Kimbrell 1980) but is probably much lower in nehu because eggs are rarely found in adult gut samples (Hiatt 1951). Mortality due to abnormal development could be significant in nehu but would be difficult to quantify because, based on our preliminary studies of egg density, dead nehu eggs sink rapidly in water with densities typical of Pearl Harbor and thus, are likely to escape capture. Because some egg mortality is likely, we believe that our estimates of egg production, based on the abundance of eggs roughly 16 hours after spawning, may be too low.

Reproductive parameters

Previous applications of the EPM (Alheit 1985; Hunter and Macewicz 1985) have used postovulatory follicles to identify spawning females. Instead, we chose to use the presence of hydrated eggs, primarily because histological preparation is not required and identification is, therefore, faster and easier. The use of hydrated ova, however, imposes a restriction on the time that the population can be sampled, to ensure correct identification of spawning females. This restriction reduces the level of precision in the biomass estimates that might have otherwise been achieved and subjects the estimates of spawning proportion to a potential bias.

The appropriate period for sampling adults is bounded between the time of day when hydrated ova are sufficiently enlarged to be recognisable and sunset when migration to deeper water begins (Clarke 1987). Recognisable hydrated ova may occasionally be observed during the early afternoon, but Clarke (1987) has found that not until two hours before sunset did all prespawning females have some ova $\geq$ 0.7 mm. Since the definition of the correct time window for sampling is so important, our study repeated part of Clarke's (1987) study by sampling adult nehu at approximately half-hour intervals throughout the late afternoon and found that a smaller time window of 1.5 hours was required to ensure correct identification. This time period was so short that we were only able to obtain an average of 1.6 adult samples each week and occasionally experienced periods in which no samples could be obtained.

Not only is the time period for adults sampling short, but it occurs immediately before spawning when segregation of sexually active and inactive fish may occur. Previous studies on northern anchovy (Picquelle and Stauffer 1985) and anchovetta (Alheit 1985) have found that active females segregate from inactive females and aggregate with males near the time of spawning. For nehu, segregation of active from inactive females, aggregation of active females with males, and segregation of adults from juveniles were tested and all were highly significant (chi-square test of homogeneity; $P<0.001$). This means that some segregation had occurred by the time the adult sampling period began. Such segregation increases biological heterogeneity among schools, and thereby increases the variance in the estimates of the proportion spawning.

In addition to its influence on the precision of the biomass estimates, the use of hydrated ova to identify spawning females is subject to one important source of sampling bias: differential

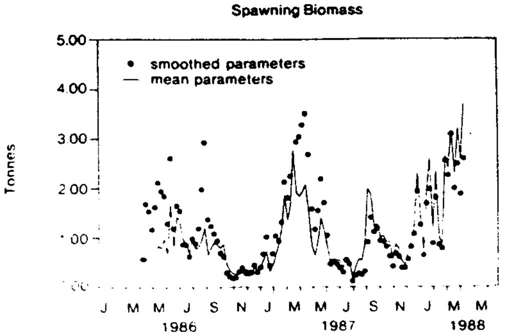

Figure 2. Spawning biomass calculated by using the smoothed weekly mean estimates of the adult parameters (F and R) and by using the total overall mean of the adult parameters.

catchability between sexually active and inactive females. For both northern anchovy (Picquelle and Stauffer 1985) and anchovetta (Alheit 1985), active females have a higher vulnerability to the sampling gear, perhaps due to a difference either in net avoidance or in depth distribution. For nehu, however, active and inactive females are apparently equally vulnerable, at least during the adult sampling period, because both occur in the shallow, nearshore areas sampled during the day (Clarke 1987). Furthermore, using postovulatory follicles and estimated rates of ova development from fish sampled at all times of day, Clarke (1987) estimated that individual nehu spawn every other day or, expressed as the inverse, that nehu have a daily spawning proportion of approximately 0.50. Since our estimate of mean proportion spawning, 0.53 (95% confidence interval, ± .04), is not significantly different from Clarke's (1987), it is unlikely that our sampling was biased.

Considerations for other baitfish applications

Other applications of the EPM on baitfishes are likely to also require frequent sampling, but lack sufficient resources for processing a large number of adult samples. Our approach to this problem was to save time by using hydrated ova rather than postovulatory follicles to identify spawning females. However, other shortcuts could be taken, especially when there is little temporal variation in adult parameters. One approach is to save time in sampling and sample processing by calculating biomass estimates based on long-term mean parameter values instead of weekly estimates. To determine how accurate such biomass estimates would have been for nehu, we compared the estimates calculated using the smoothed parameter values to those computed using the overall mean parameter values. The results indicated that roughly 58% of the variability in spawning biomass and 35% of the variability in commercial biomass were explained by the variability in the biomass estimates computed from long-term means. Much of the loss in predictability, however, was at high frequencies, while most of the low frequency and seasonal fluctuations are preserved (Fig. 2). For still further savings in time, this approach could be extended by simply eliminating adult sampling altogether and treating egg abundance as a proxy for fish abundance similar to the way catch per unit effort is used.

Factors influencing nehu population fluctuations

Since the nehu population in Pearl Harbor is closed, receiving little or no immigration (Tester and Hiatt 1952), the observed variation in abundance (Fig. 1) must be due to variations in the recruitment of juveniles and the mortality rate of adults. Recruitment to the nehu population depends to some degree upon the biomass of adult females, the production of feeding larvae per gram of female, and the survival of feeding larvae to adulthood. Although survival of feeding larvae in our study was not considered, the variation in larval production was examined. Larval production per gram of female can be considered as the product of three components: 1) the production of eggs per gram of female body weight (Fig. 3d), 2) the proportion of the population spawning each day (Fig. 3c), and 3) the survival of eggs through the stage of first feeding larvae (Fig. 3e). All three of these components are seasonal, but their product, that is, the production of feeding larvae per gram of adult female (Fig. 3f) has a conspicuously strong seasonal pattern that is similar to those of the mean water temperature within Pearl Harbor (Fig. 3a) and to the density of the crustacean zooplankters that are the primary component of nehu food (Fig. 3b).

The total mortality rate of adults is the sum of both natural and fishing mortalities. Based on our preliminary studies of the Pearl Harbor population, natural mortality appears to be nearly constant over time, but fishing mortality varies greatly. Since the catch rises and falls almost synchronously with the estimated spawning biomass (Fig. 1), the fishery is bait limited and responds quickly to changes in bait availability. This response occurs in two ways. First, when bait abundance increases, the bait catch of individual vessels tends to increase, at least until capacity is reached. Second, more vessels fish in Pearl Harbor when the catch rates there are high relative to other baitfishing areas. The link between changes

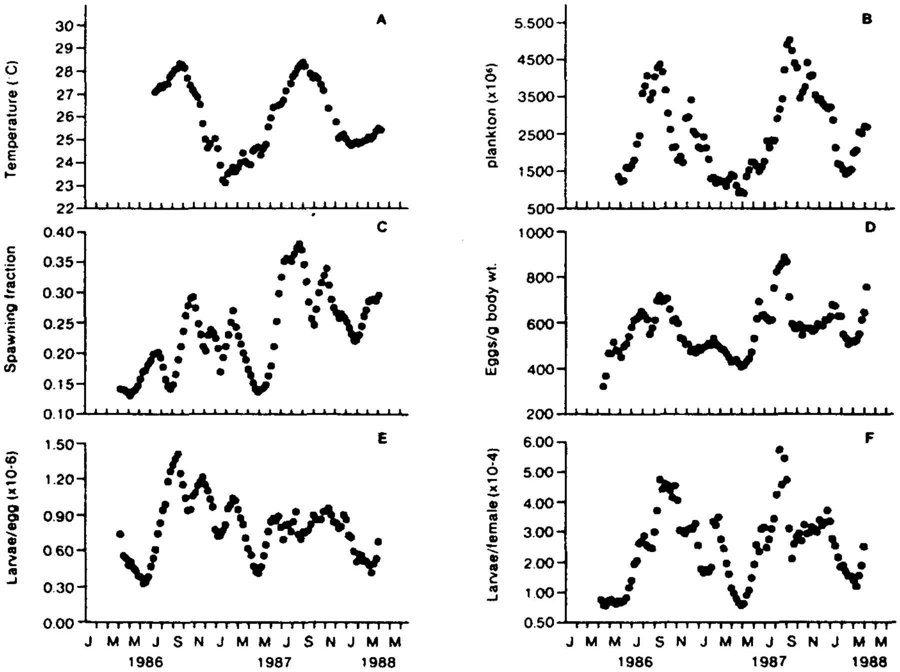

Figure 3. Weekly estimates of A) mean water temperature in Pearl Harbor, B) total abundance of all crustacean zooplankters identified as nehu food, C) proportion of the population, by weight, comprised of spawning females, D) batch fecundity expressed as eggs per gram body weight of spawning female, E) a crude estimate of early larval survival calculated as the total number of feeding larvae caught in one week divided by the number of eggs produced in the previous week, and F) number of feeding larvae produced by 1 gm of spawning female, that is, the product of C, D, and E.

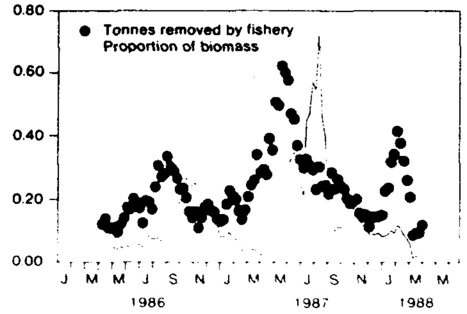

Figure 4. Catch of nehu within Pearl Harbor, expressed in tonnes and as a proportion of the estimated commercial biomass. This proportion is also known as the exploitation rate.

in effort and changes in nehu abundance is subject to inertia, and for a time, effort can remain high, or even increase, when the population begins to fall. As a result, the proportion of the population taken, or the exploitation rate, increases markedly (Fig. 4) and the abundance of nehu rapidly falls.

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