# GENETIC IMPACT OF THE REPRODUCTION DYNAMICS IN THE EUROPEAN FLAT OYSTER *Ostrea edulis*

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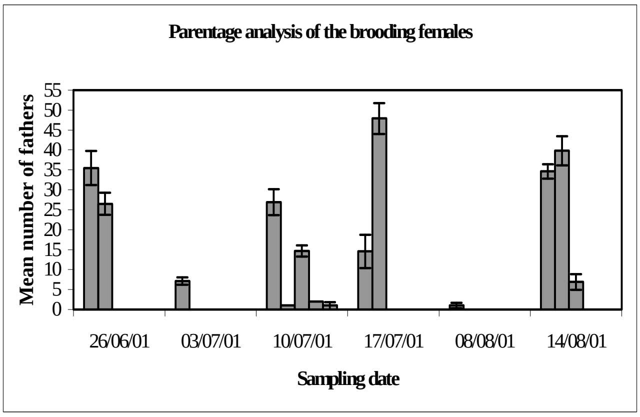
The European flat oyster (*Ostrea edulis* L.) is a marine bivalve whose natural geographical distribution ranges along the European Atlantic coast from Norway to Morocco, in addition to the Mediterranean and Black Sea. The latest results obtained on the genetic differentiation between these populations have led us to persue studies at a finer scale, in order to estimate the effective number of breeders and the temporal dynamics of reproduction and, more specially, recruitment. Several experiments were performed to document (1) the variance in allele frequencies during a natural settlement period, (2) the paternal contribution to fertilization by analyzing larvae sampled at the brooding stage within individual females, (3) the variance of individual reproductive success within an experimental population.

Firstly, 3 sets of collectors were successively deployed every two weeks and one set left during the whole recruitment period in 2001. In addition, adults oysters were sampled including 14 brooding females (i.e. females presenting larvae in their paleal cavity). Their larvae were sampled and preserved in ethanol. Mitochondrial (12S fragment) and microsatellite (four loci) analyses were performed.




Although the temporal cohorts did not exhibit any differentiation on the basis of the microsatellite markers, a slight but significant differentiation was observed with the mitochondrial marker. Moreover, our data on the genetic variability of single-female progenies show that females can be fertilized by a highly variable number of males, which can be, in some cases, very low (See Figure). In such cases, a temporary low effective could lead to a level of inbreeding (even low) which would explain the correlation between growth and heterosis often observed.

More investigations are needed to directly demonstrate that each male fertilizes several females. If males succeed in fertilizing several females, variability in environmental conditions should increase female variance in reproductive success more than male variance in reproductive success, thus reducing mitochondrial relative to nuclear effective size as has been observed in the study of the genetic structure of the populations along the species range.

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# *Ostrea edulis*

**+ introduction in USA, Canada, Japan...**

# *O. edulis* population genetics:

## *How is the genetic variability distributed...*

### *... in space ?*

- Within *vs* among populations ?
- Atlantic *vs* Mediterranean populations ?
- Isolation by distance ?

### *... among genomes ?*

- Nuclear *vs* Cytoplasmic ?

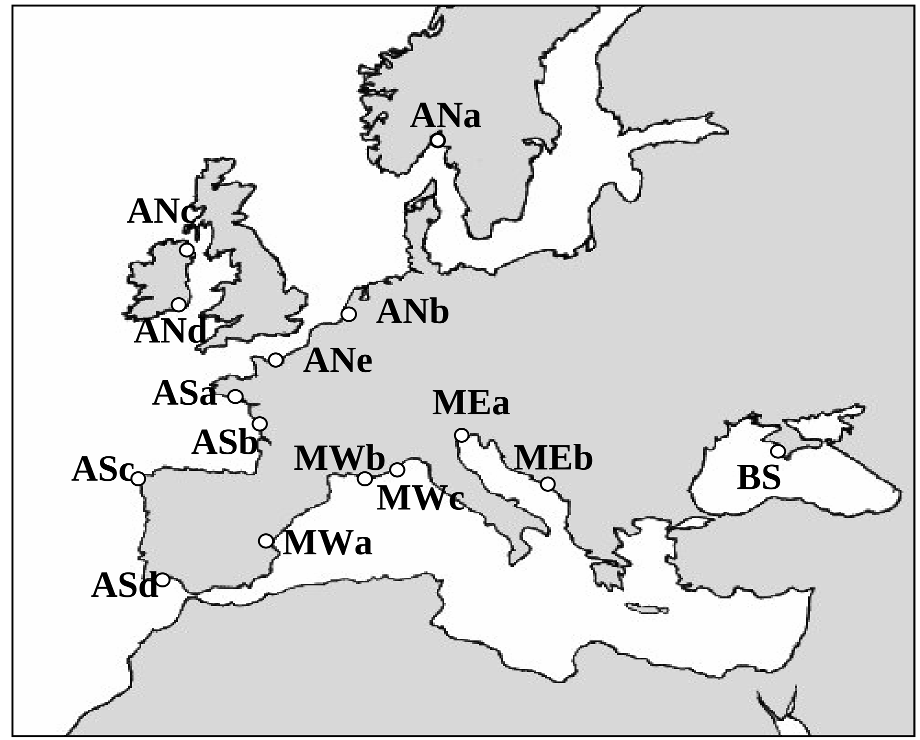
### *... over time and/or generations ?*

- Adults *vs* juveniles
- Females *vs* larvae ?

# Spatial distribution ?

**Sampling**:

15 populations sampled
14 to 50 individuals per location



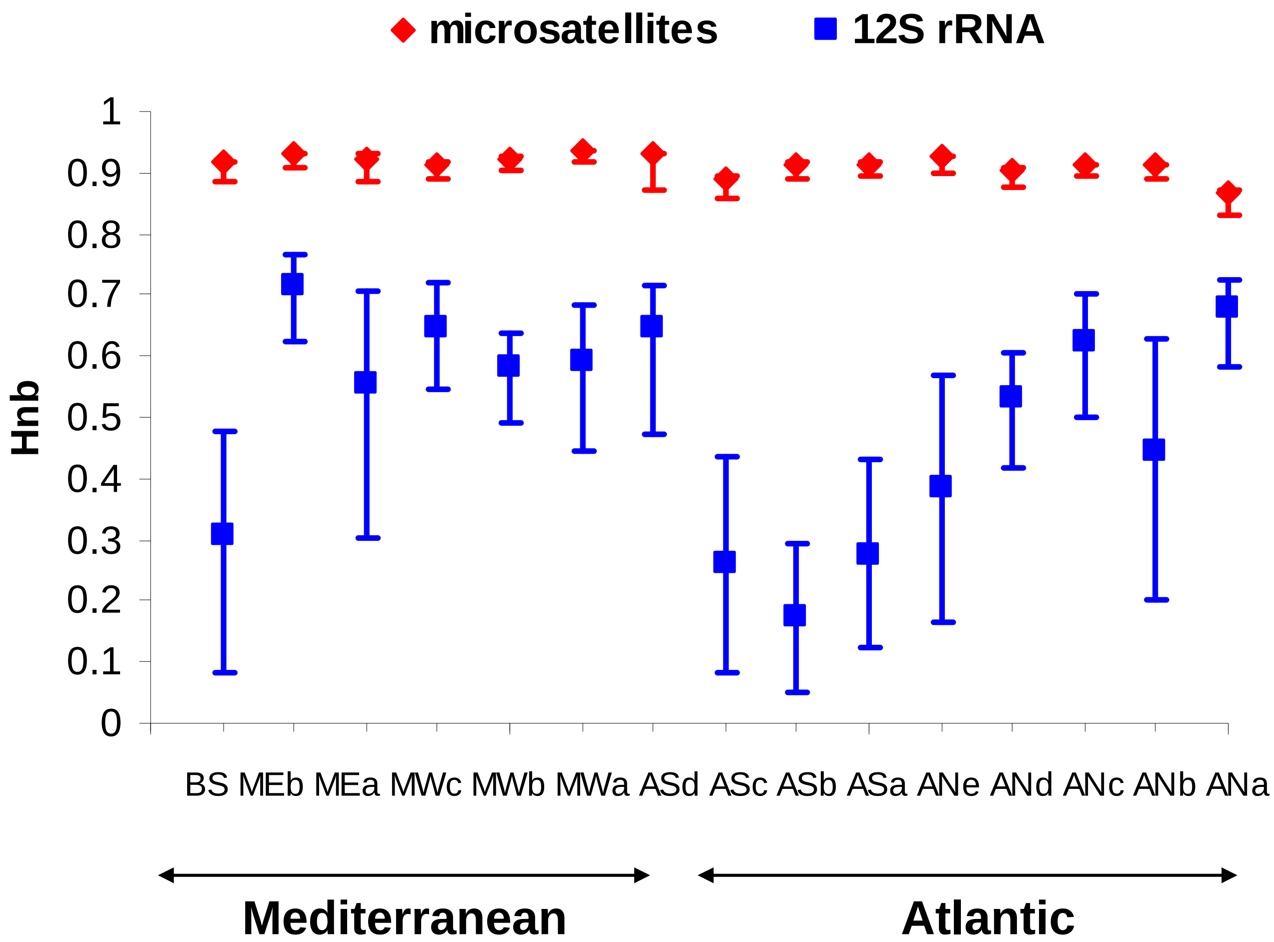

**Markers**:

- Allozyme loci (Saavedra *et al.*, 1993, 1995)
- microsatellite loci (Launey *et al.*, 2002)
- 12S rRNA SSCP (Diaz Almela *et al.* in press)

# Within population diversity :

**<u>Microsatellites</u>:** **allele/locus/pop = 18.5 ± 4.5**
**mean He = 0.914 ± 0.018**

**<u>12S rRNA</u>:** **14 SSCP haplotypes**

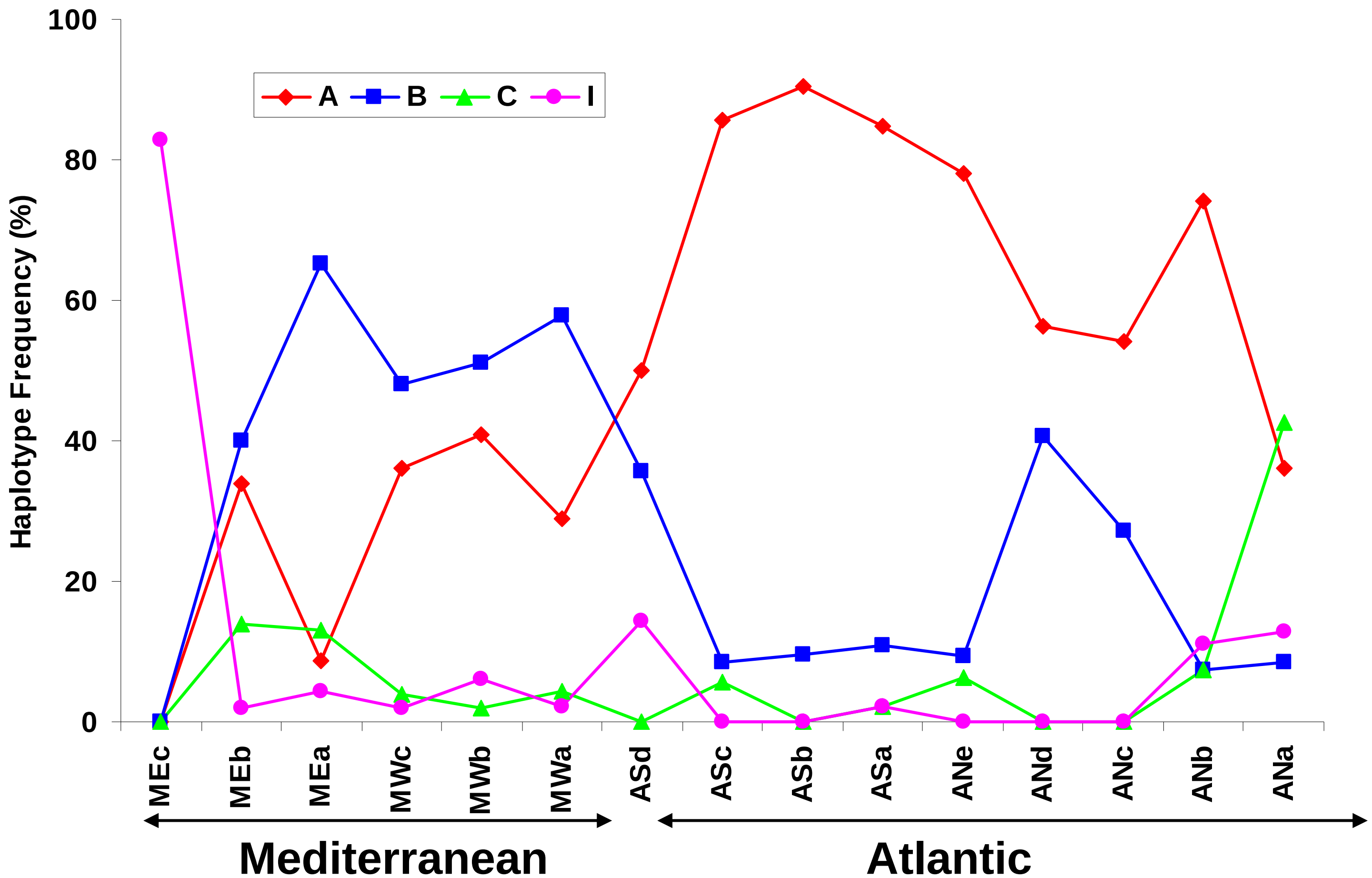


# Among population differentiation :

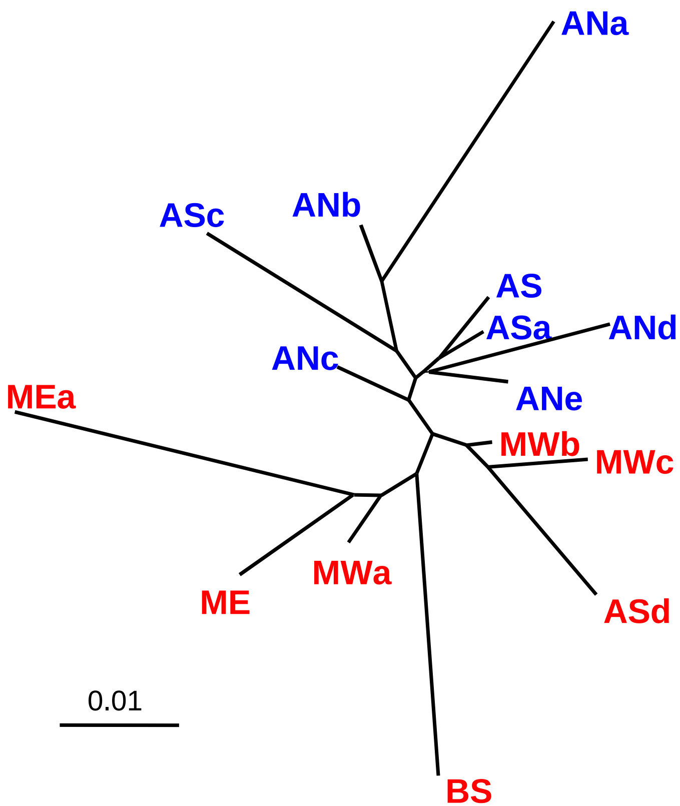
**Microsatellites** : ***Fst*** **= 0.019*****

**12S rRNA** : ***Fst*** **= 0.224*****

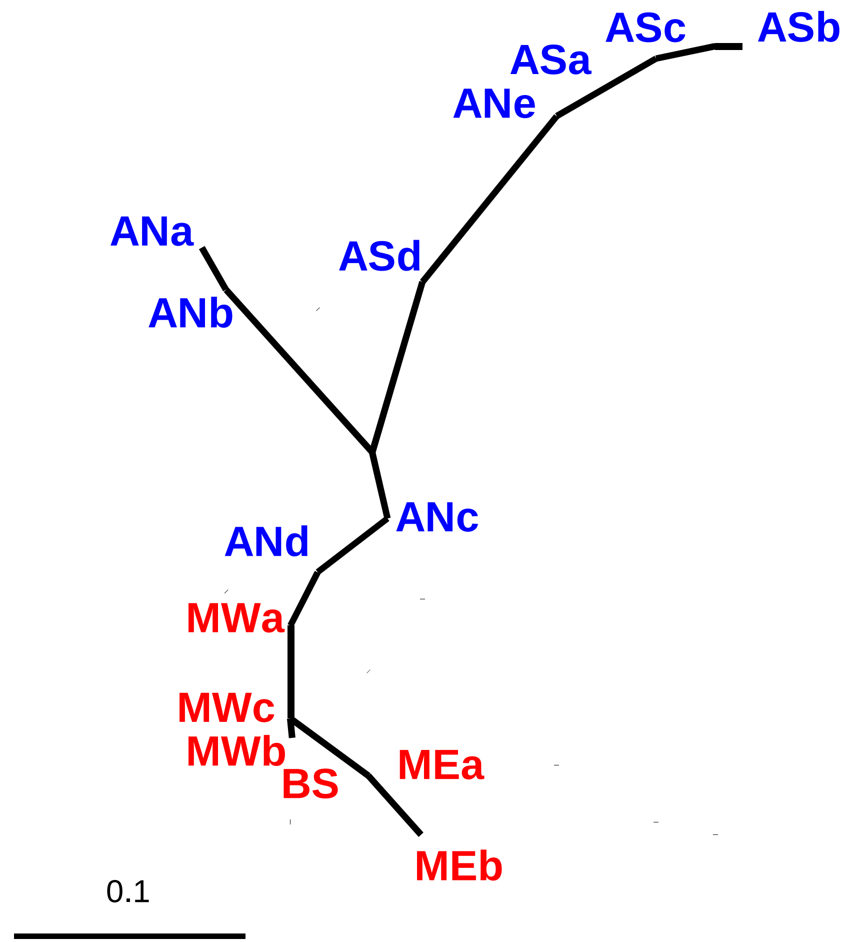
***10 X***



# Among population differentiation :

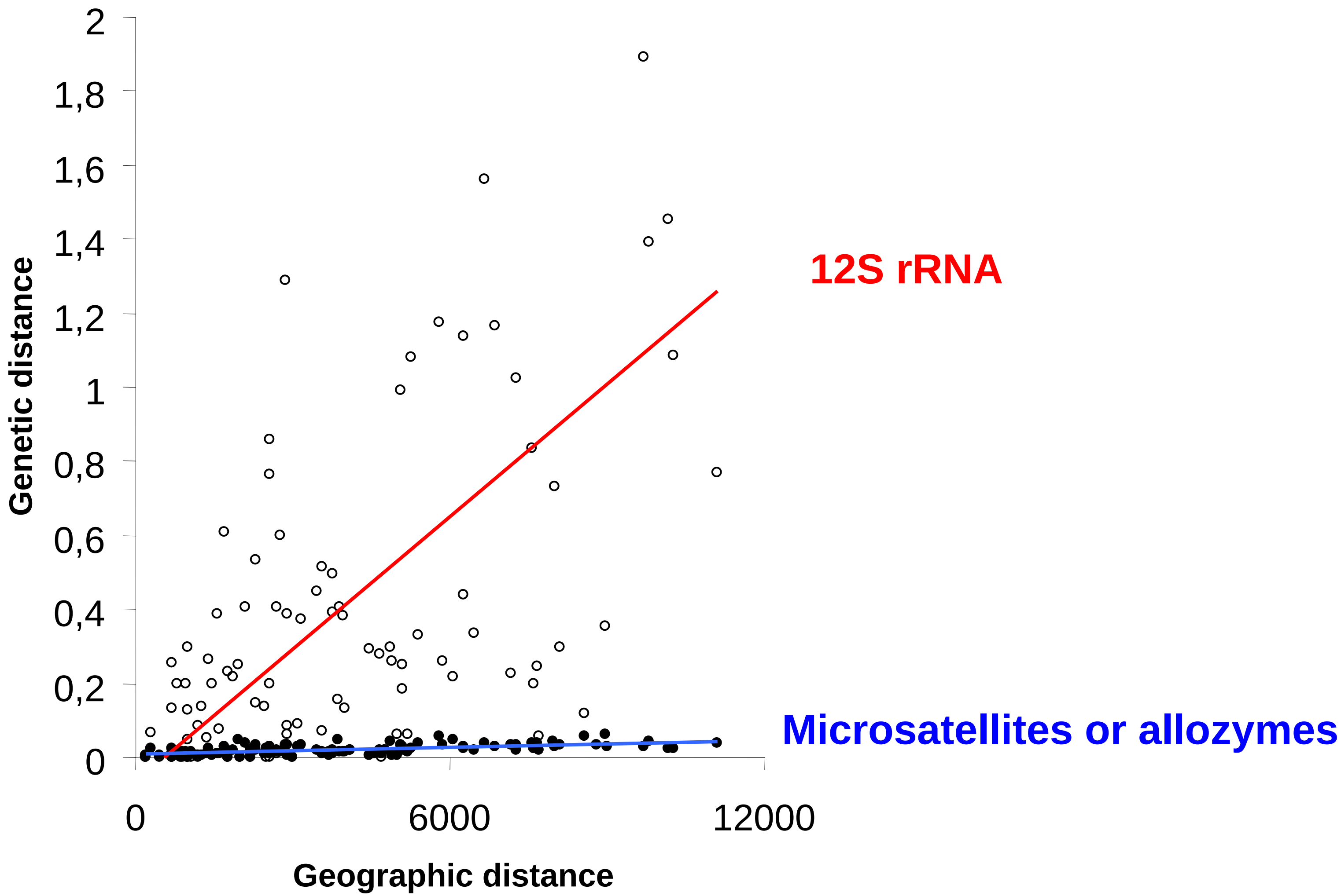



Microsatellites



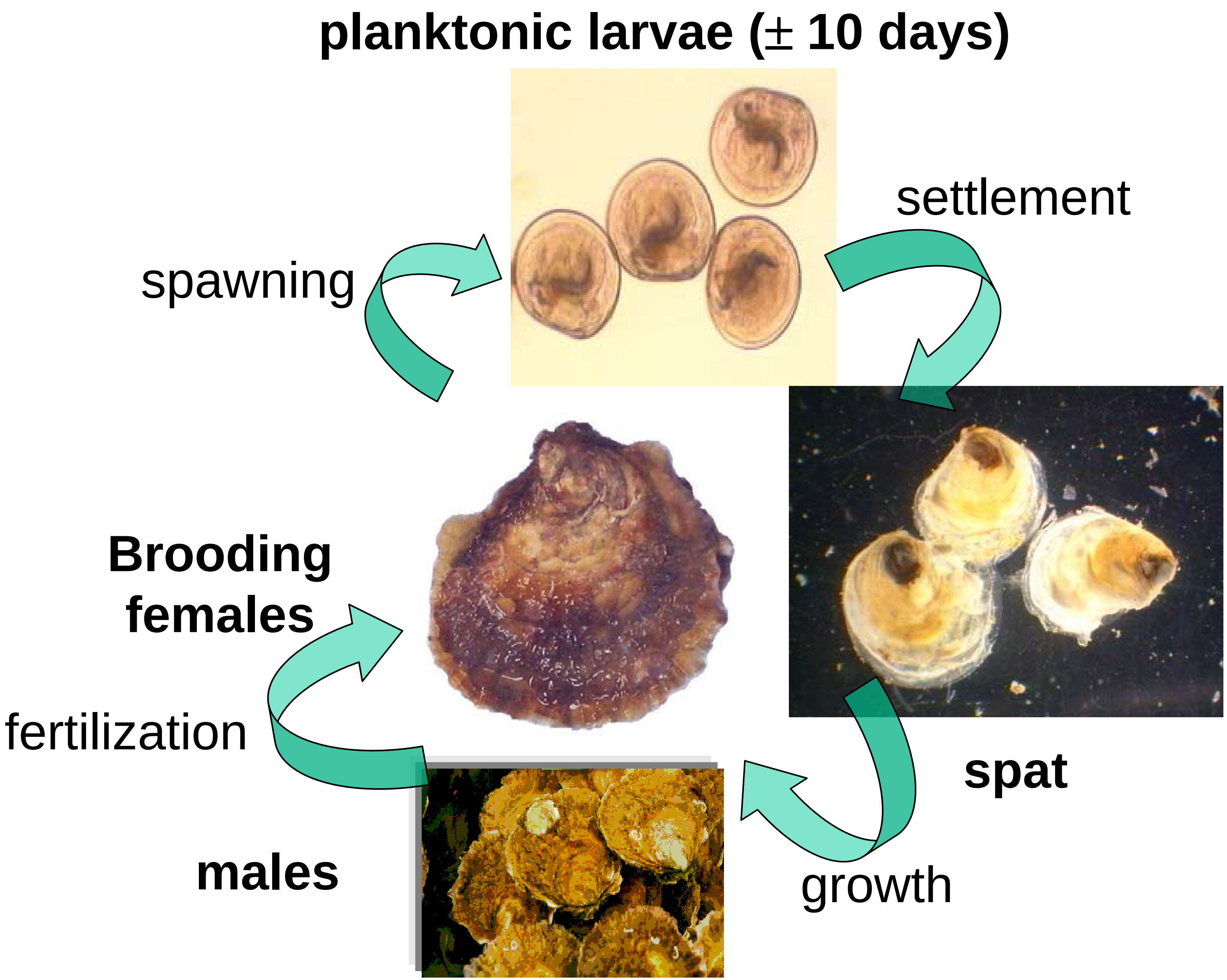

12S rRNA

# Isolation by distance :

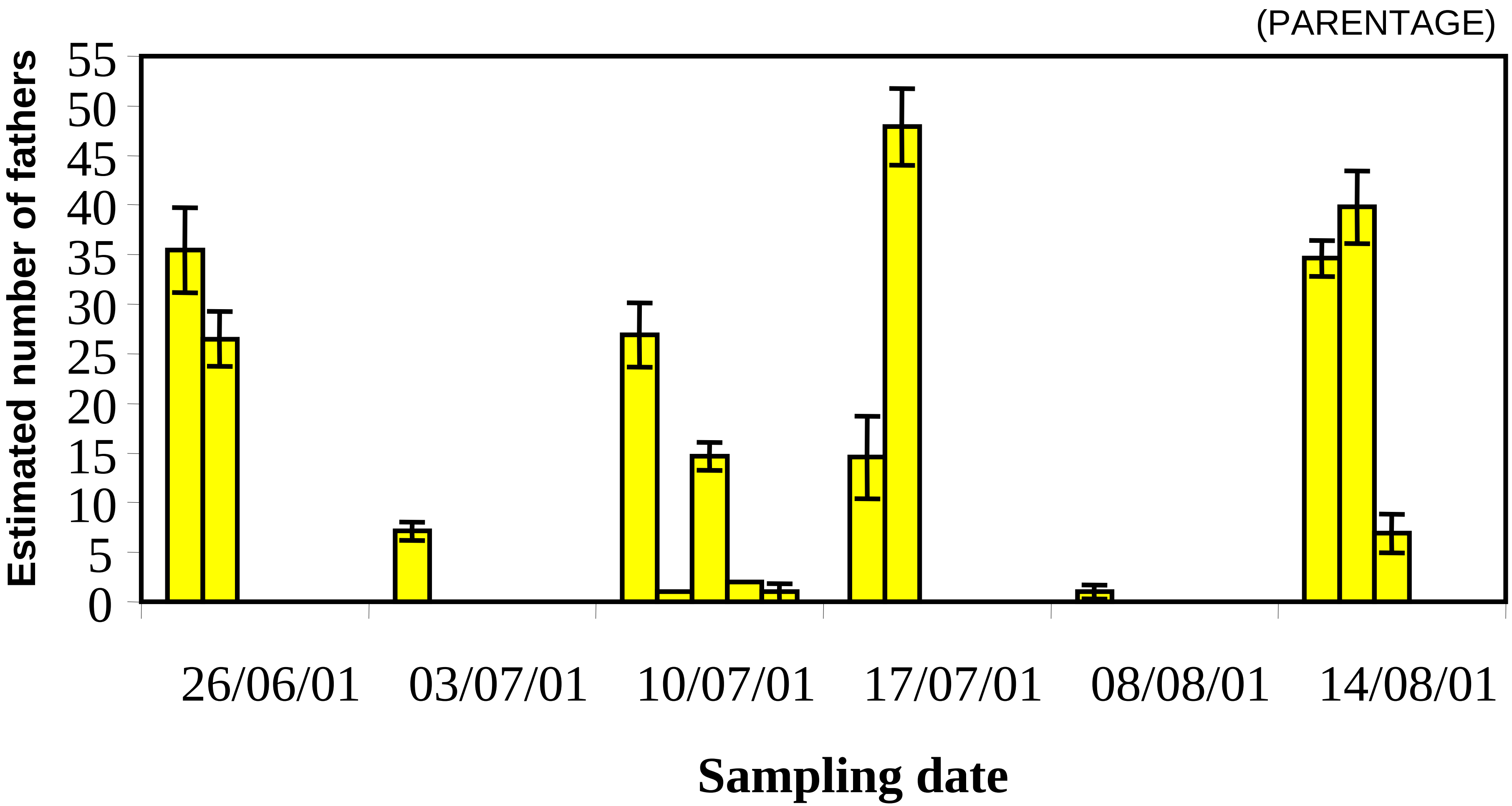



**Higher variance in reproductive success in the female than in the male ?**

# Reproductive cycle

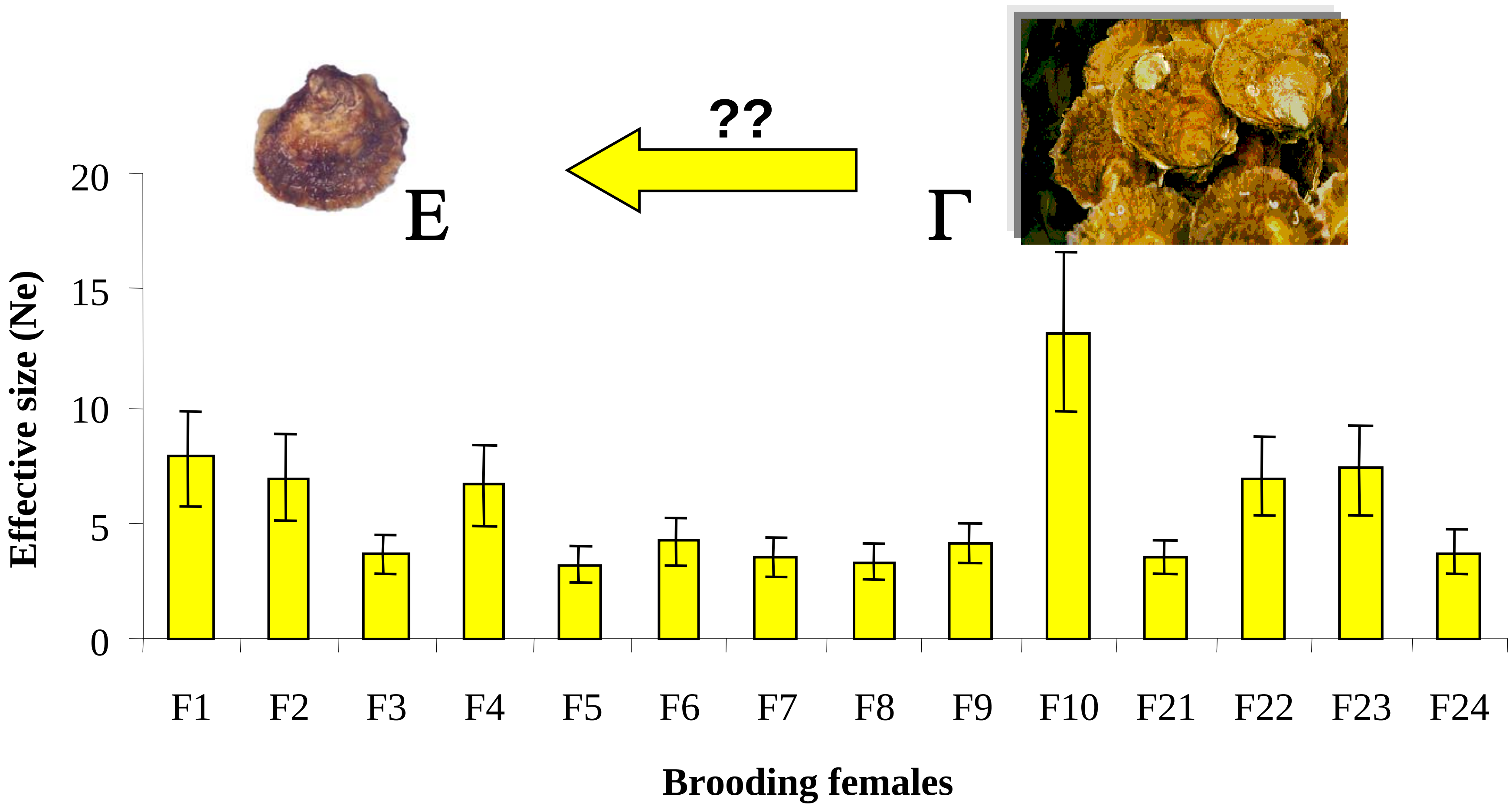


# how many males / female ?

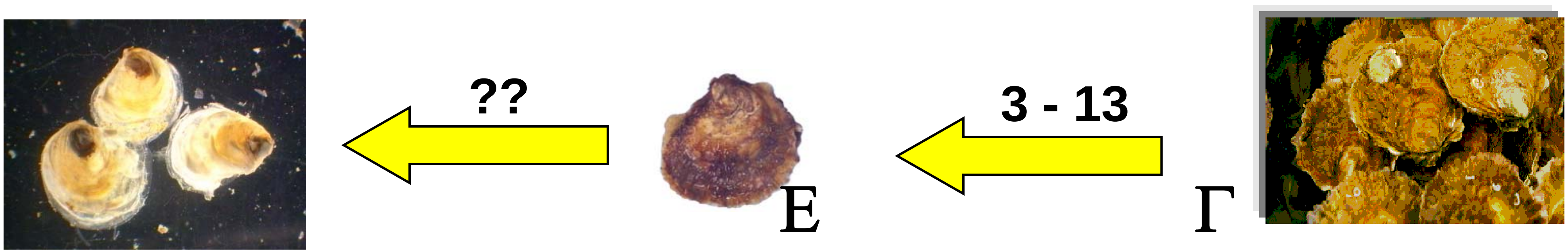



**Very variable number of males / female**

# Variability over generation:
# how many males / female ?



# Variability over generations : *Ne* = ?



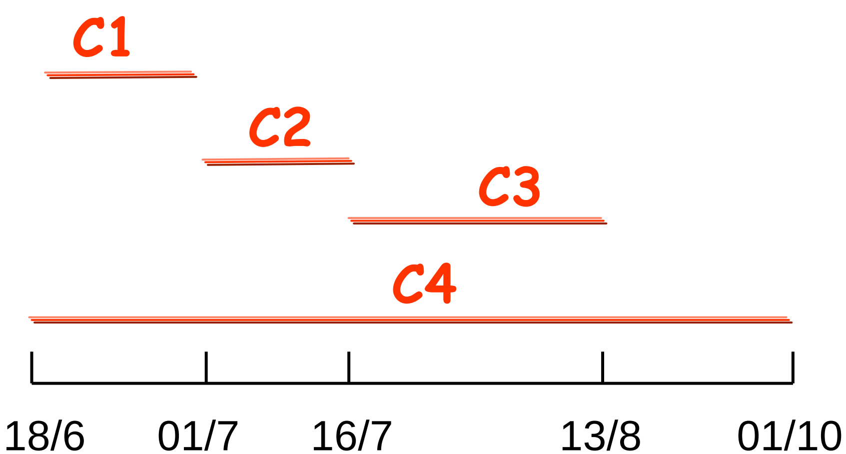

- **1994** : **one cohort recruited over 15 days in Sète**

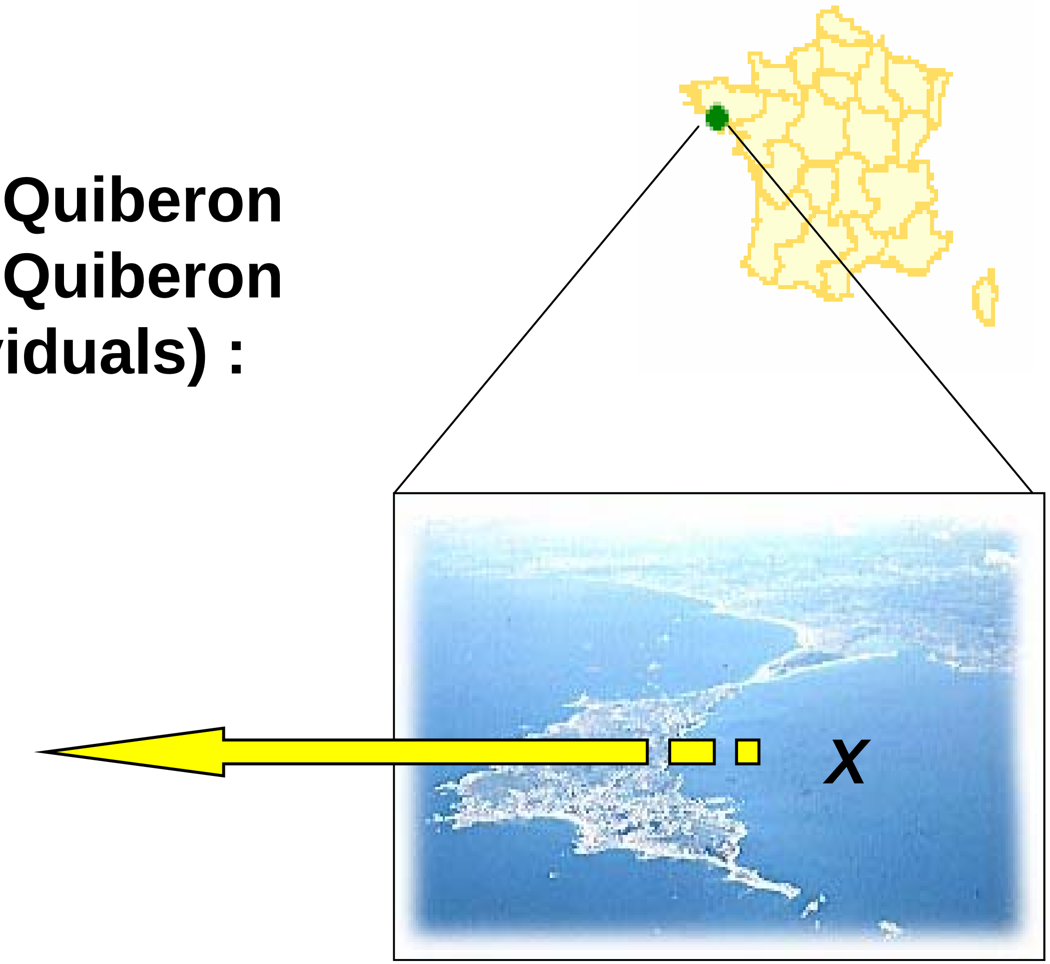
  **alleles / locus : spat (16.4) ≠ adults (21.8)**

  **Ne = 16** [11,23] **(Launey *et al*., in prep)**

# Variability over generations: *Ne* = ?

**1995 : 49 adults collected in Quiberon**
**2001 : 68 adults collected in Quiberon**
**+ 4 cohorts (672 individuals) :**




**4 microsatellites**
**12S sequence**

# Differentiation among cohorts (*Fst*)

| | Cohort 1 | Cohort 2 | Cohort 3 | Cohort 4 | Adults |
|---|---|---|---|---|---|
| Cohort 1 | | -0.019 | -0.014 | **0.052*** | **0.058*** |
| Cohort 2 | 0.000 | | -0.024 | 0.041 | 0.049 |
| Cohort 3 | 0.001 | -0.001 | | 0.021 | 0.029 |
| Cohort 4 | 0.000 | 0.002 | 0.002 | | -0.019 |
| Adults | 0.001 | 0.002 | 0.002 | 0.000 | |

*** $p < 0.05$**

**No differentiation using microsatellites**

**Low mitochondrial differentiation (cohort 1)**

# Variability over generations: *Ne* = ?

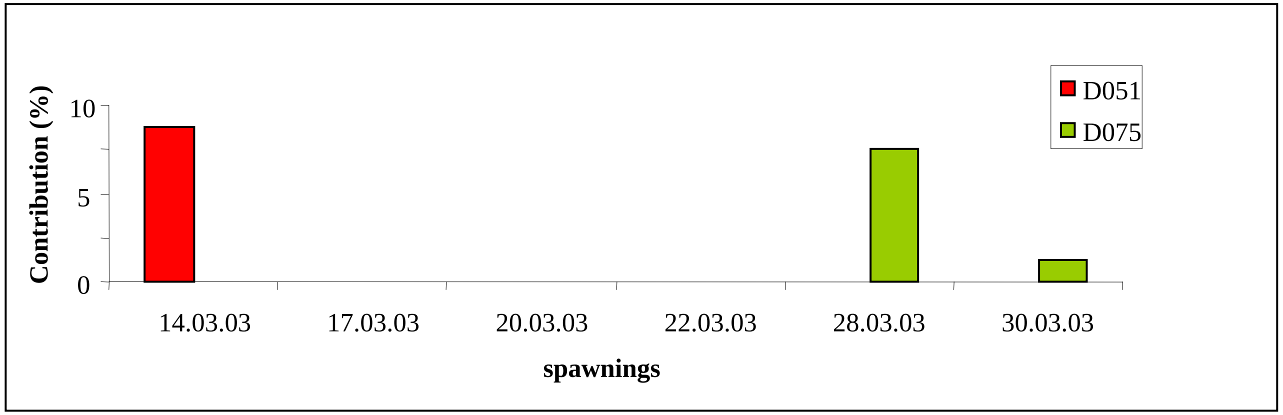
- **alleles / locus : spat = adults**

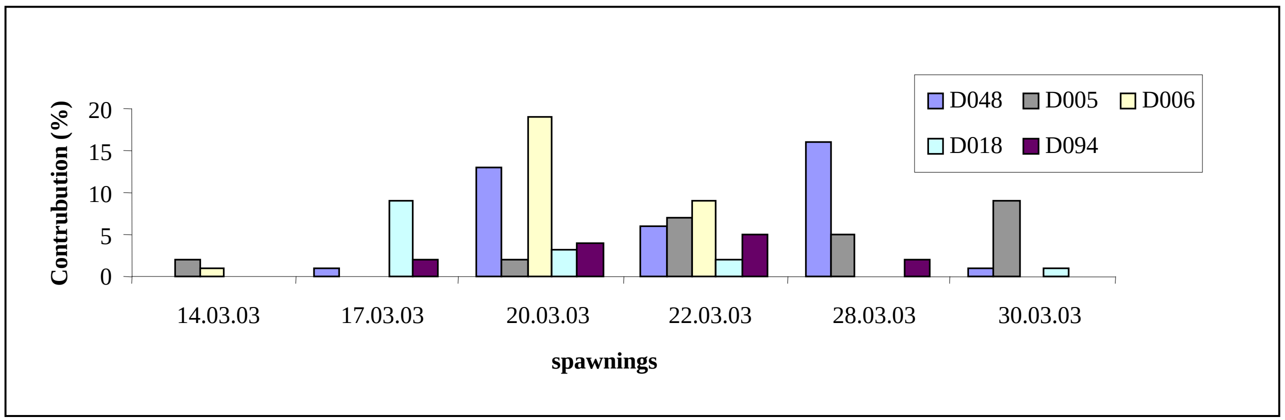
- **adults /spat : *Ne* = 135** [44, +∞]

- **adults 1994 / adults 2001 : Ne = 137** [ 60, +∞]


# Spawning behaviour under controlled conditions :

## 60 individuals in a spawning tank
## Ne / spawning event = 7 to 17




## Variation of reproductive success

# Risks and benefits of hatchery propagated spat to enhance production ?

- **Breeding programs have been initiated to improve disease resistance** (Naciri-Graven et al., 1998; Culloty et al., 2001)

- **Hatchery populations usually have low *Ne*** (Hedgecock et al., 1992) **in *O. edulis* :**
  - ✓ **Saavedra & Guerra (1996): *Ne* ≈ 4**
  - ✓ **Launey et al. (2001): *Ne* = 3 to 20**

- **Today hatchery-based production is still very limited**

- **Its development could have a positive impact in terms of aquaculture but might have a strong negative impact on genetic diversity of wild populations.**