# PHYLOGEOGRAPHY OF MANGROVE OYSTERS FROM THE SOUTHERN ATLANTIC OCEAN: CRASSOSTREA GASAR AND CRASSOSTREA RHIZOPHORAE.

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# Introduction

Mangrove oysters encompass several species and their taxomony is unclear. Among then, *Crassostrea rhizophorae* has been described along the Central and South American Atlantic coasts and *Crassostrea gasar* along African Atlantic coasts. Most taxonomic studies have so far been based on morphological criteria which are particularly susceptible to plasticity and new insight can be provided by molecular markers. The present work aims to provide information on the phylogeography of these two taxa.

## **Materials and Methods**

Nine populations of mangrove oysters were sampled in Senegal, Martinique, French Guyana and Brazil, and their polymorphism for the 16S mitochondrial fragment was studied using different methods (PCR-RFLP, SSCP, sequencing). The karyotypes of some specimens were also examined.

# Results

The three methods gave congruent results and allowed the samples to be separated into two types: an African type, corresponding to *C. gasar*, and an American type, corresponding to *C. rhizophorae*. But the *C. gasar* type was not only observed in Africa (Senegal) but also in South America (French Guyana and Brazil), whereas the *C. rhizophorae* type occurred only on the South American coasts (Brazil, Martinique). Both types were therefore observed in the Brazilian sample. The karyological studies were fully congruent with these results. This is the first report of the presence of *C. gasar* in South America as well as the first molecular genetics study on this species. A phylogenetic tree was built with the information from the 16S fragment sequences of different *Crassostrea* species and showed *C. gasar* in an intermediate position between the *C. gigas - C. angulata* and *C. virginica - C. rhizophorae* groups. These results raise questions on the possible endemic status of *C. gasar* in South America and the sympatric status of both species in Brazil.

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#### INTRODUCTION

Mangrove oysters are encompassing several species growing on different coasts. Crassostrea rhizophorae (Figure 1a) has been described on the South American Atlantic coast and Crassostrea gasar (Figure 1b) on African Atlantic coasts. Taxonomic study of oysters has long been based on morphological criteria particularly susceptible to plasticity. Thanks to molecular biology new insights can be given to taxonomic and phylogenetic studies. The present one aims to bring information on the phylogeography of these two species in South Atlantic.

#### MATERIAL AND METHODS

Nine populations of Mangrove oysters have been sampled (Figure 2) in Senegal (Africa), French Guyana, Martinique Island and Brazil (South America) and their polymorphism on the 16S mitochondrial fragment was studied with different methods (PCR-RFLP, SSCP, sequencing). The karyotype of some oysters was also examined



### **RESULTS**

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All the three methods were congruent and allowed the populations to be separated in two genetic types, corresponding to the two species. But the *C. gasar* type was both found in Africa and South America, whereas the *C. rhizophorae* type occurred only on the South American coasts. Moreover both species were observed on a same location in Brazil

Figure 2: Location and genetic type of the populations studied

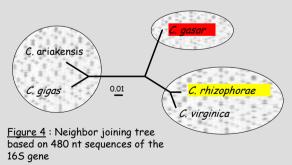
The karyotypes of the French Guyana and Senegal sample were very similar (Figure 3), corresponding to *C. gasar* with 6 metacentric (m) and 4 submetacentric (sm) chromosome pairs. These were clearly different from the *C. rhizophorae* karyotype previously described in Mexico Samples (see Leitão *et al.*, 1999, *Malacologia*, 41(1), 175-186).



<u>Figure 3</u>: Giemsa-stained karyotype of C. gasar samples from Senegal (A) and French Guyana (B).



The information of the 165 fragment sequences from different *Crassostrea* species (O' Foighil *et al.*, 1995, *JEMBE*, 192, 211-220) indicated an intermediate position of *C. gasar* between the *C. gigas* - *C. ariakensis* and *C. virginica* - *C. rhizophorae* groups (Figure 4).



#### CONCLUSIONS

This is the first description of the presence of C. gasar in South America as well as the first genetic study on this species. These results are raising questions on:

- Was C. gasar man-introduced into South America or is it naturally present?
- What is the nuclear genetic divergence between the two species and do they inter-cross?
- How many oyster species are present on the South American Atlantic coasts?