FECUNDITY AND SPAWNING FREQUENCY OF THE HAWAIIAN ANCHOVY OR NEHU, *ENCRASICHOLINA PURPUREA*

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ABSTRACT

Female nehu can begin spawning at 35 mm standard length; almost all fish over 40 mm SL from Kaneohe Bay were mature and in spawning condition. Mature females were found in all months of the year. Females from summer (May-October) had higher fecundity and relative cost per batch than fish from winter (November-April). In nehu and most other anchovies, fecundity appears to increase exponentially with weight. Nehu appear to be distinguished from other species by a higher exponent and consequently greater increase in relative fecundity over the reproductive size range. Nehu spawn during a short period 1 or 2 hours after sunset and begin hydrating ova only a few hours before spawning. Data on presence or absence of hydrated ova or postovulatory follicles along with differences in oocyte size in fish collected from throughout the diel cycle indicated that, after spawning, nehu can ripen a new batch of oocytes in 2 days and that most females spawn every other day. The estimated requirements for continued spawning at this rate indicate that individual variation in recent feeding success or stress could be responsible for observed scatter about fecundity-weight relationships and deviation from the normal spawning frequency.

The nehu, *Encrasicholina purpurea*, is a small anchovy endemic to the Hawaiian Islands. It is one of the dominant planktivorous fishes in enclosed, semi-estuarine areas and is the major source of bait for the local skipjack tuna fishery. Nehu are short-lived; growth increments on otoliths indicate a maximum age of about 6 mo (Struhsaker and Uchiyama 1976). Leary et al. (1975) showed that nehu can reach maturity at 35 mm standard length (SL) and presented fecundity data for 41 females. Leary et al. found very few females with hydrated ova and, on that basis, suggested that nehu spawn only once per lifetime.

Reexamination of Leary et al.'s (1975) conclusions was prompted both by the great variability in their fecundity vs. weight relationship and by discovery in recent collections that female nehu with hydrated ova are not at all rare, but rather are found only at restricted times of the day. This paper presents results of more detailed investigations of fecundity and spawning frequency in nehu in order to compare and contrast aspects of reproductive output of a tropical anchovy with those of better studied temperate species.

MATERIALS AND METHODS

All nehu examined for this study were collected from Kaneohe Bay, HI. Day samples were collected by beach seine or dip net in shallow water (1-2 m deep) or were taken from bait recently collected from similar areas by skipjack tuna vessels. Night samples were taken by blind sets with a ca. 67 m long by 13 m deep purse seine over deeper (12-14 m) areas of the bay. Forty-four night samples and two day samples were taken in 1974-79, while 5 night samples and 18 day samples were taken in 1983-85. Samples with adult nehu were available from all months of the annual cycle and, for most months, from at least two different years. One or more samples with adults were available from all hours of the diel cycle except the period between midnight and dawn, when there were few samples and very few adults collected.

In order to follow short-term oocyte development in the same group of fish, on two occasions a school of nehu was surrounded with a 60 m long beach seine in shallow water and sampled initially and twice later in the day. Samples were taken at the hours of 1300, 1500, and 1700 on 13 January 1984 and at 1000, 1300, and 1600 on 27 January 1984. Although the school was obviously disrupted by initial surrounding and subsequent dipnetting of samples, the fish held in the net appeared to resume normal daytime behavior shortly after each disturbance and spent most of the time loosely schooled with other nehu on the outside of the net. The oocyte size-frequency data from these samples did not differ in any obvious manner from data taken from other

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Manuscript accepted November 1986.
FISHERY BULLETIN: VOL. 85, NO. 1, 1987.

samples at approximately the same times on other days; consequently, the data from these "enclose and hold" samples were pooled with the others for all analyses.

Time of collection was recorded as the beginning of the set of the net; usually 15-30 min elapsed before the sample was actually preserved. For samples taken from skipjack tuna vessels, the time of collection was often only known within ±15 min and the delay between collection and preservation of the sample was often somewhat longer than 30 min.

For analyses of oocyte development rate and spawning frequency, collection time was adjusted to hours since the most recent spawning. Data on appearance of newly spawned eggs in the plankton (Clarke unpubl. data) indicate that spawning begins 1-2 h after sunset and is nearly over in about an hour; the delay after sunset is greatest during the summer. For samples considered here, spawning time was assumed to be 1 h after sunset for dates between mid-October and the end of April and 2 h after sunset for the remainder of the year. Given the frequent uncertainty in actual time of capture, this crude correction for spawning time was satisfactory for the purposes of the present study.

All specimens were preserved and held in ca. 4% formaldehyde/seawater solution. The recently collected samples were held at least 1 wk before measurement and further analyses; by this time most shrinkage in length had occurred. Although the older samples had been in preservative for several years, there was no evidence that long-term storage had affected any parameters considered here, e.g., length-weight relationships were similar for both recent and older samples.

For each sample, standard length (SL) of all or a subsample of ca. 100 specimens was measured to the nearest mm. Individuals for further examination were selected from throughout the size range of nehu >35 mm SL in the sample. The selected individuals were measured to the nearest 0.5 mm, opened, and the gonads examined under a dissecting microscope. Females were classed as immature —ovaries translucent and maximum oocyte length <0.40 mm; mature—ovaries mostly opaque, oocytes visibly yolked and over 0.40 mm; or hydrated—mature and at least some oocytes with translucent, globular yolk and the perivitelline space visible. For mature females the length of the apparent largest oocyte was estimated to the nearest 0.1 mm using an ocular micrometer.

To determine oocyte size frequency of mature females, a portion of the ovary was teased apart on a glass slide, placed under a compound microscope at 100×, and the lengths of oocytes over 0.40 mm measured to the nearest 0.01 mm until 20-30 of the largest oocytes were measured. Spawned nehu eggs are ellipsoidal with the length about twice the width (Yamashita 1951). Oocytes >0.3-0.4 mm are also elongate but are more variable in shape. "Length" as used here refers to the maximum dimension. Extremely elongate (length to width ca. 3 or more) and nearly round (length to width less than ca. 1.5) oocytes were noted as was the relative opacity of each oocyte measured. These observations were necessary in many cases to separate nearly round, heavily yolked oocytes that belonged to an advanced mode from very elongate, more nearly translucent oocytes of the same "length" that clearly belonged with a less developed mode.

As reported by Leary et al. (1975), mature female nehu may carry 0-2 separate size-frequency modes of oocytes. If a distinct advanced mode of oocytes was evident from the measurements and associated notes, the maximum, minimum, and median lengths of oocytes in this "largest" mode were used for subsequent analyses. These parameters will be abbreviated as LMX, LMN, and LMD, respectively. If all ova in the most advanced mode were hydrated, all lengths were arbitrarily assigned a value of 1 mm. If the largest mode was incompletely separated from smaller oocytes, only LMX and an estimate of LMD were recorded; if there was no separating mode evident, LMX (the largest oocyte in the subsample) was the only datum recorded. If an advanced mode was present and a second or "next" mode was also separated from yet smaller oocytes; the maximum, minimum, and median lengths of oocytes in the next mode will be abbreviated NMX, NMN, and NMD. In most females, however, the next mode was either only partially separated or not evident and, similarly to the case for unseparated advanced modes, only NMX and an estimate of NMD or only NMX, the largest oocyte not in the advanced mode, could be recorded.

For 107 specimens for which size-frequency measurements were made from a sample of the right ovary, the left ovary was prepared, sectioned, and stained with eosin/hemotoxylin as described by Hunter and Goldberg (1980). The slides, identified by only a code number, were examined for presence of postovulatory follicles (POF).

For determination of batch fecundity and dry weight, the fish was first rinsed with distilled water. The ovaries were removed and placed on a clean glass slide. Oocyte size frequency was determined as described above. If a distinct mode of advanced oocytes was present and oocytes in this mode could

be unequivocally discriminated under a dissecting microscope on the basis of size or opacity, the ovaries were teased apart and all ova in the advanced mode counted. This technique eliminated any error in fecundity determination due to subsampling of the ovaries, but meant that very few determinations were based on specimens with oocytes smaller than ca. 0.65 mm. In most of the latter cases, even if an advanced mode was clearly evident from the size-frequency determinations, it could not be unequivocally discriminated for total counts under the dissecting scope. Females with hydrated ova free from the follicles and segregated from the smaller oocytes were not used for fecundity determinations.

After the oocytes in the largest mode were counted, the entire ovaries were rinsed with distilled water from the slide into a preweighed aluminum pan. The stomach contents were removed from the fish and the body cavity was examined for parasites, specifically the presence of ca. 5 mm long nematodes around the liver and pyloric caeca. The fish was placed in a preweighed pan, and any tissue remaining on the slide was rinsed with distilled water into the same pan. The fish and gonads were dried at 60°C for 24 h after which the pans were reweighed to the nearest 0.1 mg, and dry weights of the fish and gonads determined by subtraction.

In all cases, fecundity and relative fecundity refer strictly to batch fecundity. Relative fecundity will be given as eggs per gram ovary-free dry weight, and gonad to somatic weight ratio (G/S) will be given as percent of dry weight values. Dry weights were used because of the difficulty in making consistent wet weight determinations on such small fish and even smaller ovaries. Careful wet-dry weight determinations on 10 females and gonads indicated that preserved nehu without gonads are about 73% water and that ovaries with yolked, but unhydrated, oocytes are about 60% water. To compare nehu fecundity data with those from other studies which had used wet weights, individual nehu dry weights were divided by 0.27, and relative fecundity and fecundity weight relationships were recalculated. This procedure admittedly ignored any variability in the wet-dry weight relationship. The G/S values given here can be multiplied by 0.675 (0.27/0.40) to make them roughly comparable to values based on wet weight from other studies.

Unless otherwise noted, all regressions given below are Model II (or "functional"), GM regressions (Ricker 1973). Results of regressions using natural logarithms are expressed as power curves (antilog form). The 95% confidence limits for slopes of linear regressions and exponents of power curves (= slopes of ln-ln regressions) were calculated from formulae in Ricker (1975). For any previous studies which had given results from Model I regressions, original fecundity and weight data were used to calculate functional regressions.

RESULTS

Maturity and Oocyte Development

The smallest mature females were 35 mm SL, the same minimum size reported by Leary et al. (1975), but in many of the samples most of the fish <40 mm SL were immature. Among the fish from the 36 samples from which more than cursory examinations were made, 30% of the 134 specimens <40 mm SL were immature. Only 8% of the 227 between 41 and 45 mm SL and <2% of the 284 over 45 mm were immature.

Nehu oocytes begin to elongate at about 0.3 mm in length but remain relatively translucent with little visual evidence of vitellogenesis until about 0.4 mm long. Oocytes longer than 0.5 mm were almost always opaque, and those over about 0.6 mm were densely opaque and yellow to yellow-brown in color. The first signs of hydration appeared in oocytes about 0.75 mm long. The yolk became more translucent and globular rather than granular in apparent texture, and the perivitelline space was evident at one or both ends. All ova longer than 0.8 mm were white in appearance and had an evident perivitelline space. At about this size or slightly larger, ova had left the follicles and begun moving to the main oviduct. Comparisons of fish from closely spaced purse seine samples taken just before and during spawning indicated that migration of hydrated ova from the follicles to the oviduct occurred in <0.5 h. Only in a few fish with the ova segregated or partially spawned were one or two hydrated ova left in the follicles. Apparently once hydration begins, all ova in a batch are normally ovulated and spawned at one time.

Separate batches of maturing oocytes become distinct from the numerous small oocytes between 0.45 and ca. 0.60 mm. In fish with LMX <0.45-0.50 mm there was little or no evidence of a separating size-frequency mode of oocytes. Variably separated modes with LMD at 0.45-0.55 mm were present in fish with the LMX at 0.55-0.65 mm. Usually modes centered at 0.60 mm or larger and with LMX over 0.65 mm were clearly separated from smaller and less opaque oocytes. There was no evidence from size-frequency data that, once oocytes reached ca.

0.65-0.70 mm long, any were "left behind" and not spawned with the ripening batch.

For 248 fish which either had a clearly defined and separated advanced mode of unhydrated oocytes or carried hydrated ova with a clearly defined and separated next mode, the largest (LMX or NMX) and median-sized (LMD or NMD) oocyte in the mode were significantly correlated ($r = 0.94$, $P < 0.01$), and the slope of the Model II regression was nearly 1 (1.042). The correlation was essentially unchanged by addition of data from 51 more fish where the median size of an incompletely separated mode was only estimated. These results indicate that, even if a mode is incompletely separated, the estimated LMD is a useful parameter and, furthermore, that for purposes of comparing different fish, LMX is as appropriate an indicator of size of oocytes in a mode as LMD. Consequently, in subsequent analyses of LMD data both unequivocal and estimated values were used, and in other cases LMX was used to analyze change in oocyte size during ripening. Both decisions were made primarily to include data from specimens with small oocytes and without completely or even partially separated modes.

Fecundity

Fecundity of 222 females 35-58 mm SL ranged from <100 to >1,600, and relative fecundity ranged from 432 to 4,098 eggs/gram. Although low relative fecundities were observed in samples from almost all months, most values over 2,000 were from fish taken in summer and fall (Fig. 1); consequently, the fecundity data from "winter" (November through April) and "summer" (May through October) fish were treated separately for all subsequent analyses. There were no significant differences in size composition between the summer and winter specimens (Kolmogorov-Smirnov test, $P > 0.20$). The mean relative fecundity for winter (1,363, $n = 93$, range = 496-2,763) was significantly different ($P < 0.01$, t-test) from that for summer (2,097, $n = 128$, range = 433-4,099). Regressions between fecundity and length or weight (Table 1) also indicated that winter fish were less fecund than summer fish.

When relative fecundity data for each season were partitioned according to LMD (<0.65 mm, 0.65-0.75 mm, and >0.75 mm), there were no significant differences between groups in the summer data (analysis of variance, $P > 0.05$), but there were significant differences in the winter data ($P < 0.001$). Inspection of the data indicated that the latter was due mostly to low values for the fish with LMD <0.65 mm. This could result from incomplete recruitment of oocytes to modes barely separated from smaller oocytes. There were, however, only 12 fish in this category, and the small sample size plus the absence of similar evidence in summer fish indicates

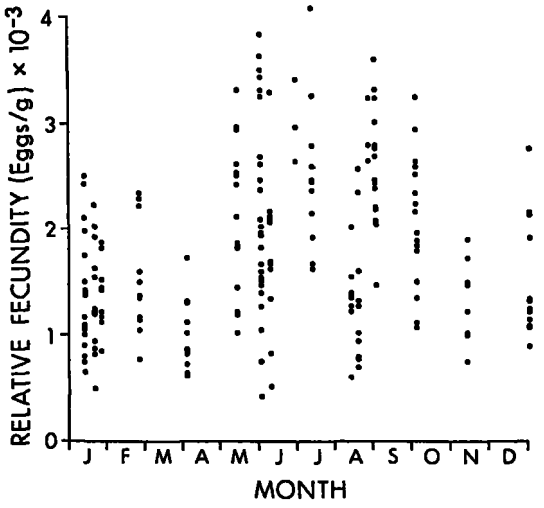

FIGURE 1.—Relative fecundity in thousands of eggs/g ovary-free dry weight vs. date of collection for 222 nehu from Kaneohe Bay, HI.

TABLE 1.—Summary of Model II regression statistics for relationships between length and weight and between fecundity and size based on data from 128 "summer" and 94 "winter" nehu plus relationship between gonad weight and bodily weight for 67 summer and 44 winter nehu with hydrated ova. Variables are standard length in mm (SL), ovary-free bodily dry weight (S) and total dry ovary weight (G) in g, and fecundity in numbers of ova in the most advanced mode (F). Results of regressions based on natural logarithms are given as power curves (antilog form). The 95% confidence limits are given for either the slopes of linear equations or the exponents of power curves.

X,Y	Summer	(95% CL)	r^2	Winter	(95% CL)	r^2
ln SL, ln S	$S = 8.868 \times 10^{-7}\ SL^{3.25}$	(3.11-3.40)	0.94	$S = 3.696 \times 10^{-7}\ SL^{3.47}$	(3.30-3.65)	0.94
SL, F	$F = -2352 + 63.1\ SL$	(56.3-70.6)	0.59	$F = -1465 + 38.9\ SL$	(33.6-45.0)	0.51
ln SL, ln F	$F = 6.073 \times 10^{-8}\ SL^{5.95}$	(5.24-6.75)	0.49	$F = 1.226 \times 10^{-8}\ SL^{6.24}$	(5.45-7.15)	0.56
S, F	$F = -351 + 3787\ S$	(3,420-4,194)	0.66	$F = -223 + 2444\ S$	(2,119-2,819)	0.53
ln S, ln F	$F = 7094\ S^{1.83}$	(1.63-2.05)	0.56	$F = 4{,}538\ S^{1.80}$	(1.57-2.05)	0.59
ln S, ln G	$G = 0.2339\ S^{1.88}$	(1.65-2.14)	0.72	$G = 0.1192\ S^{1.61}$	(1.32-1.95)	0.60

that the significant differences for winter may have resulted from chance alone.

The regression statistics (Table 1) for data from each season indicate a great deal of variability about the functional relationships between fecundity and length or weight or between the logarithms of these. The correlation coefficients (r) for all regressions were significantly ($P < 0.05$) different from zero, but the coefficients of determination (r^2) indicated that only about half the variance of fecundity or ln fecundity was accounted for by the regression. The exponents from the logarithmic regressions of fecundity on length are considerably higher than those of the weight-length relationships, and the exponents from the logarithmic regressions of fecundity on weight are significantly greater than one. Both indicate that fecundity is not linear with weight and that the appropriate expressions for the functional relationship with size are the power curves for fecundity vs. weight. The exponents of the curves for the two seasons were nearly the same, while the summer-winter ratio of the preexponential factors (antilogs of the regression intercepts), 1.56, was almost identical with the ratio of mean relative fecundities, 1.54.

A small, but significant part of the variability in fecundity within seasons was related to variation in length-weight relationships of the fish. Using predictions of weight and fecundity from Model I (least squares) logarithmic regressions on standard length, I tested for correlations between relative deviations (observed-predicted/predicted) of fecundity and weight. The relative deviations were positively and significantly correlated for both seasons (summer: $r = 0.44$, $P < 0.01$; winter : $r = 0.24$, $P < 0.05$). The coefficients of determination, however, indicate that the variation in relative deviation from predicted weight accounted for small percentages of the variation in deviation from predicted fecundity. Maximum relative deviations in weight were ca. $\pm 20\%$ about the predicted value, while deviations in fecundity were much broader: $\pm 75\%$ in summer and $\pm 60\%$ in winter. Thus there was a tendency for relatively "fat" individuals to have higher fecundity, but this did not account for much of the scatter in the fecundity data.

Nematodes were the only parasites noted frequently, and their presence had a minor and insignificant effect on fecundity. About half of the summer fish and about a third of the winter fish had nematodes. For both seasons, the exponent from the logarithmic regression of fecundity on weight was higher for fish without nematodes than for those with them, but the 95% confidence limits overlapped.

G/S values ranged from under 2% to about 12% in summer fish and to about 7% in winter fish. For females with maturing oocytes, G/S is a function of both the number and size of oocytes. LeCluse (1979) showed for *Sardinops ocellata* that ovum dry weight does not increase once hydration begins, and my own preliminary data indicated that this was also true for nehu. Thus effects of variation in oocyte size could be eliminated by considering only fish with LMD >0.75 mm—the size at which hydration begins. The mean G/S for such fish from winter was 4.8% ($n = 67$: range: 2.4-7.1%) and from summer, 6.3% ($n = 44$; range: 2.1-12.0%). Among fish with LMD >0.75 mm, the exponents from logarithmic regressions of gonad weight on fish weight were significantly greater than one for both seasonal groups (Table 1).

Postovulatory Follicle Deterioration

Although the number of specimens examined for postovulatory follicles (POF) was limited (107 from 13 different samples), the results indicated that POF were a reliable indicator of recent spawning up to about 16 h after spawning. Among the 80 specimens from 9 samples taken 1-5 h after estimated spawning time, follicles were either present and obvious or completely absent. Only seven mature females were available from between midnight and dawn. There were no traces of POF in one specimen; in the others, POF were obvious but showed some signs of degradation similar to that described for northern anchovy, *Engraulis mordax*, by Hunter and Goldberg (1980). Among the 20 specimens from two samples taken 14-16 h after spawning, POF were further degraded but still distinguishable from other structures in half the fish, while the others showed no traces. Judged from descriptions of POF in *E. mordax* by Hunter and Goldberg, 14-16 h in nehu appears roughly equivalent to 24 h in *E. mordax*. Although controlled experiments such as those of Hunter and Goldberg were not conducted, it seems likely that, later in the day, POF cannot be distinguished reliably enough to indicate spawning the previous night. Since POF were either present and very obvious or totally absent in fish collected during the night, all traces of previous spawning are apparently gone after about 24 h.

Spawning

Examination of fish from purse seine samples

taken over deep water after sunset indicated that spawning began and ended during a relatively brief period near the predicted spawning time and that most females in the early night samples were spawners. Ten samples were taken within ±40 min of the predicted spawning time. In four of these, 96-100% of the mature females in each (a total of 85 examined) carried hydrated ova. Most of those with hydrated ova appeared to have not yet started to spawn, i.e., the hydrated ova were not completely separated from the ovarian tissue and smaller oocytes. In the other six samples, 0-75% of the females (total = 114) carried hydrated ova; most of these appeared to be either partially or nearly completely spent. The largest oocytes in those with no hydrated ova were usually <0.65 mm—about the same size as the largest unhydrated oocytes in those with some hydrated ova present. Only 17 of the specimens without hydrated ova were examined histologically; POF were present in 13. Although it is not possible to separate spawners from nonspawners unequivocally on the basis of oocyte size (see below), the small size of the oocytes and the high fraction with POF among those examined indicate that most of the fish without hydrated ova from these samples had just finished spawning.

Later in the night, the frequency of females with hydrated ova decreased, and nonspawning fish appeared to occur more frequently. In 16 samples taken between 40 min and 2 h after predicted spawning time the percentage of females that carried hydrated ova ranged from 0 to 80%. Most values were <25%, and only 24% of total of 332 examined carried hydrated ova. Most of those with hydrated ova appeared at least partially spent; many carried only a few at the posterior end of the oviducts. In 14 of these samples, most of the females without hydrated ova were probably recent spawners. The largest oocytes present were <0.65 mm long, and POF were present in 19 of 20 fish examined histologically. In two other samples, however, several of the females carried larger unhydrated oocytes; POF were present in only 6 of 10 examined from one of these samples. Among the 20 samples taken later in the night (2-4 h after predicted spawning time), only 5 of 254 mature females carried hydrated ova, and oocytes >0.65 mm were present in many of the others. POF were present in only 10 of 20 females examined from two of these samples.

Spawning Frequency and Oocyte Development Rate

Oocyte size-frequency data for 135 fish taken between 0 and 3.25 h after spawning time indicated that spawners carried smaller oocytes than nonspawners. Few of these fish had clearly defined modes of unhydrated oocytes, so LMX (or NMX if hydrated ova were present) was used as a measure of oocyte development. The 20 specimens without POF carried significantly larger oocytes than those with either POF or some hydrated ova present (Table 2). Although there was some overlap in the size ranges of the two groups (Fig. 2), most of the other fish taken during this period but not examined histologically had relatively small oocytes and were probably recent spawners.

Oocyte size-frequency data from fish taken inshore in the morning, 14-16 h after last spawning time,

TABLE 2.—Means and ranges of largest oocyte in the most advanced (first) or next mode of oocytes for nehu taken over spawning areas at night and in shallow areas during the morning and afternoon. For night fish which had some hydrated ova present, the datum used was the largest unhydrated oocyte. Collection times were adjusted to hours since estimated time of the most recent spawning. For night and morning, "S" indicates fish that spawned the night of or the night before collection; for afternoon, "S" indicates fish about to spawn the next night. Similarly, "NS" indicates fish that had not spawned the night of or before collection or were not about to spawn the next night. Probability values between three pairs are based on *t*-tests.

Time	Hours since spawning time	Group	*N*	Mode	Largest oocyte (mm) Mean	(Range)	
Night	0-3.25	S	115	First or next	0.56	(0.46-0.71)	$P < 0.001$
	0-3.25	NS	20	First	0.66	(0.50-0.72)	
Morning	14-16	S	10	First	0.64	(0.60-0.70)	$P < 0.001$
	14-16	NS	10	First	0.72	(0.69-0.75)	
Afternoon	20-24	S	59	Next	0.52	(0.42-0.63)	$P < 0.001$
	20-24	NS	45	First	0.68	(0.60-0.75)	

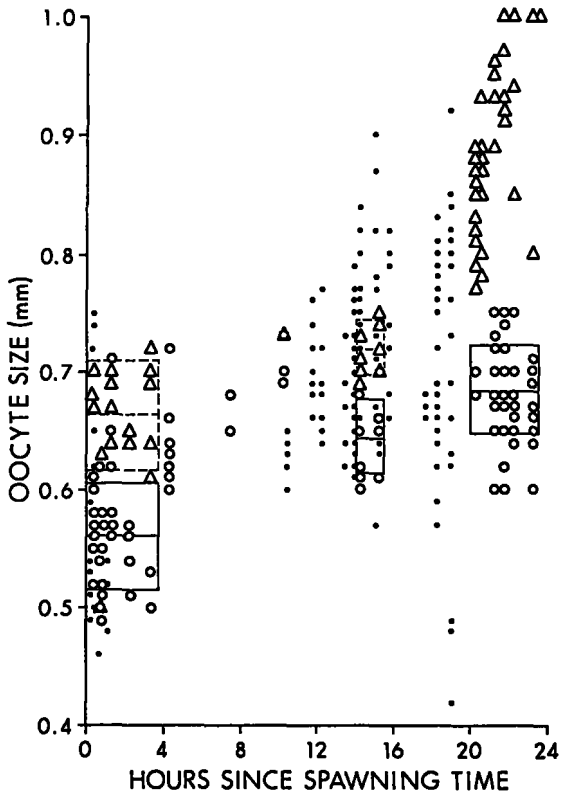

FIGURE 2.—Maximum oocyte size vs. estimated hours since the most recent spawning time for 460 nehu collected at different times of the day from Kaneohe Bay, HI. Large open circles represent fish that either were spawning or had spawned at or near 0 hours (i.e., the night of or the night before collection) or were unlikely to spawn at 24 h (i.e., the next night). For such fish taken 0-3 h after estimated spawning time, the data points indicate the largest unhydrated oocyte. Triangles represent fish that had not spawned at 0 h or were apparently going to spawn at 24 h. Small solid circles represent fish which were not examined for postovulatory follicles and could not be assigned to the above groups on the basis of oocyte size alone. Solid and dashed lines indicate the mean and ±1 standard deviation of maximum oocyte size (horizontal lines) for fish represented by circles and triangles, respectively, for three different time intervals (vertical lines) considered in Table 2.

indicated that oocyte size of both spawners and nonspawners had increased considerably over nighttime values. The LMX of the 10 specimens without POF averaged significantly higher than that of the 10 with POF (Table 2). The mean LMX for spawners had increased to almost the same value observed for nonspawners in the night samples, while that for nonspawners was almost at the size at which hydration begins. Other fish taken at the same times, but not examined histologically, already carried some hydrated ova (Fig. 2).

Data from later in the day indicated that the previous night's nonspawning fish do in fact begin hydrating ova and eventually become the spawners of the next night. Hydrated or hydrating ova occurred in about half the fish taken inshore between 16 and 24 h after last spawning, and most of the rest of the fish had considerably smaller oocytes (Fig. 2). In 67 of 138 specimens taken inshore more than 20 h after the last spawning time (and thus 4 h before the next spawning time), the largest mode was clearly separated from smaller oocytes; LMX was >0.75 mm; and at least some ova were hydrated. The NMX of 59 fish in this group averaged slightly but significantly (t-test, $P > 0.001$) smaller than the largest unhydrated oocytes of spawning fish from offshore early night samples (Table 2, line 5 vs. line 1). This indicates that during hydration and spawning of the current batch, which require no further increase in dry weight or energy content, the oocytes in the next batch were already starting to advance toward the next spawning.

Although some of the remaining fish in the late afternoon (+20 h) samples could conceivably have begun hydrating oocytes and spawned by evening, most appeared to be spawning fish from the night before that were to become the nonspawners of the next night. LMX was >0.70 mm in only 9 of the 71 fish in this group, and about 25% of the 45 examined for oocyte size frequency did not have a clearly separated mode. The mean LMX of these 45 fish did not differ significantly ($P > 0.05$) from that of the nonspawners from the early night samples (Table 2, line 6 vs. line 2). The LMX did, however, average significantly ($P < 0.005$) higher than that of the previous night's spawners in the morning samples (Table 2, line 6 vs. line 3), thus indicating continued oocyte growth between morning and late afternoon.

In summary, the data indicate that most mature female nehu spawn every other day. The largest oocytes present just after spawning increase substantially in size by the next morning and appear to reach the size found in nonspawning night fish by late afternoon. The largest oocytes in nonspawning fish at night are almost at the point where hydration begins by morning, and appear to begin hydrating then or shortly afterwards such that the previous night's nonspawners are nearly ready to spawn by late afternoon.

Alternative cycles are either impossible or difficult to reconcile with the data. If spawning were more frequent, i.e., every day, there would be no nonspawners. This was essentially the case in most night samples taken over the spawning areas, but the day samples averaged about 50% spawners as would result from an every other day cycle. Less

frequent spawning is not consonant with the apparent growth of oocytes in the 24 h after spawning and the near absence of fish with LMX <0.60 mm in the late afternoon samples. Some individuals may, however, spawn more or less frequently than every other day. In the early night samples, some spawners carried larger oocytes than most nonspawners and some of the latter carried smaller oocytes than most of the former (Fig. 2). Thus a few spawners appeared to be capable of ripening the next batch in 24 h rather than 48 h, and the largest oocytes of some nonspawners appeared unlikely to be ready for spawning within 24 h.

DISCUSSION

Results of the present study indicate that the rate of oocyte development in nehu is much faster than in the northern anchovy, *Engraulis mordax*, or the Peruvian anchovy, *E. ringens*, the only other species for which comparable data are available. Hunter and Goldberg (1980) showed that oocytes of *E. mordax* which had spawned within 24 h averaged 0.46 mm long and, during the peak spawning season, grew to the size at which hydration begins in about 7 days. Alheit et al. (1984) indicated that about 6 d are required in *E. ringens*. In nehu, oocytes in largest mode just after spawning averaged 0.52 mm (mean LMD of 54 spawners taken within 3 h after spawning); these appear to advance to hydration stage in <48 h. Hunter and Goldberg's results also indicated that about 7% of the oocytes in the largest mode are not hydrated and spawned; whereas, in nehu it appears that once a batch of oocytes is separated from smaller oocytes, oocytes in that batch are rarely left behind and not spawned with majority of the batch.

Hydration, spawning, and degeneration of POF after spawning are also more rapid in nehu than in the *Engraulis* species. In *E. mordax* hydration begins in the morning about 12 h before spawning begins (Hunter and Macewicz 1980); Alheit et al.'s (1984) data indicated that *E. ringens* is similar. Both studies indicated that the *Engraulis* species spawn over a broad period after sunset with peak spawning just before or near midnight. Nehu ova to be spawned on a given night begin hydrating only a few hours before spawning, and spawning occurs over a rather brief period shortly after sunset. Whereas POF are reliably identifiable up to 24 h after spawning in *E. mordax* and even longer in *E. ringens* (Hunter and Goldberg 1980; Alheit et al. 1984), they appear to degenerate to a similar point in about 16 h in nehu.

My estimates of spawning timing and duration conflict with those of Yamashita (1951) upon which Tester (1955) apparently based his statements that nehu spawn around midnight. As mentioned earlier, studies in progress on appearance of newly spawned eggs confirm the pattern indicated by presence of females with hydrated ova in purse seine samples after sunset. These studies further indicate that Yamashita was probably not sampling deep enough in the water column to collect newly spawned eggs and that his "freshest" eggs were actually one or more hours old.

One of the broader implications of this study is that, when dealing with tropical species, the time scale of sampling must be on the order of hours rather than weeks or days. The latter may be appropriate for investigation of species from higher latitudes, but would miss many events or stages in the reproductive cycle of nehu. Leary et al.'s (1975) conclusion that nehu spawn only once per lifetime was in part based on the rarity of females with hydrated ova in their samples. This was almost certainly due to their not sampling during the short period between late afternoon and shortly after sunset when hydrated ova are found in the current night's spawners. Leary et al. stated that all females with hydrated ova were captured between 2100 and 2300 h, i.e., well after the peak of spawning even in summer.

Both of the above studies of *Engraulis* species indicate some degree of segregation of spawning females at or near spawning time; spawners tended to be overrepresented in such samples. Segregation appears more extreme in nehu; the purse seine samples taken just before and after spawning time were almost all spawners. The greater percentage of nonspawners in some purse seine samples taken later after spawning and the nearly 1:1 ratio of spawners to nonspawners in most day samples indicate that spawning fish remix with others later during the night and that segregation of the next night's spawners does not occur until the mixed schools leave shallow day areas at or near sunset. The distribution of nonspawners early in the night is not known.

The winter-summer differences in nehu fecundity were evident from both the comparison of relative fecundity and the regressions of fecundity on either length or weight. The G/S data for fish with ova >0.75 mm also showed a higher mean and broader range in summer. Other data (Clarke unpubl. data) indicate that spawned nehu eggs are about 20% heavier in winter, but this difference is insufficient to compensate for higher fecundity in summer fish.

The summer-winter ratio of mean relative fecundity was 1.54; roughly corrected for the egg weight difference, the ratio of mean effort per spawning would be 1.28 (1.54/1.20), about the same as the ratio of mean G/S, 1.31. There was no evidence that winter fish compensated for lower effort per spawning with higher frequency.

The causes and adaptive value of the much greater range and, on the average, higher effort by summer fish are not obvious. Similar differences have been reported between different populations of other species. For example, the northern population of *E. mordax* appears to be more fecund than the central population (Table 3). This difference is probably genetic and appears to reflect the shorter spawning season (and lower number of batches) in the northern population (Laroche and Richardson 1980). Since nehu live <6 mo (Struhsaker and Uchiyama 1976), it is difficult to postulate that the differences between summer and winter fish are genetic. It is, however, possible the winter fish may spawn for longer periods and thus to some degree compensate for lower effort per spawning.

The winter-summer differences in nehu reproductive effort per batch may simply be physiological consequences, perhaps with neutral or even negative adaptive value, which result from seasonal differences in the environment. If output in nehu is closely linked to recent feeding success (see below), the output could be lower in winter fish if average daily ration were lower. There is, however, no evidence of major seasonal differences in standing crop of the macrozooplankton upon which adult nehu feed (Hirota and Szyper 1976). Also, nehu feed almost exclusively at night (Clarke unpubl. obs.), and actually have a longer feeding period per diel cycle during the winter. Although the difference between summer maxima and winter minima of temperature in Kaneohe Bay is only about 5°C, it is possible that metabolic processes overall, and consequently both daily ration and reproductive output are slowed enough in winter to account for the observed difference.

Regardless of season, the relative fecundity data combined with minimal estimates of spawner abundance from purse seine catches predicts planktonic egg densities 2 or 3 orders of magnitude higher than those reported by egg surveys of Tester (1955) or Watson and Leis (1974). Assuming all fish in a ca. 300 m^2 area were captured, catches of several purse seine sets indicated 0.3-0.5 g dry weight of spawning females/m^2 and predicted egg densities of 10^2-10^3/m^2. Studies in progress have shown that such egg densities do in fact occur routinely, but that most of the eggs are deeper than 5 m in the water column. Thus the earlier egg surveys, which used surface plankton tows, had missed over 90% of the spawned eggs.

Comparable fecundity data are available for only a few other species of anchovies (Table 3), and most

TABLE 3.—Fecundity-weight relationships for winter and summer nehu, *Encrasicholina purpurea*, and five other species of anchovies. Means and standard deviations of relative fecundity and power curves for fecundity vs. weight were calculated from available fecundity and weight data. Fish weight were ovary-free wet weights except for nehu, whose wet weights were estimated from dry ovary-free weight data, and *Engraulis ringens*, for which the data were given as total fish wet weight. Power curves are the antilog forms of equations based on Model II linear regressions of the natural logarithms; 95% confidence limits are for the exponents. Relative fecundities of the smallest and largest female from each group were calculated from the extremes of weight values and the appropriate power curve.

Species	*N*	Fish weights (g)	Relative fecundity (eggs/g) Mean (+2 SD), smallest-largest		Fecundity vs. weight (95% C.L.)	Reference
Encrasicholina purpurea						
Summer	128	0.4-1.8	566 (±436),	284-1,043	$F = 647\ W^{1.83}$ (1.63-2.05)	This study
Winter	94	0.4-1.3	368 (±266),	195-542	$F = 431\ W^{1.80}$ (1.57-2.05)	This study
Engraulis mordax						
Central	67	9.3-31.9	421 (±295),	261-561	$F = 65.6\ W^{1.62}$ (1.36-1.93)	Hunter and Macewicz 1980
North	21	14.4-31.3	826 (±449),	650-1,094	$F = 108.9\ W^{1.67}$ (1.19-2.34)	Laroche and Richardson 1980
Engraulis ringens	83	11.8-41.5	651 (±404),	493-709	$F = 241\ W^{1.29}$ (1.09-1.53)	Miñano 1968
Cetengraulis mysticetus	86	24.5-69.5	863 (±529),	613-1,233	$F = 71.9\ W^{1.67}$ (1.45-1.93)	Peterson 1961
Stolephorus heterolobus	9	1.6-6.3	469 (±173),	410-514	$F = 379.4\ W^{1.165}$ (0.89-1.53)	Muller 1976
Anchoa naso	12	0.8-5.6	885 (±672),	1,257-618	$F = 1,159\ W^{0.64}$ (0.43-0.94)	Joseph 1963

of these species are much larger than nehu. The reproductive size range of nehu overlaps slightly with only *Stolephorus heterolobus* and *Anchoa naso*. Unfortunately, previous studies of these two species involved very few specimens, and the summary statistics must be regarded as less reliable than those of the other species in Table 3.

Mean relative fecundities for nehu appear to be lower than those of most species; however, the usefulness of this parameter is questionable because the exponents of the power curves relating fecundity and weight are considerably (and significantly) greater than one in most of the species. Thus mean relative fecundity, a commonly used comparator, would be affected by the size range and size composition of the sample of females upon which fecundity and weight are based. When two groups of similar size composition are compared, as in the case of summer and winter nehu, the difference in mean relative fecundity is similar to that indicated by comparison of power curves, but otherwise, such as when comparing different-sized species, mean relative fecundities are likely to give erroneous or at best misleading results. Mean relative fecundity also ignores the differences between small and large individuals of the same species or population.

The exponents of the power curves for nehu are considerably higher than those of any other species. Although the 95% confidence limits for these values do not exclude those for all the other populations, this indicates that the rate of increase in relative reproductive output with increasing size is greatest in nehu. The consequences are illustrated by the relative fecundities calculated for the smallest and largest fish of each population using the power curve for that species (Table 3). Relative fecundities of the largest females are 1.2-2.2 times those of the smallest in the other species but 2.8 and 3.7 times greater in winter and summer nehu, respectively. Both the smallest and largest winter nehu appear to be less fecund per unit weight than the smallest and largest females of all or most of the other species. Small summer nehu also have considerably lower relative fecundity than most of the others, but the value for large summer nehu is among the highest. Ignoring the rather questionable results for *Anchoa naso* (only 12 individuals), the value for the largest *Cetengraulis mysticetus* is the only one substantially greater than that of the largest summer nehu.

Although these comparisons must be regarded as tentative because many between-species differences in power curve exponents are not significant, nehu seem to be distinguished from other anchovies not by differences in relative fecundity but rather by differences in the relation between relative fecundity and size. Speculation about the possible relation of this to differences in environment and other life history parameters, such as nehu's short life span and maturity soon after metamorphosis, is unwarranted without evidence that similar differences exist between large and small species in other taxa. Nevertheless, it seems possible that the pattern of allocation of resources between growth and reproduction over the reproductive life span is yet another life history parameter which could be selected for by prevailing adult mortality rates, predictability of larval survival, etc.

Comparison of fecundities alone does not adequately reflect differences in reproductive effort if there are differences in egg size. For example, nehu eggs average about two-thirds the egg weights calculated for *E. mordax* by Hunter and Leong (1981). Effort per batch would be best measured by relative cost in terms of dry weight, calories, etc., rather than numbers of eggs. Available data permit only crude comparisons of the two species.

The intercept of the regression equation for G/S vs. fecundity of nehu with ova >0.75 mm is about 2.5% for fish from both seasons and nearly the same as the mean G/S (2.4%) of 21 other fish whose largest oocytes were 0.48-0.65 mm and had presumably just spawned. (G/S data were not available for fish used for POF analyses.) Using 2.5% as the mean G/S 2 days before spawning and subtracting this from mean G/S of nehu with ova >0.75 mm, i.e., those about to spawn, gives mean relative weights per batch of 3.8% of bodily dry weight in summer and 2.3% in winter. These estimated relative costs per batch are minimal since they do not include investment in bringing oocytes to the size at 2 days before spawning.

Hunter and Leong (1981) did not give relative cost per spawning of *E. mordax* in terms of dry weight, but data in their table 4 plus an assumption of dry bodily weight equal to 25% of wet weight yield an estimate of about 4.4% of bodily weight per spawning for an average female. Hunter and Leong's data in table 1 indicated that dry weight in *E. mordax* declined about 30% during the main spawning season due to loss of fat; this loss is shown to be equal to the calories required for about 13 spawnings. If this is also true for dry weight then the loss per batch would be about 2.3% of dry bodily weight.

The above estimates of cost per batch in terms of dry weight are very crude and only indicate that nehu, particularly summer nehu, are probably similar to *E. mordax*. Additionally it is clear that nehu, like *E. mordax*, lose half or more of their ovary

weight with each spawning and must depend on bodily reserves and assimilation of food, rather than ovarian reserves, to continue spawning. As mentioned above, Hunter and Leong (1981) showed that about 65% of the caloric cost of spawning is supplied by fat reserves. Even if the same were true for nehu, the additional requirements for continued spawning would have to come from food assimilated and available for reproductive processes over a period of only 2 d rather than 7 d in *E. mordax*. Assuming cost per batch is 4% of dry bodily weight and that 65% of this comes from bodily reserves in both *E. mordax* and summer nehu, the average additional requirements per day would be 0.2% and 0.7%, respectively.

The above suggests that all aspects of reproductive output in nehu—batch fecundity, spawning frequency, and duration of spawning—would be very sensitive to any factors affecting availability of resources for reproduction. Parasite load, which has been shown to affect batch fecundity in cod (Hislop and Shanks 1981), apparently has only an insignificant effect on nehu, but since a batch is formed only 2 or 3 days before spawning and the ova to be spawned on a given evening do not attain maximum size until just a few hours before spawning, even recent events could affect the number or the growth rate of oocytes in a batch. Some of the great variation in fecundity and the indications that some fish spawn more or less often than normal could result from individual differences in recent feeding success, injury or stress from predators or the fishery, or perhaps the extent of inshore-offshore movements over the diel cycle. Unfortunately, none of these putative factors (except for serious injury) would leave any detectable trace on individual fish that might explain why fecundity or spawning frequency was higher or lower than average.

ACKNOWLEDGMENTS

I thank L. R. Johnson and K. C. Landgraf for assistance in different phases of this study. The research was supported by the University of Hawaii Sea Grant Program, PM/M-20 and PM/M-1BB1; by the University of Hawaii Research Council; and by the Hawaii Institute of Marine Biology.

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