

## Reproduction of the oyster *Crassostrea gasar* (Mollusc, Bivalve) in Southern Casamance (Senegal)

*Reproduction de l'huître Crassostrea gasar (mollusque, bivalve)  
en Casamance du sud (Sénégal)*

Hamet Diaw Diadhiou\*, Marcel Le Pennec\*\*

\*Institut sénégalais de la recherche agronomique, zone maritime, CRODT, BP 2241 Dakar, Sénégal

\*\*Biologie marine, UMR CNRS 6539, Institut universitaire européen de la mer,  
Place Nicolas Copernic, 29280 Plouzané, France

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### ABSTRACT

Diadhiou H.D., M. Le Pennec - Reproduction of the oyster *Crassostrea gasar* (Mollusc, Bivalve) in Southern Casamance (Senegal). Mar. Life, **10** (1-2) : 19-25.

Studies were undertaken to better understand some of the more fundamental biological aspects of the mangrove oyster *Crassostrea gasar* (Dautzenberg, 1891) in Southern Casamance (Senegal) with a view to its culture. Results obtained on reproduction from April 1992 to March 1994 are described. The reproductive activity was assessed by determination of a Condition Index, histology and image analysis of histological slides. Despite the fact that the level of mature individuals was high throughout the sampling period, the percentage of mature oocytes was low, varying from 38% (September 1994) to 15% (March 1994). Atretic oocytes were always numerous in the follicles and during the spawning season of October 1992 they comprised 60% of the total oocytes. There was no real sexual rest period as, for example, 25% of the oocytes are seen adhering the acini wall after the October 1992 spawn. The follicle occupancy levels by oocytes, in the gonad, never reach 100% and a maximum of 45% was observed in October 1992. The maximum of spawning individuals, 50%, was found in January 1993. The main spawning periods occur at the end of the rainy season and during the transition rainy-dry colder seasons. This study was undertaken to determine the best period to collect larvae of this species in the wild.

### RÉSUMÉ

Diadhiou H.D., M. Le Pennec - [Reproduction de l'huître *Crassostrea gasar* (mollusque, bivalve) en Casamance du sud (Sénégal)]. Mar. Life, **10** (1-2) : 19-25.

En raison de l'intérêt manifesté pour l'huître de mangrove *Crassostrea gasar* (Dautzenberg, 1891), espèce-cible potentielle dans le domaine de l'aquaculture des mollusques bivalves en Casamance du sud (Sénégal), le besoin de mieux connaître certains aspects de sa biologie s'est imposé. C'est dans ce but qu'une étude a été réalisée d'avril 1992 à mars 1994 afin de connaître son cycle sexuel et déterminer les périodes les plus favorables pour la collecte des larves en voie de fixation. Trois méthodes ont été utilisées: l'indice de condition, l'histologie et l'analyse d'images. Bien que le taux d'individus matures soit resté élevé durant la période d'étude, le pourcentage d'ovocytes matures était faible, variant entre 15% (mars 1994) et 38% (septembre 1994). Les ovocytes atrétiques sont toujours très nombreux dans les acini, pouvant atteindre 60% (octobre 1992) du total des ovocytes. Le taux d'occupation de la gonade par les acini est au maximum de 45% (octobre 1992). La ponte la plus forte observée, a intéressé 50% des individus (janvier 1993). C'est durant la transition entre la saison des pluies, chaude, et la saison sèche, plus froide, que les principales émissions gamétiques se produisent.



## INTRODUCTION

*Crassostrea gasar* (Dautzenberg, 1891) or *Ostrea tulipa* (Lamarck, 1891), commonly known as the mangrove oyster, is widespread in the tropical western regions of the African continent, from Senegal to Angola (Nickles, 1955; von Cosel, 1995). The intertidal zones of estuaries are the preferred habitat of this oyster, although it can also be found in coastal areas at depths of up to 10 m (Sandison, Hill, 1966; Marozova *et al.*, 1991). The mangrove oyster is an euryhaline species whose salinity tolerance range in Southern Casamance is from 6 to 60 (Gilles, Le Pennec, 1992).

Due to its pleasant taste, *C. gasar* is especially appreciated by the coastal populations (Afinowi, 1975; Cormier, 1984), who collect it on mangrove tree roots. According to Cormier-Salem (1986), 10,000 t.year<sup>-1</sup> are collected in this way in Casamance. In several African countries, such as Sierra Leone (Kamara, 1982), Nigeria (Ajana, 1980a,b) and Senegal (Dioh, 1976; Blanc, 1962), attempts have been made to farm *C. gasar*. These efforts, however, have met with little success. For example, the aquaculture of *C. gasar* represents about 5% of the national production in Senegal (Anonymous, 1992). For several reasons, including a poor understanding of the organism's biology, the farming of *C. gasar* is at this time almost non-existent in Africa despite the potential of this industry. Consequently, studies are underway in Southern Casamance (Senegal) to remedy this situation and to better understand some of the more fundamental biological aspects of this species. These studies include the description of the reproduction, the larval phase, the recruitment on artificial collectors and juvenile growth of the mangrove oyster. In this paper, we describe the reproduction of *C. gasar*.

## MATERIAL AND METHODS

Specimens of *C. gasar* were obtained from mangroves on the island of Karabane situated in the mouth of the Casamance River (Senegal). Monthly sampling of 15 to 25 individuals 60 mm in length was carried out from April 1992 to March 1994. Temperature and salinity measurements were taken using a mercury thermometer and a refractometer, respectively, every three days in the morning between 08:00 and 09:00 hrs, and in the afternoon between 14:00 and 15:00 hrs. These time periods correspond to the maximum and minimum of temperature during the day (Dacosta, 1989). The reproductive condition of *C. gasar* was assessed in 3 ways:

- By determination of the Condition Index (C.I.), defined by Walne and Mann (1975):

$$C.I. = \frac{\text{dry flesh weight} \times 100}{\text{dry shell weight}}$$

The drying of the flesh and shells was performed at 60°C for 72h.

- By histology: gonadal tissues were fixed in aqueous Bouin's solution for 48h. After dehydration in successive alcohol baths (70-100%), the samples were set in liquid paraffin. Sections 5 to 7 µm in thickness were then made with a microtome and coloured with Masson's trichromatic staining method (Gabe, 1968). The different stages of gonadal development can be classified according to the scale proposed by Tranter (1958) for the Australian pearl oyster *Pinctada albina* and grouped as follows for female individuals:
  - A: gametogenesis: phase Fd1 (oogonia and previtellogenic oocytes) + phase Fd2 (oogonia and vitellogenic oocytes) + phase Fd3 (adherent or attached oocytes in the same proportion);
  - B: maturity: phase Fd4 (attached and free oocytes) + phase Fd5 (free oocytes, acini almost full);
  - C: spawning and regression of gonadal tissues: phase Fr1 (partial emptying of the acini) + phase Fr2 (acini almost empty) + phase Fr3 (few residual oocytes).
- By image analysis: in this method, five gonad samples were taken from five different individuals during the characteristic periods of the reproductive cycle, namely gametogenesis, spawning, and rest periods. These slides were observed with a Leitz microscope. Images were numerized with an image analyser (Silicon Graphics station PI 25, Visilog software-Noesys, France). The percentage of oocytes that were either adherent, mature or atretic was determined manually and a measurement of gonad occupation levels by these three categories of oocytes was performed using a binary segmentation.

## RESULTS

### Abiotic factors

Changes in fluctuations in salinity are shown in figure 1a. From December 1992 to August 1993, salinity was at a maximum and varied from 42 to 50. From May to July 1992, from November to December 1992 and in September 1993, salinity fluctuated by 1 to 2 ‰ from 40. From July to October 1992 and from October to March 1994 the values were low and ranged from 35 to 30 (figure 1a). The fluctuations in salinity are not identical from one year to the next.

Two periods of temperature fluctuations were discernible (figure 1b). The first is characterised by high temperatures and occurred from May to September 1992 and from May to September 1993 at which times the temperatures fluctuated around 30°C. The second, characterised by colder temperatures, can be seen to occur in December 1992 and in January 1993 and 1994 at which time the temperatures varied between 25°C and 20°C (figure 1b). The fluctuations in temperature appear to be annual.

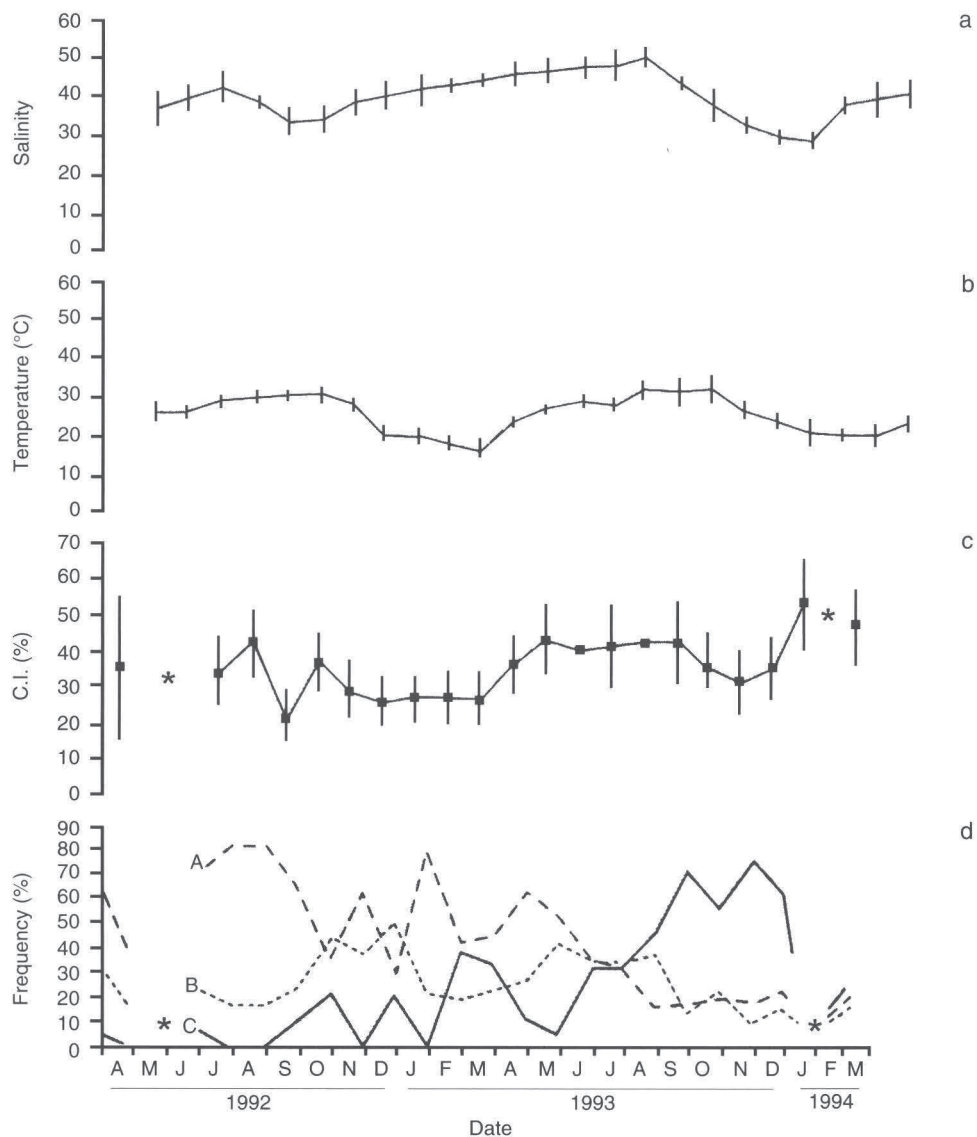


Figure 1 - Salinity (a) and temperature (b) variations in the sampling site of Karabane (Casamance River). Reproductive cycle obtained from the studies of a Condition Index (c) and histology (d) of the gonad of *Crassostrea gasar*. A, B, C: gametogenesis, maturity and spawning. \*: no data. / Variations de salinité (a) et de température (b) au site de prélèvement de Karabane (fleuve Casamance). Cycle reproductif réalisé d'après l'indice de condition (c) et l'étude histologique de la gonade de *Crassostrea gasar*. A, B et C : gamétogenèse, maturité et ponte. \*: absence de donnée.

### Condition Index

Assuming that variations in the C.I. are the result of an accumulation or a loss of organic matter associated with the reproductive cycle (Lucas, Beninger, 1985), four maturation periods and three spawning periods can be identified (figure 1c).

The rapid maturation of the gonad observed from July to August 1992 was followed by a spawn-

ing of gametes after which the C.I. fell to the lowest value observed throughout the two year study period. The maturation after this period was also very rapid and led to an additional spawning period between October and November 1992. From this period to March 1993, the C.I. remained constant and was very low. From March to May 1993, gametogenesis caused a substantial increase in the index



values which then remained at this high level up until September at which time another spawning event occurred. In December 1993, gametogenesis resumed and, by January 1994, the index increased to reach its highest value throughout the two year study period.

### Histology

The monthly percentages of individuals in A, B, and C allow the identification of five spawning periods of varying magnitudes from April 1992 to March 1994 (figure 1d). Maturity was at a maximum in November 1992 (45% of individuals) and in January 1993 (50%). Lower maturity occurred in June 1993 (near 40%) and September 1993 (35%) and a final maturity of low intensity (20%) was observed in November 1993.

The percentage of individuals in phase A was high, between 80 and 20%, throughout the sampling period with particularly high percentages from August to September 1992 and in February 1993 when values reached 80%. Concerning the C phase, 20% of individuals are in this phase in November and January 1993, 30% in March 1993 and 70 and 80% in October and December 1993, respectively (figure 1d).

The occupancy level of the gonad by acini and the quality of oocytes measured by image analysis, during the characteristic periods of the reproductive cycle given by the C.I. results, are presented in table I. The observed periods are: August 1992, maturity; September 1992, spawning; October 1992, resumption of maturation; November 1992, spawning; March 1993, weak values of the C.I.; March 1994, atretic oocytes.

The lowest occupancy level of the gonad was observed in March and April 1993 (about 22% and 28%) at which time the atretic oocytes represented approximately 55% and 48% of the sexual cells present. Occupation levels are at their highest level in

November 1992 being in the order of 69% although the atretic oocytes are dominant at this time representing 55% of the observed oocytes. A high occupancy level of 56% was also seen in July 1993 with the adhering (38%) and atretic oocytes (37%) being the most dominant. The percentage levels of mature oocytes are at their lowest (less than 20%) in September 1992, November 1992, March, April and May 1993 and March 1994 (table I).

### DISCUSSION

In tropical areas, the recognition of the great potential of the farming of several oyster species has led, over the last two decades, to an increased understanding of their reproductive cycles. This is true for *Crassostrea rhizophorae* whose distribution extends along the western coast of South America from Mexico to Venezuela (Velez, 1977), for *Crassostrea madrasensis* occurring along the Indian coast (Stephen, 1980) and for *Saccostrea cucullata tuberculata* and *Crassostrea echinata* present on many islands of the Pacific and in Queensland (Australia) (Braley, 1984).

The results that have been obtained reveal that, for a given Ostreidae species, there occur wide variations in the sexual cycle and in particular the reproductive periods and the intensity of gametogenesis which are a function of the organism's geographic location. These results are in agreement with the generally conceived notion that the variations through time in the reproductive cycle of Bivalves are directly linked to the environmental conditions, namely temperature, salinity, light intensity and food, as well as to a certain number of endogenous factors such as the nervous system and biogenic amines (Sastry 1979; Barber, Blake, 1991).

For certain tropical Ostreidae species, temperature plays an important role. This is true for *C. rhizophorae*. According to Velez (1977), gametogenesis

Table I - Percentage of adherent, mature and atretic oocytes, with standard deviation, and acini occupancy level in the gonad of *Crassostrea gasar*. / Pourcentage d'ovocytes adhérents, mûrs et atrétiques, avec écart-type et taux d'occupation de la gonade de *Crassostrea gasar* par les acini.

		Adherent oocytes		Mature oocytes		Atretic oocytes		Occupancy level of the gonad	
		%	±	%	±	%	±	%	±
1992	August	19.65	10.27	21.36	10.11	59	11.33	38.27	11.33
	September	32.25	14.77	15.11	16.69	52.64	8	32.73	8
	October	17.25	5.70	22.73	11.93	60.02	5.91	44.38	5.91
	November	24.48	19.38	19.34	15.80	56.19	12.12	69.77	12.12
1993	March	26.07	4.40	18.85	4.86	55.08	13.38	22.53	13.38
	April	33.53	6.71	18.20	6.30	48.28	6.27	28.63	6.27
	May	32.40	15.98	18.08	5.86	49.52	8.70	42.01	8.70
	July	38.06	6.08	24.48	8.66	37.46	20.84	55.10	20.84
	August	25.45	4.66	28.28	6.31	46.27	28.82	49.48	28.82
	September	25.74	8.99	37.59	5.06	36.67	14.88	39.92	14.88
1994	March	30.86	6.41	14.40	9.71	54.75	20.59	26.03	20.59



in this last species occurs all year round although high temperatures are responsible for the acceleration of oocyte maturation and spawning. Conversely, lower temperatures cause a regression of the gonad and bring about a reproductive rest period. Therefore, for species which follow the same reproductive pattern as *C. rhizophorae*, gamete release can be observed to occur throughout the year in an asynchronous manner, in response to the fluctuations of certain environmental factors including temperature.

For other Ostreidae, however, salinity seems to be the most important abiotic factor. This is particularly true in regions subjected to monsoons (Rao, 1956; Giese, Pearse, 1974; Braley, 1984) and for species colonising estuaries (Stephen, 1980; Joseph, Madhyastha, 1982). In some species, maximum salinity levels can trigger spawning, such as in *C. madrasensis* (Stephen, 1980) and *C. echinata* (Braley, 1984). As a general rule, however, it is the minimum salinity levels that favour the spawning (Rao, 1956; Giese, Pearse, 1974; Stephen, 1980) as is seen in *Crassostrea gryphoides* (Durve, 1965) and *C. gasar*. Salinity can act directly or, as observed in *Crassostrea virginica* by Bulker (1949) cited by Stephen (1980), indirectly through salinity dependent fluctuation of the phytoplankton population.

*C. gasar* possesses an extensive range of distribution which extends from 15°N (Senegal) to 15°S (Angola). The data available on the reproduction of this species, however, remains fragmentary and concerns mainly the countries situated north of the equator: Nigeria, Sierra Leone, Guinea, and Senegal. In Nigeria, the gonad C.I. is high in July and the oysters are ready to spawn while in December the C.I. is at its lowest level. These results suggest that the reproductive period for these oysters is between September and November (Afinowi, 1975; Ajana, 1980a,b).

In Sierra Leone, spawning can be observed all year long although maturation is particularly important between April and May. The main spawning events therefore occur during the rainy season (starting in June) which allows spat to be collected from September to November (Wellesley-Cole, Kamara, 1978; Kamara, 1982).

A three year study on *C. gasar* in Guinea (Valovaya, Kaba, 1990) showed that there occurs a main spawning period during the rainy season although four such spawning periods can occasionally be observed, two during the dry season and two during the rainy season.

In the Senegal region of Joal (100 km south of Dakar), Blanc (1970) found that there were four periods of spat settlement. The first was sparse and occurs at the beginning of August, the second, of greater magnitude, was seen in mid-August. A period of variable spat settlement was then observed in September, the magnitude of which depends on the amount of rainfall during this month and a last settlement period was seen to occur at the end of October if the rains continue to fall. For Blanc

(1970), the main reproductive period occurs during the rainy season, from July to August. Conversely, Leung-Tack and Pages (1987) described the main period of spat settlement in the same geographical area as occurring later in the year between October and the end of November.

In Casamance, the only available data concerning the period of *C. gasar* reproduction are those provided by Gilles (1991) who used larval settlement to situate the oyster's reproductive period between March and October so long as the salinity is not above 39. The data generated in the present study using the condition index method enabled us to detect the occurrence, during the two year study period, of three main spawning periods in August-September 1992, in October-November 1992, and in September-October 1993. By histological examination, five periods can be identified: November 1992, January, June, September and November 1993. There is agreement for two of these periods which differ by only a few days; October-November 1992 and September-October 1993.

During these periods the salinity levels are low, between 30 and 35, these values being due to the heavy rains that fall in June, July and August in Casamance (Le Reste *et al.*, 1986). The water temperatures at this time are between 25 and 28°C.

It would therefore appear that the main spawning periods in this southern Senegal region occur during the periods of high Casamance River flooding at the end of the rainy season (salinity approximately 35, temperature between 30 and 35°C) in September, and during the transition period between the rainy season and the dry colder season (temperature less 25°C) in October-November.

The results generated by image analysis of the characteristic periods of the sexual cycle, namely maturation, spawning and the post spawning period, help to describe more clearly the sexual cycle of this species:

- mature oocytes are still present within the gonad and their percentages vary between approximately 14% in March 1994, at which time the Condition Index is at its highest value (more than 50), and 37% in September 1993 just before their emission;
- the atretic oocytes are always abundant as a minimum of 36% and a maximum of 60% are observed. Also, these atretic oocytes are numerous during the main spawning periods of October-November 1992 and September-October 1993. Indeed, they represent 60% of all oocytes in October 1992, 56% in November 1992 and 36% in September 1993;
- despite the fact that the C.I. remains constant and low from the September-October spawn to March 1993 there is no true sexual rest period as about 24% of the oocytes are seen adhering to the acini wall;
- the acini occupancy levels in the gonad never reach 100% as levels of 44 and 39% are observed prior to the October 1992 and September 1993 spawns, respectively;



- spawning does not occur in all mature individuals. Indeed, only 10% (November 1993) to 50% (January 1993) of mature individuals examined were seen to spawn.

The data presented in the present study on *Crassostrea gasar* in Casamance help to better understand the reproductive strategy of this species. Throughout the year, it is possible to find mature individuals capable of producing gametes after triggering by environmental factors. It is the rainy season, however, which causes a drop in salinity levels and triggers the main spawning events. In addition, the flooding of the Casamance River at this time brings about an increase in available mineral resources which causes the phytoplankton populations to reach their maximum density in September-October (Diouf, 1987). This increased availability of food may be beneficial to the oyster adults, some of which experience a resumption in gonad maturation, to the larvae and to the young spat.

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