

Ovarian Maturation of Japanese Anchovy, *Engraulis japonica* T. and S., from I-Lan Bay, Northeastern Taiwan

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Shuh-Sen Young and Tai-Shen Chiu (1994) Ovarian maturation of Japanese anchovy, *Engraulis japonica* T. and S., from I-Lan Bay, northeastern Taiwan. *Zoological Studies* 33(4): 302-309. The maturation of Japanese anchovy, *Engraulis japonica* Temminck and Schlegel, was studied from specimens collected by local fishermen and targeted for herring-like fish fishery (HELFF) in northeastern Taiwan. Two hundred forty-eight ovaries collected during the period of March 12, 1992 to April 30, 1993, were examined. Oocyte development were categorized in four stages according to cytological appearances. The size of the oocyte was represented by the length of the major axis. During the period, March through May, the migrating schools of Japanese anchovy in I-Lan Bay were 11-12 cm in standard length, and the gonadosomatic index (GSI) of 4.0-6.0 that indicated the reproductive status of this species in the Bay. Both GSI and percent of maturation (PM) are valid indices to measure ovarian maturity. The group maturation remained almost constant "intra-annually" and "inter-annually" during the HELFF fishing season.

Key words: Reproduction, *Engraulis japonica*, HELFF, Taiwan.

The herring-like-fish fishery (HELFF), which utilizes the post larval fish, is one of important coastal fisheries in Taiwan, and the fishery is composed of three major species: Japanese anchovy (*Engraulis japonica* T. and S.), *Encrasicholina heteroloba* (Ruppel) and *Encrasicholina punctifer* Fowler (Young et al. 1992). Two distinct fishing seasons, spring and fall, are readily identified; Japanese anchovy, the major species in I-Lan Bay, is from March until May, the others from September to November. The landing from the I-Lan Bay, one of two major HELFF fishing sites in the coastal waters around Taiwan, contributes approximately one-third of the total Taiwan fishery catch (Anon. 1992). The other site is located in the coastal waters of Fang-Liao in the southwestern Taiwan. As over-fishing of Japanese anchovy imposed a significant impact on fish stock, the mechanism of stock replenishment is of great interest to fishery biologists, and comprehension of the reproductive biology of this species is crucial for resource management.

The reproductive biology of *Stolephorus zolingeri* (= *E. punctifer*, current legal name) and of

Stolephorus heterolobus (= *E. heteroloba*) in the Fang-Liao area were reported elsewhere (Chen 1984 1986). Although the Japanese anchovy is a dominant species in either of the HELFF fishing sites, information of the reproductive biology of this species is still lacking. This work is intended to present results on oocyte development and group maturation of Japanese anchovies collected from the I-Lan Bay. Information obtained is fundamental for estimating batch fecundity and recruitment size of Japanese anchovy.

MATERIALS AND METHODS

Samples

The fishing ground for HELFF is located about 30 m isobath in the I-Lan Bay, (around 25°50' N, 121°55' E; Fig. 1). Specimens for reproductive studies were collected regularly from the catches of local fishermen during the fishing season. A two-strata-sampling technique was employed.

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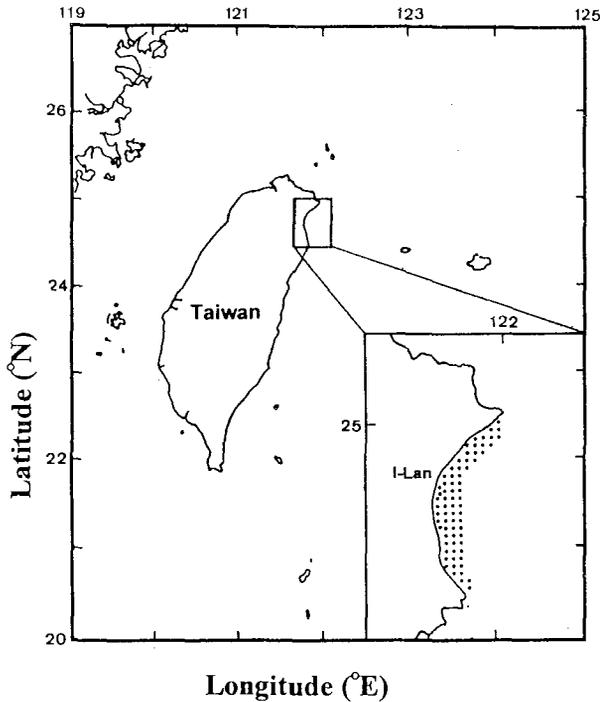


Fig. 1. Sampling area of Japanese anchovy, *Engraulis japonica*, in I-Lan Bay, NE Taiwan during 1992-1993.

Table 1. Summary data on female specimens examined in the study of maturation for Japanese anchovy, *Engraulis japonica*

Year/ Date	Sample size (N)	Fork length (mm) mean \pm SD	Range (mm)
1992			
03/12	24	111.8 \pm 3.7	118.5-101.4
03/19	37	111.2 \pm 6.1	122.4- 99.7
04/01	14	117.7 \pm 6.4	133.5-109.4
04/07	16	113.9 \pm 7.3	128.6-102.1
04/14	13	113.8 \pm 6.4	125.1-103.7
05/03	20	116.2 \pm 7.5	131.8-104.0
Mean		113.5 \pm 6.1	
1993			
02/17	19	121.1 \pm 8.2	146.6-112.8
02/20	10	126.1 \pm 9.4	144.3-110.3
03/04	9	121.6 \pm 5.6	135.2-114.4
03/07	15	119.6 \pm 5.4	131.4-112.9
03/19	12	122.0 \pm 7.0	140.4-113.5
03/26	9	120.1 \pm 5.7	130.6-112.2
03/31	8	124.3 \pm 6.8	135.7-116.1
04/15	10	121.1 \pm 3.7	126.6-116.3
04/17	12	120.9 \pm 6.1	129.4-111.0
04/28	7	117.0 \pm 4.5	122.8-110.6
04/30	13	121.2 \pm 5.5	133.0-114.1
Mean		121.3 \pm 6.3	
Grand mean		117.4 \pm 6.2	

Specimens were first sampled from a fishing boat, and then sub-sampled from the daily catch by local fishermen. Sampling data in the fishing seasons of 1992 and 1993 are summarized in Table 1.

Procedures

The fork length and body weight of the specimens collected was measured to the nearest of 0.1 mm and 0.01 g, respectively. Fish were biopsied and the ovaries were then extracted, weighed, and preserved in 5% formalin. The preserved ovaries were examined and the oocytes from each ovary were separated from the ovarian tissue in glycerin. The ovarian oocyte were counted and measured under a binocular microscope. From each ovary, thirty eggs were randomly selected for double verification; i.e., an image of the egg was fetched with an image analyzer and the corresponding photographs were measured in a more precise manner. The staging of oocyte development is based on the cytological peculiarities described by Yamamoto (1956), Takeshita and Tsukahara (1971), Kuo and Nash (1975), and Wallace and Selman (1981). The oocytes of Japanese anchovy were arbitrarily categorized into four groups; primary oocyte, yolk vesicle, yolk globule, and mature oocyte stages.

The fish maturity was represented by gonadosomatic index (GSI) and oocyte size spectrum. The GSI is defined as: $GSI (\%) = [Ovarian\ Wt. / (Body\ Wt. - Ovarian\ Wt.)] \times 100$, here total ovarian weight and body weight are all in grams. Since the major axis of a developing oocyte was in the range 0.1-0.9 mm before reaching full maturation, the oocyte was scored by a percent of maturation, which is defined as:

$$(\text{length of major axis (mm)} - 0.1) \times 100 / 0.8 (\%)$$

Mean percent of maturation of the oocytes is defined as percent maturation (PM) of ovary. The spectrum of oocyte diameters from each fish were correlated with the magnitude of the GSI in order to evaluate the validity of GSI for maturity measurement.

RESULTS

Oocyte development

Oocyte development were categorized into four stages based on the gross appearance of the ovarian oocytes.

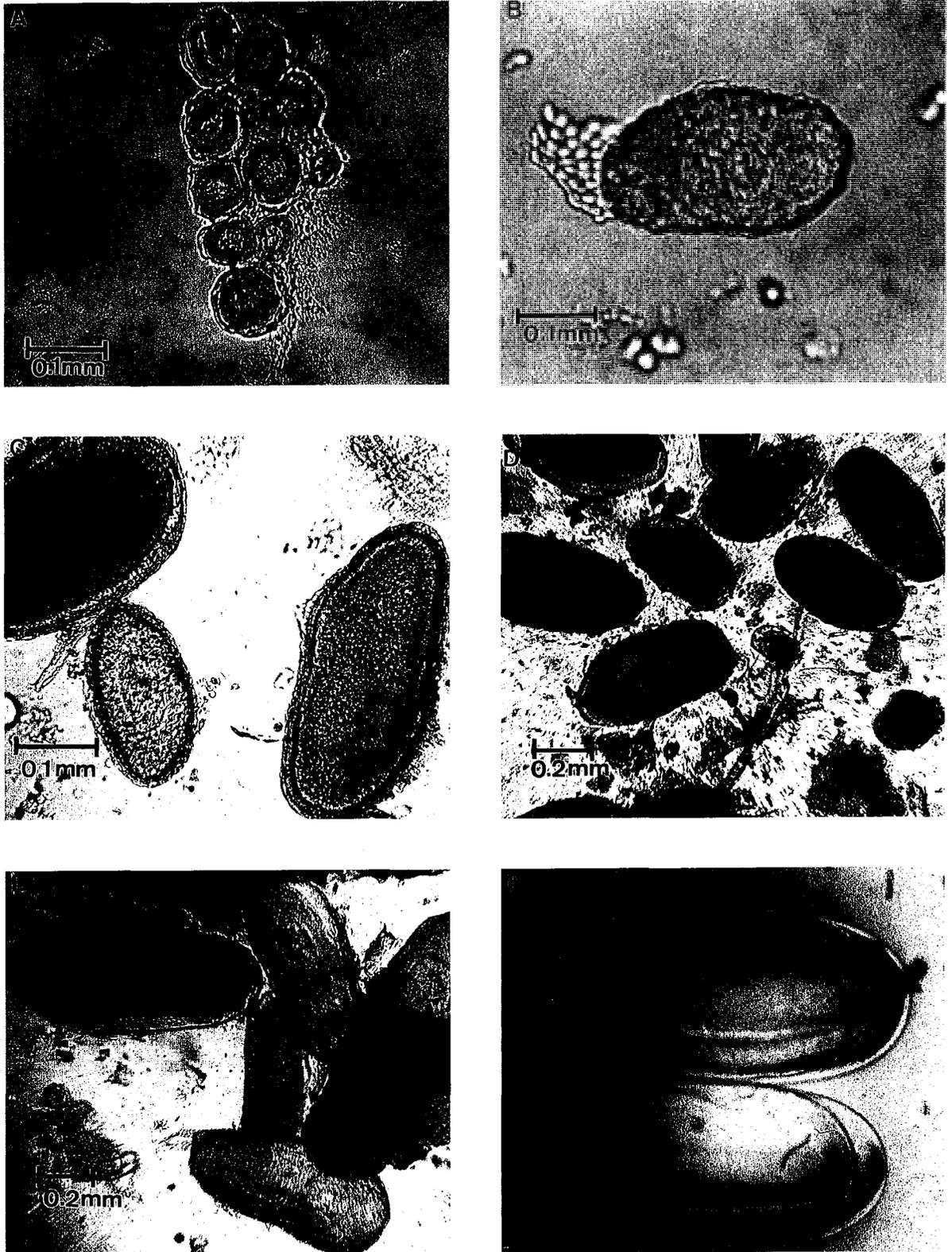


Fig. 2. Stages of oocyte development of *Engraulis japonica*. A: Primary oocyte stage; B: Yolk vesicle stage; C,D,E: Yolk globule stage (primary, secondary, and tertiary yolk globule stages); F: Spawned egg collected by field survey.

Stage I — Primary oocyte stage. The oocyte is small and transparent with a few vacuous nucleolus (Fig. 2A). The size of the primary oocyte is small, about 0.1 mm in diameter.

Primary oocytes are numerous and present in the ovaries throughout the year.

Stage II — Yolk vesicle stage. The oocyte is characterized by granular ooplasm and having a slightly darkened zone surrounding the germinal vesicle. The size of the major axis is less than 0.23 mm (Fig. 2B). This stage is considered as being the onset of oocyte vitellogenesis.

Stage III — Yolk globule stage (primary, secondary and tertiary yolk globule stages) In the beginning of the yolk globule stage, the yolk vesicle spreads to the entire ooplasm. The oocytes are granular in appearance (Fig. 2C). As vitellogenesis advances, the oocyte is notably enlarged isometrically with an extensive granule deposition (Fig. 2D). The major axis of the oocyte is greater than 0.9, close to the ovulated oocyte at the end of this stage (Fig. 2E).

Stage IV — Mature oocyte stage. The oocytes that undergo the final maturation are all grouped into this categories. Theoretically, the oocytes at germinal vesicle migration, germinal vesicle breakdown, ovulation, and forthcoming readily spawning are included. Since the process of oocyte final maturation is very rapid and short, difficulties are often encountered collecting the ovary at this stage.

Some spawned-out eggs of the Japanese anchovy were occasionally collected by plankton tows from the surface water in the I-Lan Bay during this work. The range of major axis of the eggs, elliptical in shape, is 1.2-1.5 mm with a minor axis of 0.6-0.8 mm (Fig. 2F).

Measurement of oocyte size

The shape of the Japanese anchovy oocyte is elliptical from the early stage of oogenesis. This shape is determined from the length of the major and minor axes. Equal increases in both directions is confirmed by scatter plots of the two measurements (Fig. 3). The relationship between major and minor oocyte axes length is represented by $Y = 0.053 + 0.39 X$ ($r = 0.90$, $p < 0.001$), in which X is the length (mm) of the major axis and Y of the minor axis in mm.

The linear relationship between major and minor axes provides a convenient tool for measuring of the size of the oocyte. The measurement of the major axis however is exclusively used to represent the oocyte in this study.

Gonadosomatic index (GSI) and percent of maturation (PM)

The spawning migrating school of Japanese anchovy collected in the coastal water of the I-Lan Bay have a minimum standard length of 10 cm (Table 1). Oocytes of all three stages were present in the ovaries, and it is therefore considered that the Japanese anchovy belongs to the multiple spawning class.

The ovary weight to body weight proportion is expressed as a gonadosomatic index (GSI), which indicates the degree of gonadal development. The mean GSI for each size group is shown in Table 2. The mean GSI ranged between 3.75 and 5.44 and between 3.81 and 5.24 in 1992 and 1993, respectively. The oocytes larger than 0.9 mm with major axis were only found in the ovaries of which the GSI values are in the range of 4.0-6.0. A significant monotonic correlation of GSI with body size was not observed (Table 2).

The spectrum of oocyte diameters and the corresponding GSI are illustrated in Fig. 4. Specimens with a major axis less than 0.2 mm of previtellogenic oocytes are numerous in each ovary, but not shown in the figure. The spectrum of oocytes in the range of 0.1-0.85 mm, 0.1-1.1 mm and 0.1-0.9 mm, for the GSI of less than 4.0, 4.0-6.0 and greater than 6.0, respectively.

Relationships of body size and maturation indices

The GSI is a compound quantity based on body weight and ovarian weight (OW). A linear relation-

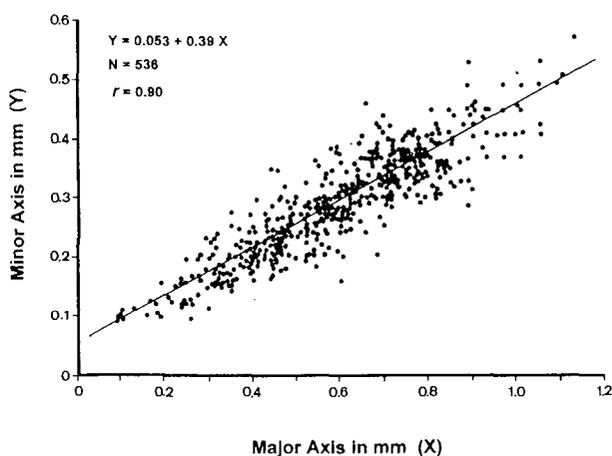


Fig. 3. Relationship of oocyte size measurements, the major and minor axes, of Japanese anchovy (*Engraulis japonica*).

ship is established between GSI and OW: $GSI = 4.981 * OW + 1.636$ ($r = 0.865$; $p < 0.001$). This result exhibits that the GSI can adequately represent the maturation status of the Japanese anchovy.

The percent of ovarian maturation (PM), estimated from averaged stages of the oocytes, is the other valid index for maturity expression. The GSI and PM relationship is significantly linear: $GSI = 0.24 * PM - 9.11$ ($r = 0.67$; $p < 0.001$).

Group maturation

Maturing Japanese anchovies in the I-Lan Bay were collected from March through May in 1992. The GSI is not very different between the pre-spawning and postspawning seasons (Fig. 5). In 1993, a larger magnitude of GSI was found in the

early spawning season, but this value subsided after March 2, 1993 (Fig. 5). Averages of PM and GSI estimated from population of 1992 are 58.05% and 4.61, respectively. The values of 1993 are 57.58% and 4.51 (Table 2). The “inter-annual variation” of group maturity on the spawning population is insignificant.

DISCUSSION

Herring-like fish have been utilized by Taiwanese local fishermen for over forty years. Various fishing gear (trawl, set net, torch net, etc.) have been employed for exploring this resource. Ever since the innovation of the efficient bull trawler in 1970, necessity for resource management of this fishery has gained importance. For the purpose of efficient resource utilization and management, information on spawning biomass in fishing season, maturity status of spawning population and fecundity are essential. Spawning biomass

Table 2. Summary on the maturation of female Japanese anchovy, *Engraulis japonica*, in the I-Lan Bay during the fishing seasons of 1992-1993

Year/Interval of FL (mm)	Sample size (N)	percentage of maturation	Mean GSI (%)
1992			
090-100	2	61.7	5.44
100-110	37	58.5	4.72
110-120	69	57.8	4.55
120-130	19	58.1	4.63
130-140	2	54.3	3.75
Mean		58.1	4.61
1993			
110-120	61	57.4	4.47
120-130	49	56.0	4.15
130-140	11	60.9	5.24
140-150	3	54.6	3.81
Mean		57.1	4.40
Grand mean		57.6	4.51

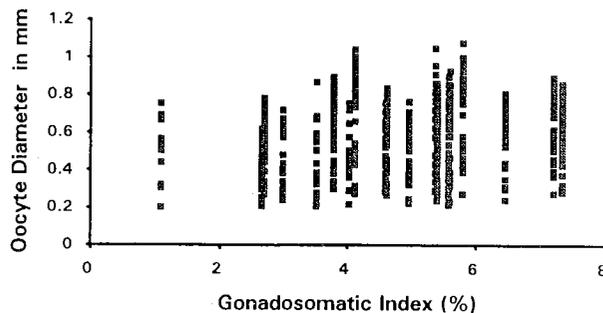


Fig. 4. Scatter plot of measured length of the oocyte's major axis and the gonadosomatic index (GSI) of Japanese anchovy, *Engraulis japonica*.

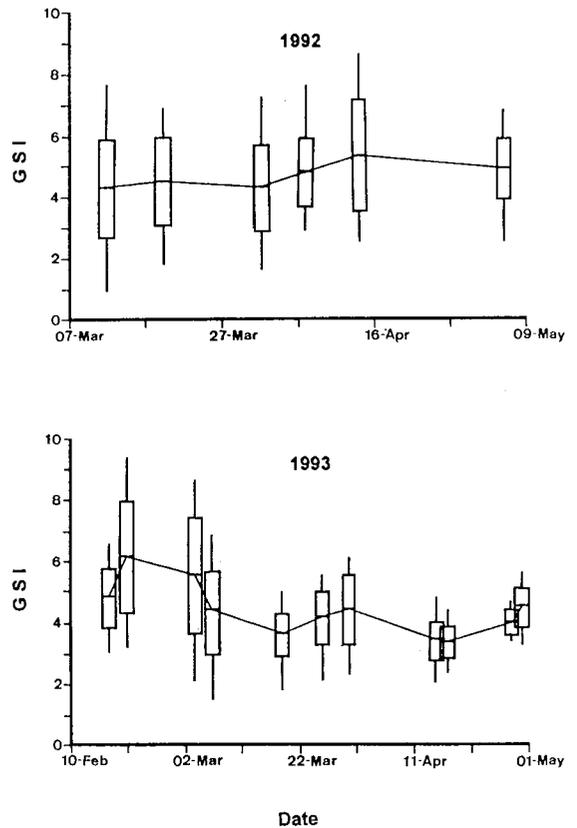


Fig. 5. Group maturity of Japanese anchovy, *Engraulis japonica*, from I-Lan Bay, as shown by time-series gonadosomatic index (GSI) during the fishing seasons of 1992-1993, box with 2 SD, mean and range.

can be estimated from the catch data of fishermen's daily reports and information on maturation are presented in this study.

Several methods may be used to estimate the fecundity from the maturing ovary. For a synchronous spawning species, the batch fecundity (approximation to annual total fecundity) is estimated from the size of standing stock in cooperation with the number of vitellogenic oocytes in the ovaries prior to spawning in the breeding season (Hislop et al. 1978). For heterochronal spawners, annual fecundity is seasonally indeterminated; therefore estimation of batch fecundity based on calculated group maturity becomes an alternative tool of practice (Hunter et al. 1992). The adult Japanese anchovy is a multiple spawner, and the heterochronal nature of this species during spawning season complicates the estimation of batch fecundity; therefore estimation of group maturity is useful to monitor the replenishment process of the Japanese anchovy.

The number of hydrated oocytes is a measure used to estimate the batch fecundity of multiply spawning fishes (Hunter et al. 1985). However, this method has the disadvantage that it must be undertaken when the parent fish are still alive. Counting all yolked oocytes in the ovaries gives neither total fecundity nor batch fecundity for asynchronous multiple spawners. This counting measure could be accepted appropriately only for fish with determined seasonal fecundity. The Japanese anchovy spawners migrate to I-Lan Bay in the period of March to May annually, but reliable estimation on the potential breeders is however, remains to be developed. Until then, the seasonal batch fecundity can probably be an alternative for estimation.

The migration of adult Japanese anchovy to I-Lan Bay is related to reproduction purpose. The body size of migrating groups was uniformly distributed within intra-annual schools, but little variation was found in inter-annual comparison. The group maturity is also constant, about 57-58% (PM) and 4.4-4.6 (GSI).

A relevant index to represent group maturity is necessary for further evaluation of the batch fecundity. Both GSI and PM can fulfill this purpose. The GSI is more convenient than the PM, but precision might be sacrificed due to the different contents of the ovaries. Nonetheless, a gonadosomatic index has long been used to indicate the degree of maturity and spawning of commercial fish; for instance, engraulid (Chen 1984 1986, Clarke 1987, Milton and Blaber 1991, Sanz and

Uriarte 1989); clupeid (Matsuyama et al. 1991); and sparid (Huang et al. 1974). The application of GSI values for synchronous spawners with a short breeding period is more adequate. For fish with longer lasting spawning period, such as the Japanese anchovy, the spawning capacity cannot be precisely determined by the GSI cycle, and GSI application should be carried on more conservatively. In this case, an other index, such as PM derived from the size distribution of oocytes, is considered a potential alternative for maturity determination.

Histological examination of ovarian development provides a convenient tool to define the size at maturation. For the Japanese anchovy in the I-Lan Bay, the largest oocyte is not found even in the specimen with the largest GSI (> 6 , Fig. 4). Nonetheless, fishes with a GSI in the 4-6 range are considered sexually mature with a higher batch fecundity value (Fig. 4).

The multiple spawning of the Japanese anchovy has been reported elsewhere (Usami and Sugiyama 1962, Takeshita and Tsukahara 1971). Although the oocytes at varying developmental stages occurred in the ovaries of Japanese anchovy at the size of 11-12 cm in standard length, it is still risky to render a conclusion on the year-round spawning of Japanese anchovy from this study. However, the Japanese anchovy population in the Bay of Sagami of Japan spawns almost all year round with two peaks of group maturation (Usami and Sugiyama 1962, Takeshita and Tsukahara 1971). For *E. mordax*, some observation of spawning activity was almost year-round, but 73% of all spawning occur from February to April (Fielder et al. 1986). The local Japanese anchovy populations in the Yellow and the East China Seas spawn at a biological minimum size of 60 mm in fork length with a peak of group maturation from April to July (Zhu and Svein 1990). Quantitative evaluation of reproductive potential of this local population has not yet been documented.

Some elaborations on the characteristics of the HELF resource in the coastal waters of Taiwan are available for management purposes. Maturation of engraulid species of *E. punctifer* and *E. heteroloba* of southern Taiwan are well known (Chen 1984 1986). The biological minimum sizes of *E. heteroloba* and *E. punctifer* are 60 and 70 mm, respectively. The developmental process of the ovary is asynchronous for these two species of *Encrasicholina*, of which their character is analogous to the Japanese anchovy (*E. japonica*) as shown in the present work. This cogenetic species of

Encrasicholina is geographically sympatric in the coastal waters of Taiwan and are able to spawn all year round. However, this character of longer lasting spawning is not an attribute for the Japanese anchovy. The conclusion of in this work is in harmony with other work stating that most pelagic clupeid are asynchronous multiple spawners (Blaxter 1982); i.e., more than one group of yolked oocytes exist in the maturing ovaries and the spectrum of oocyte diameters indicates a pattern of continuity.

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臺灣東北宜蘭灣海域日本鰺魚(*Engraulis japonica* T. and S.) 之卵巢成熟週期

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1992年2月至1993年5月自臺灣東北宜蘭灣區(25° 50' N, 121° 55' E)的日本鰺魚(*Engraulis japonica* T. and S.)漁獲中取樣, 研究其性成熟週期, 在這段期間共檢視248隻雌性個體的生殖腺。根據卵母細胞形態差異, 可將其發育分為4個階段。3月至5月洄游至宜蘭灣之日本鰺魚其標準體長達11-12公分, 其性成熟指數(GSI)為4.0-6.0, 由此顯示其在灣內有相當活躍的生殖行為。個體成熟度之估計方法甚多, 對日本鰺魚而言其GSI及成熟百分率(PM)均為相當有效的指標。比較兩年的結果顯示, 在漁獲季節其群成熟狀態相當穩定。

關鍵字：生殖, 日本鰺, 魚鰾魚, 臺灣。

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