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**Aspects of the Reproductive Cycle of *Encrasicholina punctifer* Fowler, 1938  
 from West - Sumatra, Indonesia**

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**Abstract**

*Encrasicholina punctifer* (FOWLER, 1938) is economically by far the most important species in the coastal area of Padang (West-Sumatra, Indonesia). In this study the reproductive behaviour of *E. punctifer* is described for the first time with different methods. The relationship between length and weight of *E. punctifer* in the coastal waters of West-Sumatra can be described as  $W = 6,3 \cdot 10^6 \cdot L^{3,125}$  ( $r^2 = 0,96$ ). The *Encrasicholines* are multiple spawning fishes with asynchronous oocyte development. During one individual spawning season, oocyte development is a continuous process involving all stages of oocytes, clearly represented by the Oocyte-Size-Frequency-Distribution (OSFD). Only very shortly before spawning a new spawning batch can be identified by a gap in the OSFD. There are Post-Ovulatory Follicles (POF) inside the ovary after ovulation. From a Gonosomatic Index (GSI) of 5,5 and more the ovaries hold hydrated oocytes, meaning the anchovies are immediately before spawning. A proportion of the population is spawning at any time. The relative fecundity is 985 ( $\pm 359$ ) eggs per batch and gram body weight. The oocyte development occurs simultaneously in all areas of the ovary. The length at first maturity is at 57,5 mm TL. There is no dependence between condition and gonad weight. The necessary energy for the gonad growth of the anchovies does not come from the body fat but directly from forage.

**Introduction**

In many regions of Indonesia fish is of essential importance for the supply with proteins. At least 200 species are of commercial interest (Soesanto 1985). *Encrasicholina punctifer* Fowler, 1938 is economically by far the most important species in the coastal area of West -Sumatra (Rohdenburg

1995). *E. punctifer* is a small pelagic schooling fish, with a total length of 10 cm (Paula e Silva 1992) and a life span of less than a year (Rohdenburg 1995). This species is commonly found around the coastal waters of the Indian Ocean and the West-Pacific.

Since Delsman (1931) it is known, that *Encrasicholines* spawn virtually continuous. *E. punctifer* is a multiple spawning fish with asynchronous oocyte development, i.e. oocytes in many stages of development occur simultaneously in reproductively active ovaries. After Hunter et al. (1992) multiple spawning fishes are called indeterminate serial spawner, when the annual fecundity is not fixed at the beginning of spawning and the standing stock of the present oocytes in the ovary is continuously replaced by the new developing oocytes during one spawning cycle. In contrary fishes, that have a fixed fecundity prior to the onset of spawning, are called determinate serial spawners. *Engraulis mordax* was determined to be an example for an indeterminate spawner (Hunter and Goldberg 1980, Hunter and Leong 1981). A determinate serial spawner has a standing stock of advanced oocytes - prior to the onset of spawning - is equivalent to the annual fecundity, whereas in an indeterminate serial spawner this is not the case (Hunter et al. 1992). Annual fecundity is much higher, than the present standing stock of advanced oocytes. Determining the type of spawning is of great importance for estimates of the potential annual fecundity and biomass of a species and the ability to recover from human fishery impact. Therefore, information about the reproductive biology of this anchovy is of great interest. In this study the reproductive behaviour of *E. punctifer* is described for the first time with different methods.

## Material and Methods

*Encrasicholina punctifer* were collected twice a week from the artisanal fishery (*Bagan* fishery) at the coast of Padang (Fig. 1). Fish were attracted with light and caught with lift nets during the night. Samples were taken from different boats and pooled for each week from April to July 1994. For the length - weight relationship 2550 individuals were measured. For further studies a total number of 780 individuals from 43 - 95 mm SL were sampled. In the laboratory wet weight, length and gonad weight of the fishes were measured. Subsamples were preserved in 4 % buffered Formalin for histological investigations.

### GSI

The gonosomatic index, GSI, was determined as follows:

$$\text{GSI} = \text{gonad weight} \cdot (\text{gonad free body weight})^{-1} \cdot 100$$

## CONDITION FACTOR

The condition factor CF was determined as follows:

$$CF = \text{gonad free wet weight} \cdot (SL)^{-3,125} \cdot 100$$

Instead of the theoretical coefficient of  $b = 3$ , the actual coefficient of the length-weight regression was used.

## SIZE AT SEXUAL MATURITY

Size at Sexual Maturity was determined with the GSI- method after Grimes (1976). This method determined the 50 % maturity by calculating the mean GSI per 5 mm length-class. In the resulting sequence of mean GSI values, 100 was divided by former GSI, multiplied with the following GSI and then subtracted by 100. The resulting values showed the proportional increase or decrease of mean GSI in comparison to the previous length-class. The highest proportional increase of GSI of all classes corresponded to the mean length, where 50 % of this species reached maturity.

## ANALYSIS OF OVARIES

For counting and measuring the oocytes, the ovary tissue was preserved in Gilson's fluid (Bagenal 1978) for three months. Oocytes from Gilson's fluid fixed tissue were then separated from the ovary wall and measured to the nearest 0,01 mm on their longest axis under a microscope and a digitizer. Ripe ovaries were identified from their external appearance and the extent to which they filled the body cavity. The oocytes of the most advanced group were counted and the relative batch- fecundity (oocytes per g body weight) of each specimen was estimated. The used histological treatments of the ovaries were standard methods with Methyleneblue, suggested by Gerrits (1985). The ovaries of 30 female *E. punctifer* were histologically examined and classified by using Wright's method (1992) for *E. heteroloba*.

## Results

### LENGTH-WEIGHT RELATIONSHIP

The length-weight relationship of *E. punctifer* off the coast of West-Sumatra is

$$\text{weight} = 6,3 \cdot 10^{-6} \cdot \text{length}^{3,125} \quad r = 0,96; n = 2550$$

### GSI AND CONDITION FACTOR

There is no correlation between the GSI and the condition factor of *E. punctifer*. The mean CF-value varies minimal only (0,50 - 0,56) between the lowest GSI-class (0,01 - 2) and the highest

(GSI > 12). The condition of the fishes was not correlated to the developmental stage of the ovaries. This is found for immature females, for mature females and for individuals with hydrated oocytes either (Fig 1).

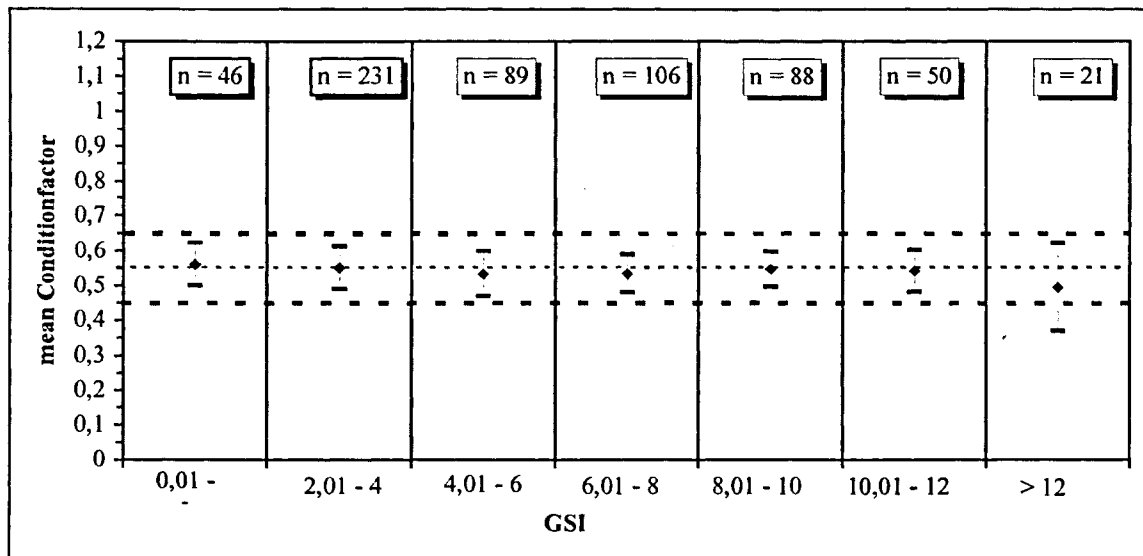


Fig. 1: Correlation of the mean CF ( $\pm$  SD) of the GSI of *E. punctifer* in different GSI- classes

The progression of maturing oocytes was evident from Fig. 2, where the maximum mode of the oocytes in the ovaries was plotted against the gonad index. Oocytes with a maximum length of 0,7 mm are vitellogenic, larger oocytes are hydrated. At a gonad index of 5,5 and above, the size of the oocytes became constant. Furthermore the nature of the scatters of points suggest that there was a rapid increase in the oocyte size at full maturity.

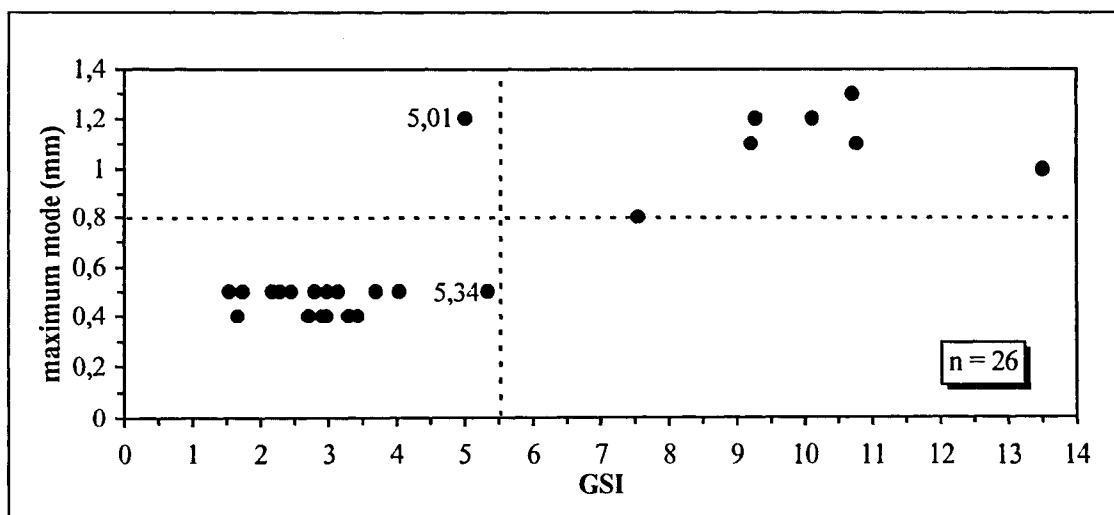


Fig. 2: Relationship between diameter of most advanced mode of oocytes in the ovary and gonad index of *E. punctifer*

### SIZE AT SEXUAL MATURITY

In this study *E. punctifer* reached sexual maturity at a length of 55 - 59 mm SL. This length corresponded with histological investigation. Vitellogenic oocytes were found at a length of 57 mm SL. At a length of 60 mm SL POFs were found inside the ovaries.

### FECUNDITY

Relative batch fecundity of *E. punctifer* was 985 ( $\pm$  359) eggs per g body weight. Table 1 compares relative batch fecundity and length at sexual maturity of *E. punctifer* from various regions of the Indo Pacific.

Tab. 1: Relative Batch fecundity and Length at Sexual Maturity (SL, mm) of *Encrasicholina punctifer* from various regions of the Indo-Pacific

Country	mean rel. Batch Fecundity	$\pm$ SD	Range	N	Sexual Maturity (SL mm)	Author
West - Sumatra	985	359	375-1851	35	55-59	This study
Papua-New-Guinea	875	301	713-1336	9	-	Dalzell (1987)
India	-	-	1236-8971	-	45-49	Luther (1989)
Philippines	-	-	-	-	65	Tiews et al. (1970)

Fig. 3 shows the proportional frequency of the different oocyte-maturity-classes in comparison to the total frequency inside the different quarters of a single ovary. There was no difference ( $p < 0,05$ ) between the different parts of a single ovary. All developmental stages of oocyte development can be found in the entire ovary at the same time. The oocytes do not move to the cranial end of the ovary during maturing. The development happens simultaneously in all parts of the ovary.

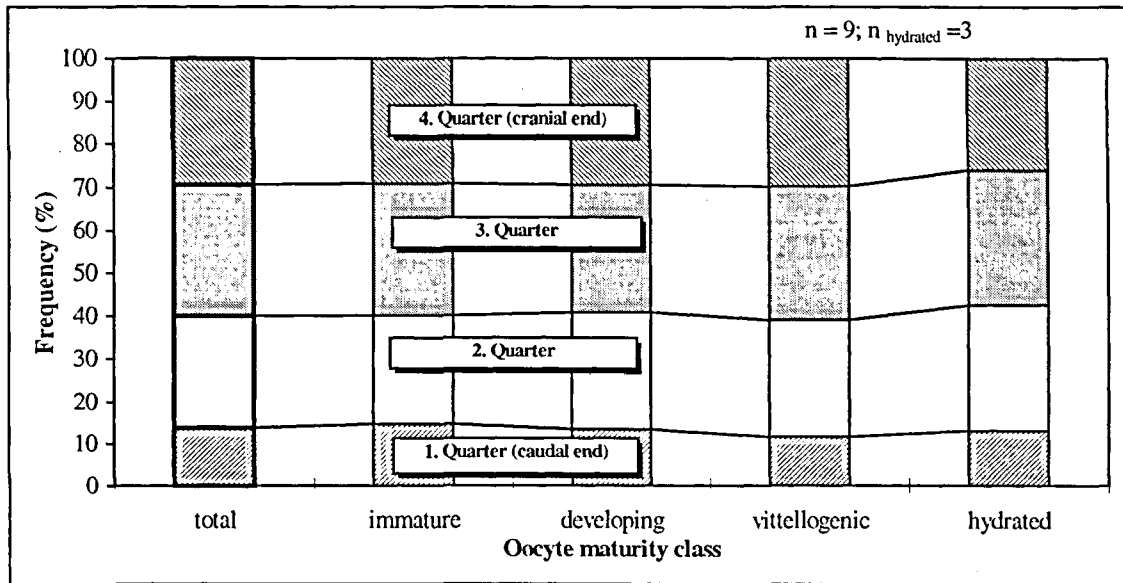
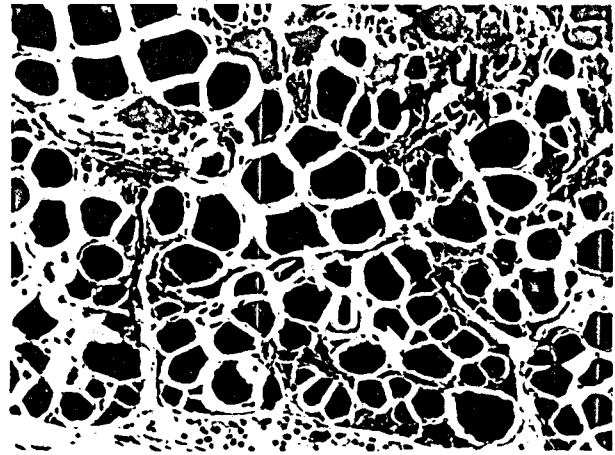
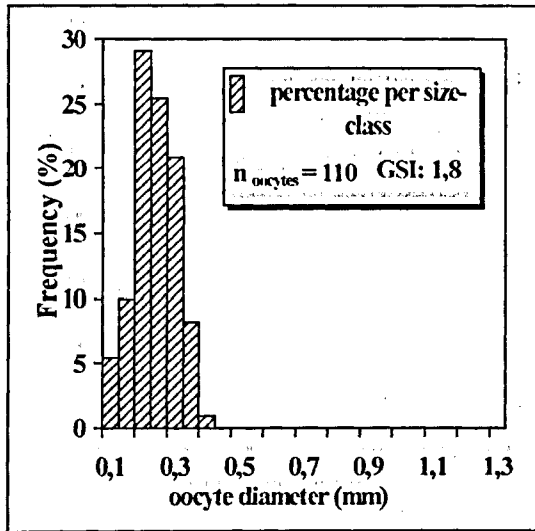


Fig. 3: percentage frequency of the different oocyte-maturity-classes in comparison to the total frequency inside the single quarter of the ovary (1.<sup>st</sup> Quarter: caudal end; 4.<sup>th</sup> Quarter: cranial end)

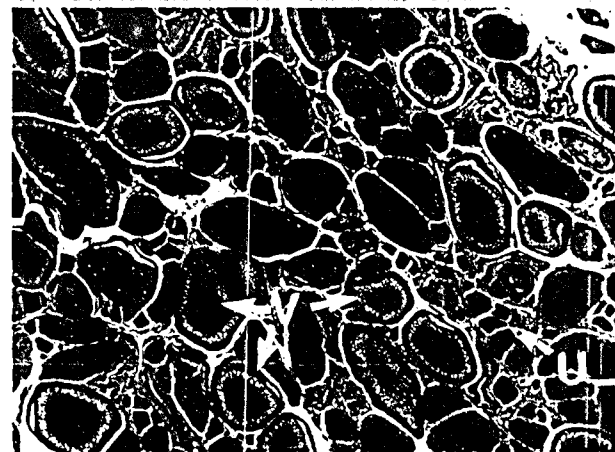
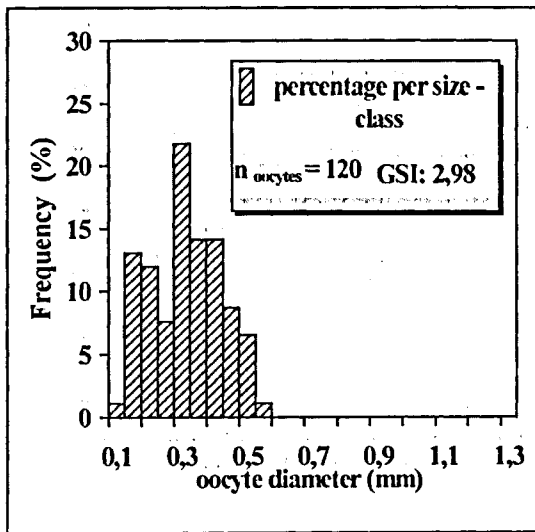
#### OOCYTE-SIZE-FREQUENCY-DISTRIBUTION (OSFD) AND HISTOLOGICAL ANALYSIS

For the main maturation stages, one representative female was taken to show the typical oocyte-size-frequency-distribution (50 $\mu$ m size classes) and the histological cross-section (Fig. 4a -c). In a reproductively active ovary, there is no gap inside the OSFD. This means the oocyte development is continuous (Fig. 4a - b). Only immediately before spawning the most advanced group of oocytes form a new batch, clearly seen in the OSFD (Fig. 4c).

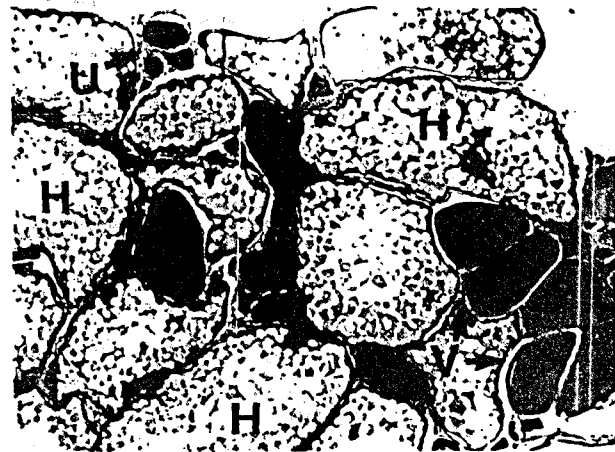
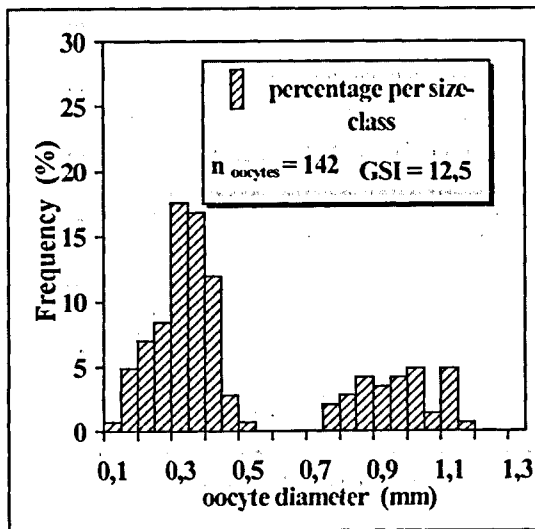
In the cross-section of an immature ovary only unyolked oocytes can be seen (Fig 4a). In a maturing ovary oocytes in different vitellogenic stages can be found additionally (Fig 4b) and an ovary with hydrated oocytes contains oocytes in all different stages of the oocyte development (Fig 4c).



4a: Scale: 0,1 mm



4b: Scale: 0,1 mm



4c: Scale: 0,1 mm

Fig. 4a-c: Oocyte-Size-Frequency-Distribution (OSFD), GSI and histological cross-section of different maturity stages in the ovary of *E. punctifer*. 4a: immature ovary with unyolked oocytes; 4b: maturing ovary with vitellogenic oocytes; 4c: ripe ovary with hydrated oocytes immediately before spawning, the batch is clearly seen. U = unyolked oocytes; V = vitellogenic oocytes; H = hydrated oocytes

## Discussion

The high percentage of reproductive active females, their presence over the whole studytime and the demonstrated OSFD of female indicate that *E. punctifer* is an indeterminate batch spawner.

The variability of the batch fecundity found for *E. punctifer* (Tab. 1) is similar for *E. heteroloba* (Dalzell 1987) and *E. purpurea* (Clarke 1987). The significant difference in the batch fecundity between winter and summer explains Clarke (1987) with different abiotic parameter. In summer water temperature is 5° C higher than in winter. Wright (1989) suggests the availability of forage as the original reason for regional and temporal differences of the batch fecundity of the genus *Engrasicholina*. Tiews et al. (1970) observed a spawning peak of *E. punctifer*, *E. heteroloba* and *E. devisi* in Manila Bay during the southwest monsoon while primary production is very high. Variation for a standard female weight is found for *Sardinops sagax* (George 1993).

The correlation of the mean CF of the GSI of *E. punctifer* (Fig. 1) confirms the suggestion of Wright and Tiews et al. There is no dependence of the condition to the gonad weight of the anchovies. The necessary energy for the gonad growth does not come from the body fat but directly from the forage. This means that the fecundity of *E. punctifer* depends directly on the availability of food. The basic energy requirement is needed for swimming, body function, etc.. Only a surplus of forage can be used for oocyte development. Hunter and Leong (1981) estimate 13 % of the body weight for *Engraulis mordax* to be necessary for a single batch.

The relationship of size at sexual maturity to maximal length with 0,504 fits very well to  $L_m/L_\infty$  ratio Beverton (1963) found for other Engraulidae. One explanation for the different sizes at sexual maturity found by other authors (see Tab. 1) certainly is, that all authors determined the sexual maturity macroscopically. In a multiple spawning fish with asynchronous oocyte development without histological analysis it is very critical to distinguish between developing ovaries from those, which are out of the individual spawning season and mature ovaries from those, which are already partially spent. Important physiological processes like starting of the vitellogenesis and the beginning of the hydration can not be seen macroscopically. In a serial spawner the identification of postovulatory follicle is the only way to find a partial spawned ovary. This is only possible after histological treatment.



## CONCLUSIONS

An extended spawning period with bimodal intensity is a characteristic of an indeterminate serial spawning fish (George 1993). Any time of the year a definitive proportion of the anchovies is able to spawn. Therefore a constant biotic environment is necessary, which is given in the tropical coastal waters off West-Sumatra. Serial spawners produce more eggs than a comparable total spawner. Small fishes like anchovies do not have the space, that would be necessary to hold the total amount of oocytes produced per year at the same time. The hydration of the oocytes has to be divided into portions (Lozan 1985).

Fig. 1 indicates that the energy for the fecundity comes directly from outside. That means a change in prey density has a direct influence on the fecundity of *E. punctifer*. To quantify the effect of variation in prey density on spawning biomass and indirect to recruitment further investigations are of great interest. Additionally the knowledge of the spawning ground is important. Tzeng and Wang (1992) found spawning areas for anchovies in mangrove estuaries. Both reasons are probably at least as important as the direct fishery impact on the standing stock. This is also of economic interest. Particularly because of a lack of alternatives more and more people have to earn their livings with artisanal fishery.

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