

Variance of reproductive success in the European flat oyster *Ostrea edulis* assessed by microsatellite-based parentage analyses in a natural population and in hatchery

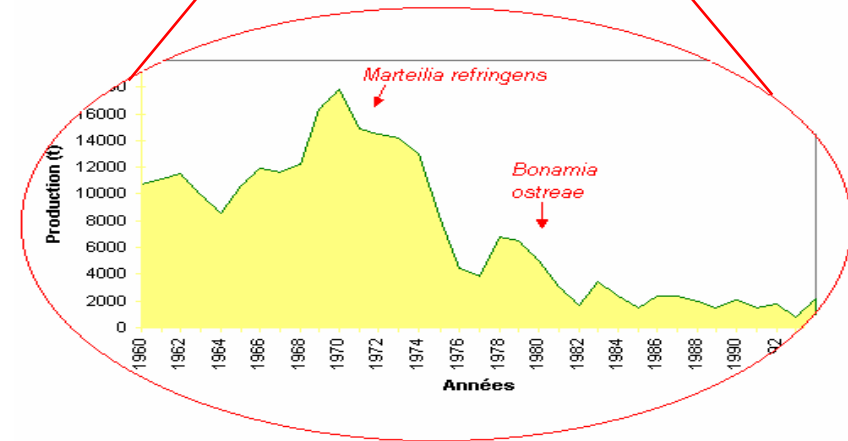
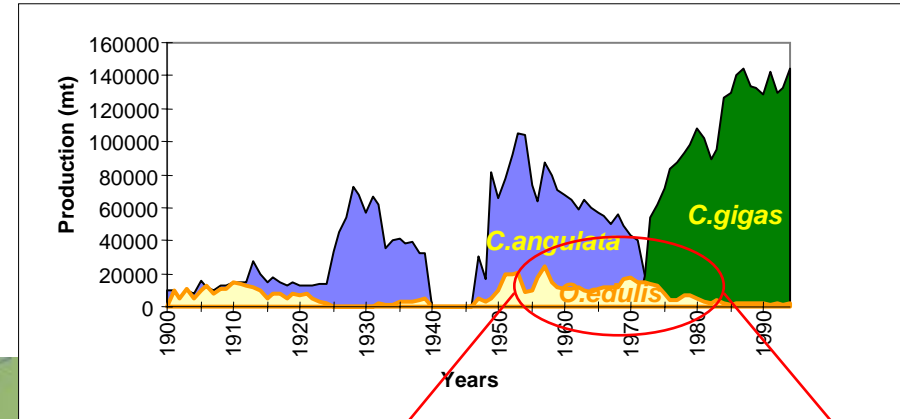
Delphine Lallias¹, Nicolas Taxis^{1,2}, **Pierre Boudry**¹,
François Bonhomme² and Sylvie Lapègue¹

¹ Laboratoire IFREMER de Génétique et Pathologie, F-17390 La Tremblade, France

² Laboratoire Génome, Populations et Interactions - CNRS UMR 5000 Station Méditerranéenne de l'Environnement Littoral, 1 quai de la daurade, 34200 Sète, France



Ostrea edulis: the native european flat oyster



Ostrea edulis in 2004 :

World production = 5.100 t/y
French production = 1.500 t/y

(JAZIRI, 1985)

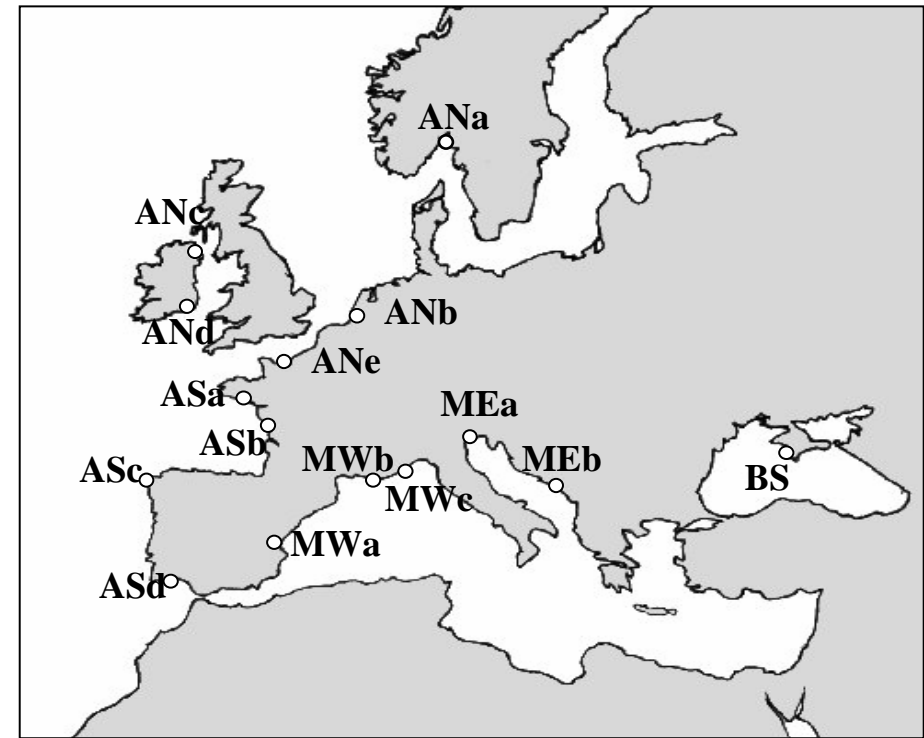
• Spatial distribution ?

Sampling:

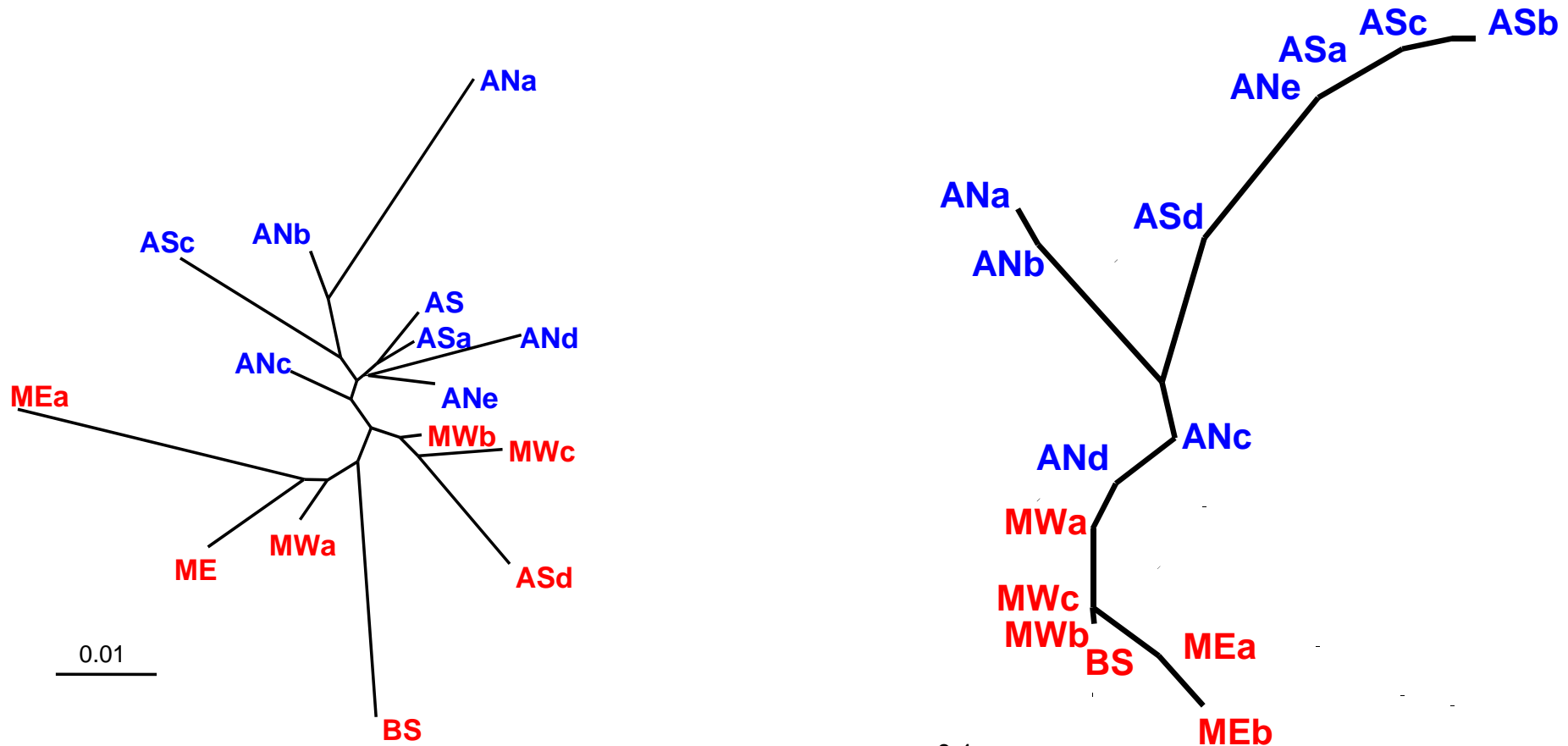
15 populations sampled
14 to 50 individuals per location

Markers:

- Allozyme loci (Saavedra *et al.*, 1993, 1995)
- microsatellite loci (Launey *et al.*, 2002)
- 12S rRNA SSCP (Diaz Almela *et al.*, 2004)



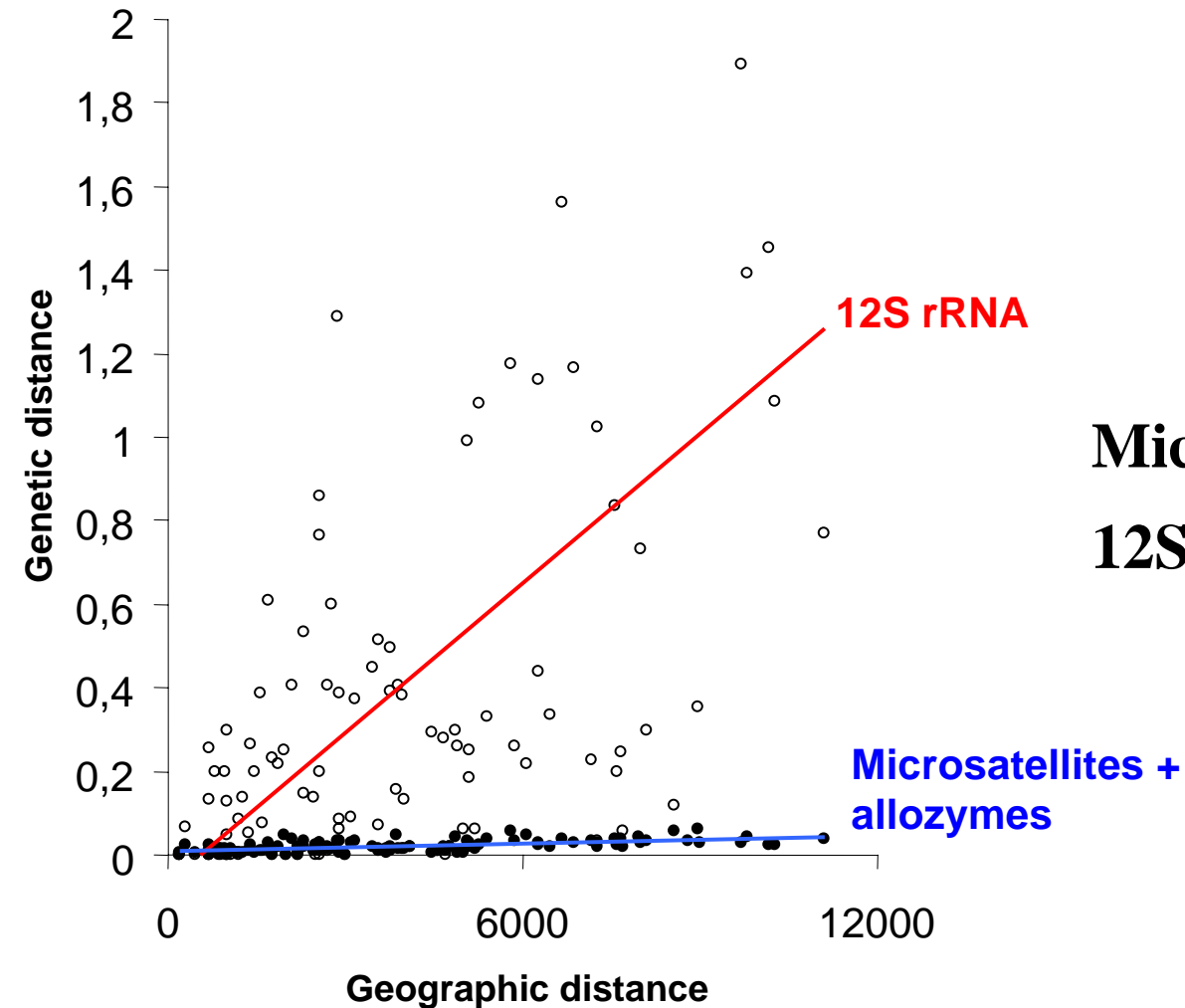
*Mediterranean sea versus Atlantic ocean
differentiation :*



Microsatellite markers

12S rRNA

Isolation by distance :



Microsatellites : $F_{st} = 0.019^{***}$

12S rRNA : $F_{st} = 0.224^{***}$

10 X



Higher variance in reproductive success in females than in males ?



**Parentage analyses in a natural population
of *Ostrea edulis***



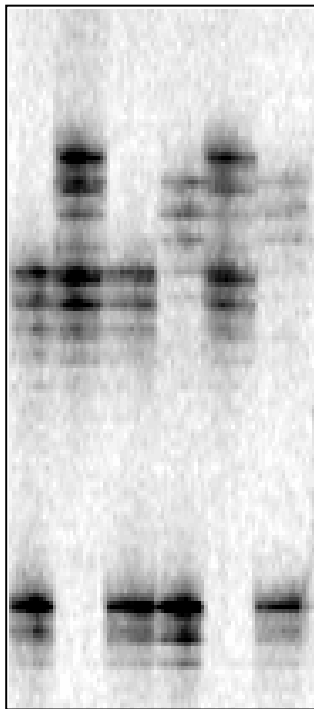
1- SAMPLING : 14 brooding females issued from a natural population (2001)



2- SCORING : 80 larvae per female
- 4 microsatellites markers : OeduJ12, U2, H15 and T5
(Launey *et al.*, 2002)

Locus OeduU2

Allele 188
→



Allele 156
→



3- PATERNITY ANALYSIS :

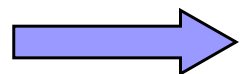
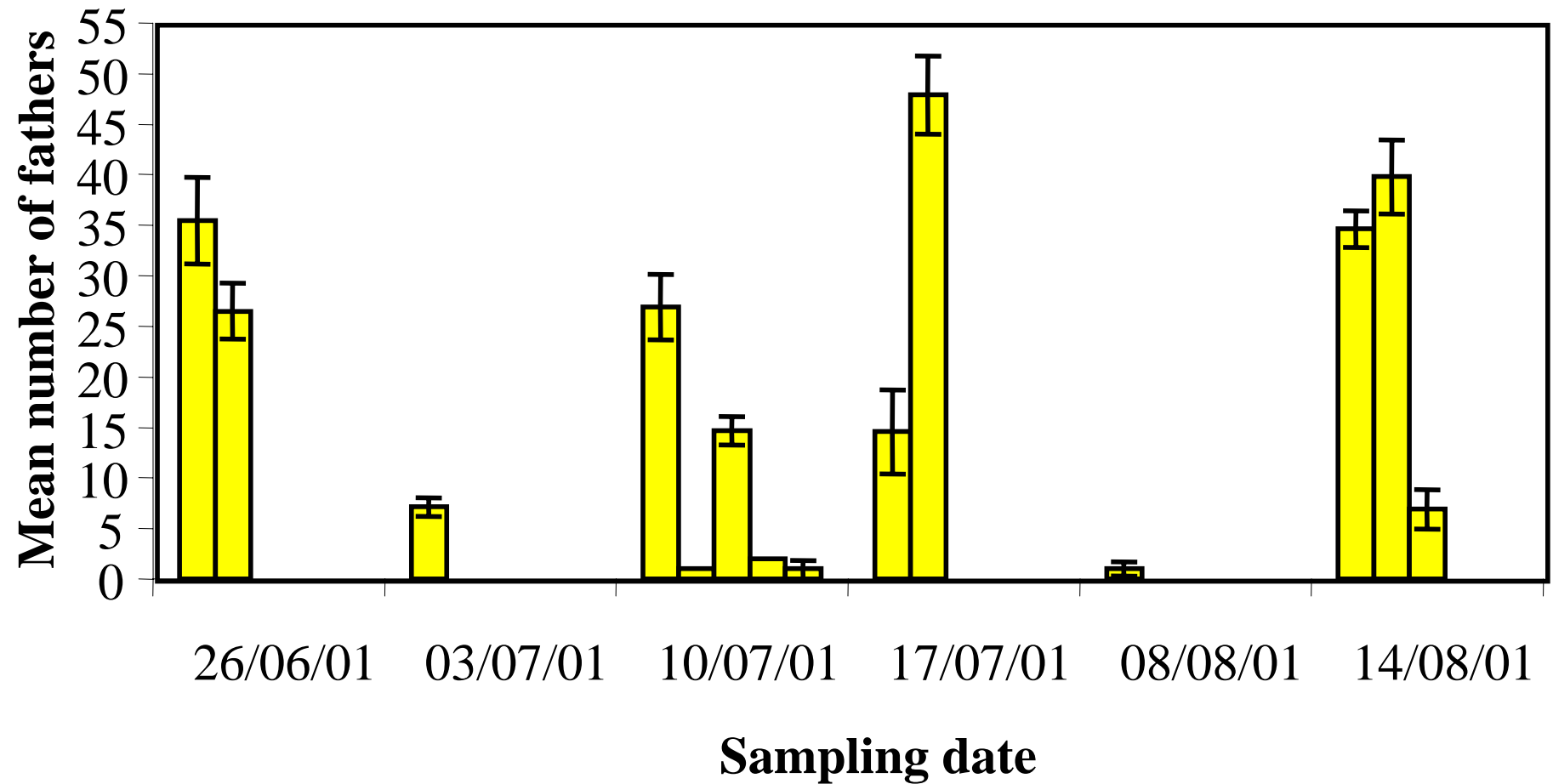
Two parental reconstruction softwares:

- PARENTAGE (Emery *et al.*, 2001) : Bayesian
- GERUD1.0 (Jones, 2001) : combinatorial

How many males fertilize a female ?



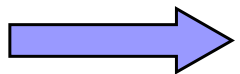
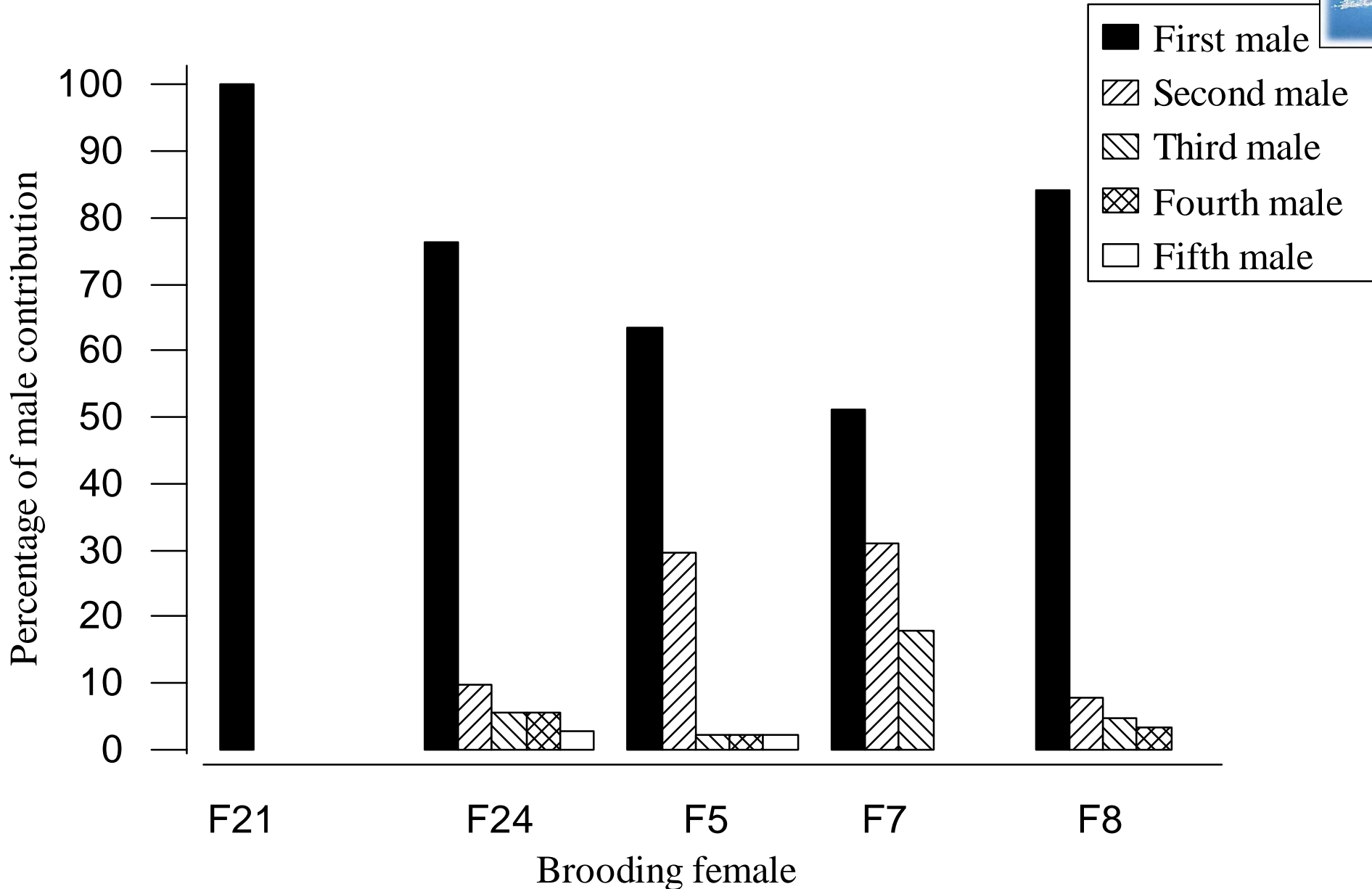
Number of males fertilizing
each of the 14 brooding females



Very variable number of males per female (1 to > 40)

(PARENTAGE)

Male contributions within each female



Variance in individual male contribution : male with the highest contribution fathered from 51 to 100% of larvae



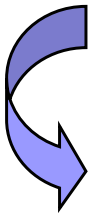
~~Parentage analyses in a natural population
of *Ostrea edulis*~~

Paternal analyses of brooding females of
Ostrea edulis in a wild population



These results are of limited value to answer our initial question because :

- The population size is poorly known
- The effective population size is difficult to estimate
- The spatial distribution of males and females is unknown
- The effective sex ratio unknown
- ...



What about experimental (hatchery) populations ?



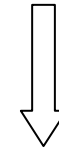


1- COLLECTION OF LARVAE :

Six successive spawning events over 2 weeks
(14/03 to 30/03)

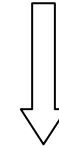


2- **GENOTYPING** : 63 oysters, 80 larvae per spawning
- 3 microsatellites loci: OeduJ12, U2, T5 (Launey *et al.*, 2002)



3- GENETIC ANALYSES :

- Genetic differentiation adult/mass spawning θ
- Effective population size (NeEstimator 1.3. Software)

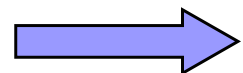
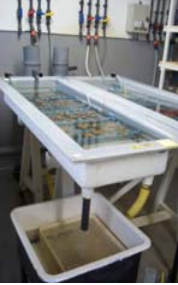


4- PARENTAGE ANALYSES :

- Combined exclusion probability 98.3%
- PAPA software (Duchesne *et al.*, 2002) : max. likelihood

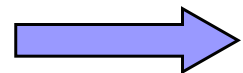
- **Effective population size**

	Ne (temporal method)
14/03/03	21.0 [12.4 36.2]
17/03/03	12.5 [7.8 19.6]
20/03/03	21.0 [12.3 36.5]
22/03/03	22.3 [12.9 39.6]
28/03/03	33.2 [17.8 70.1]
30/03/03	29.6 [16.3 58.9]



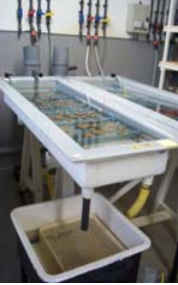
Relatively small Ne (12-33)

Genetic variability of a cohort : $Ne < 20$ (Hedgecock *et al.*, submitted)



Large 95% CI : small number of markers

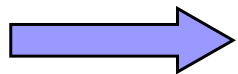
Genetic analyses



- Genetic differentiation ($F_{st} \times 100$)

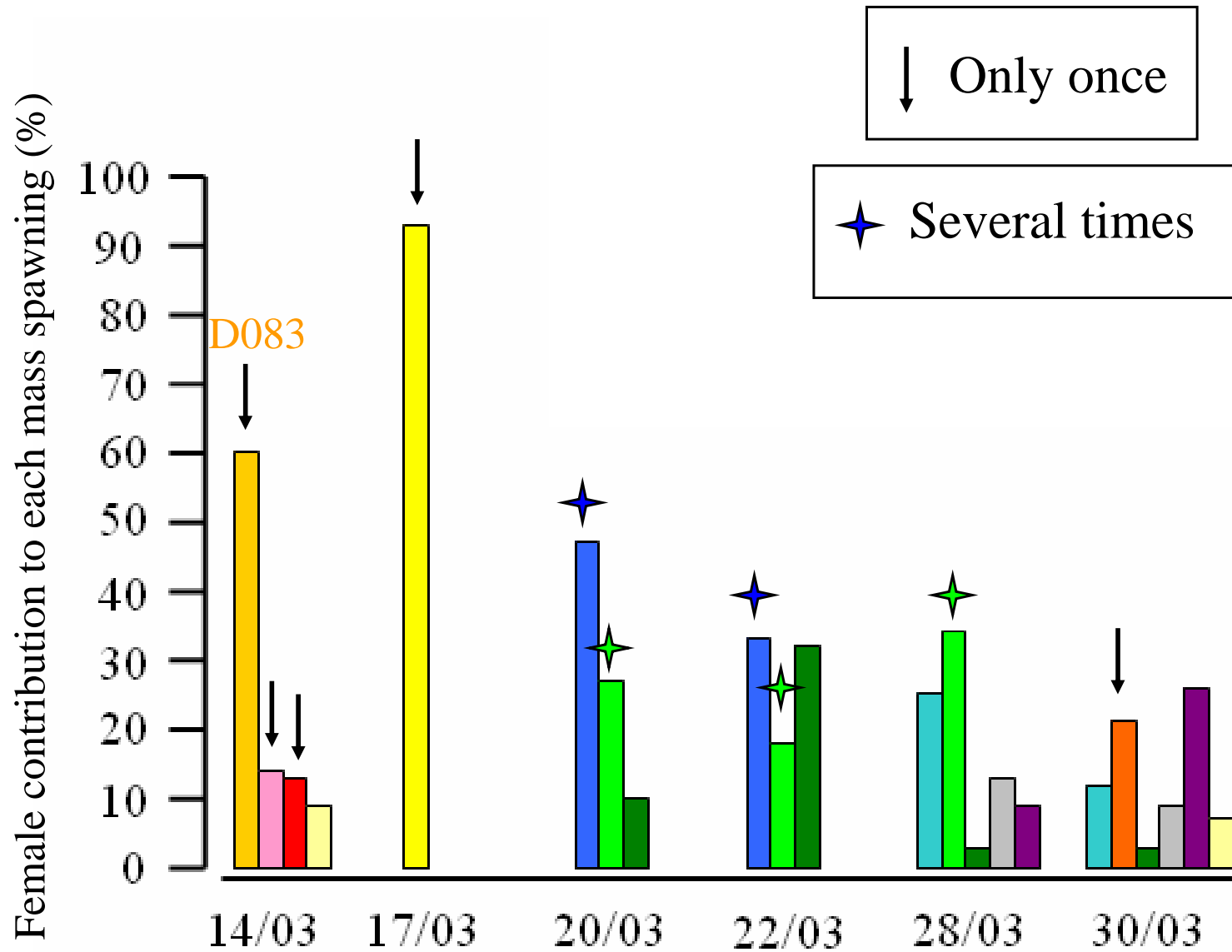
	14/03/03	17/03/03	20/03/03	22/03/03	28/03/03	30/03/03
<u>Adults</u>	3 ^{***}	5.5 ^{***}	3.1 ^{***}	1.5 ^{***}	1.3 ^{***}	1.2 ^{***}
14/03/03	-	11.9 ^{***}	7.1 ^{***}	5.9 ^{***}	5 ^{***}	5.1 ^{***}
17/03/03	-	-	9.8 ^{***}	6.8 ^{***}	7.9 ^{***}	7.8 ^{***}
20/03/03	-	-	-	0.7 ^{**}	4.2 ^{***}	4.5 ^{***}
22/03/03	-	-	-	-	3 ^{***}	3 ^{***}
28/03/03	-	-	-	-	-	1.8 ^{***}

	14	14+17	14+17+20	14+17+20+22	14+17+20+22+28	14+17+20+22+28+30
<u>Adult</u>	3 ^{***}	1.1 ^{***}	0.6 ^{**}	0.6 ^{**}	0.3 [*]	0.2 ^{NS}

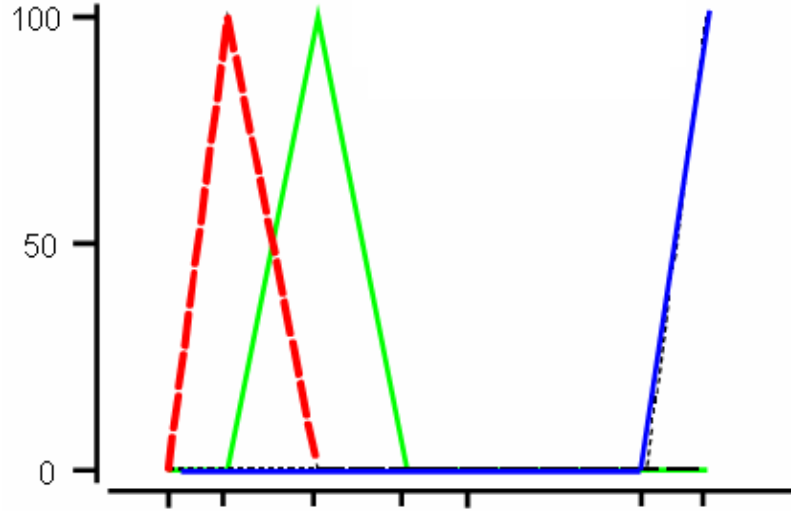
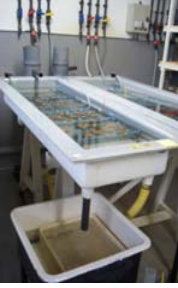


While pooling the successive spawning cohorts, genetic differentiation becomes blurred

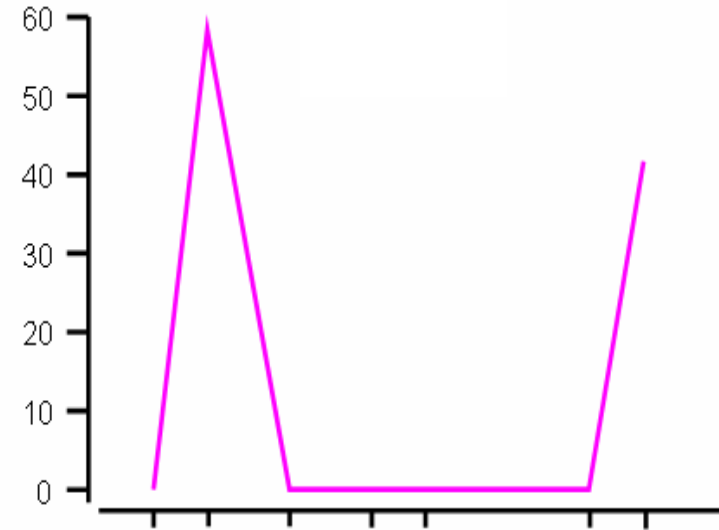
Parentage analyses : Female contributions



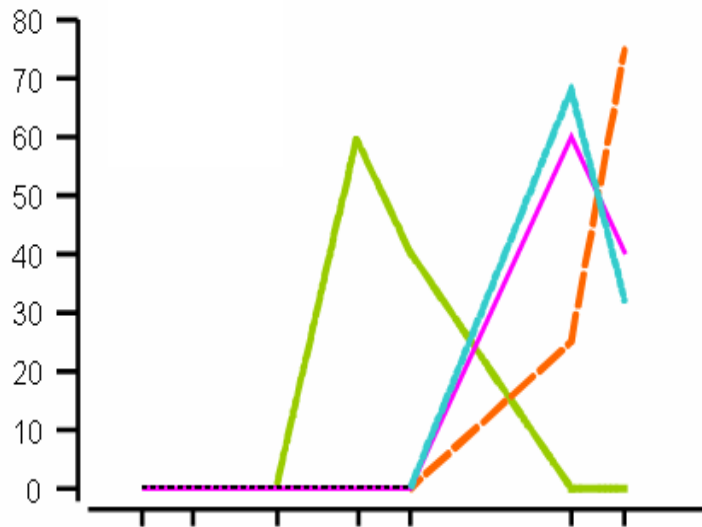
Parentage analyses : Dynamics of female larval release



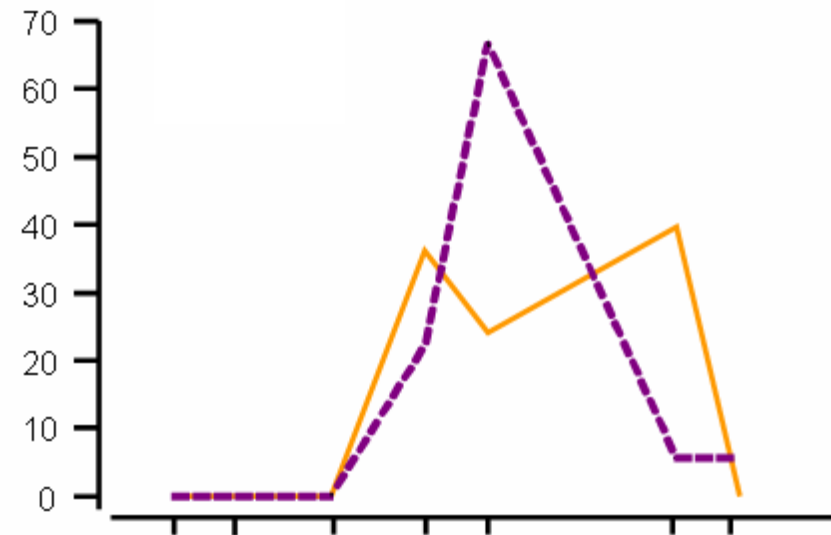
In 1 mass spawning



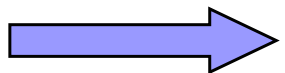
In 2 spaced mass spawning



In 2 successive mass spawning

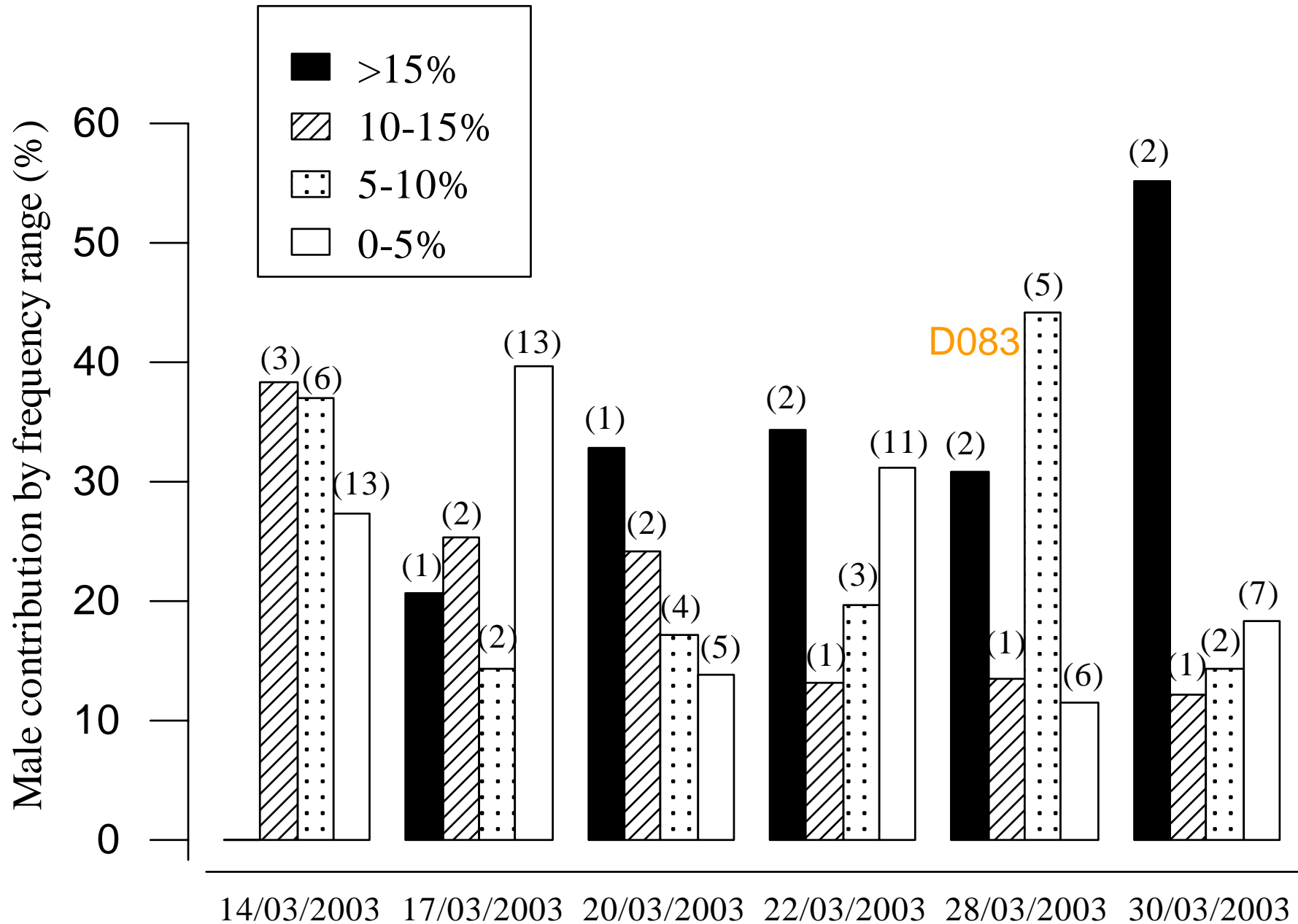


In 3 or 4 mass spawning

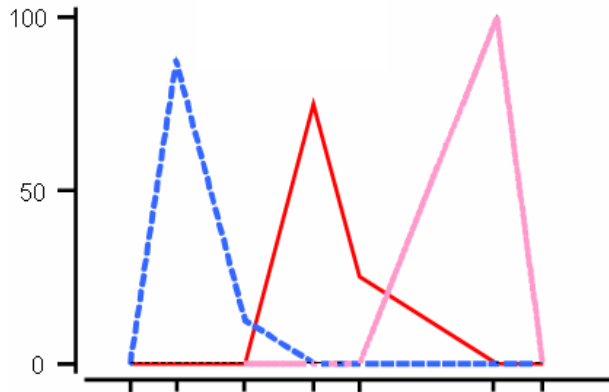


Variable extension in time of larval release and its dynamics

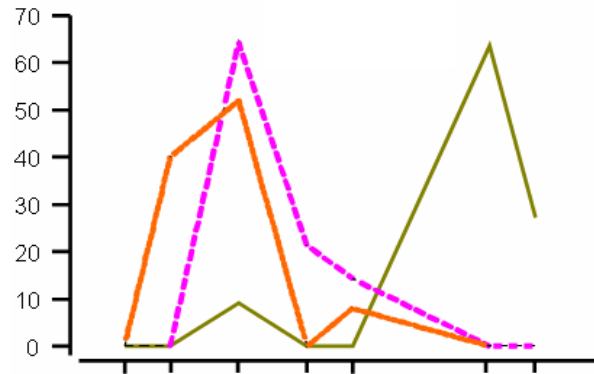
Parentage analyses : Male contributions



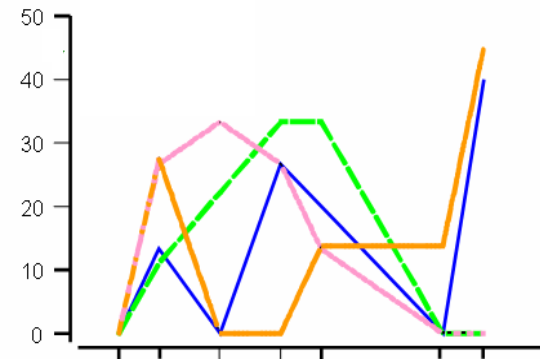
Parentage analyses : dynamics of male contributions



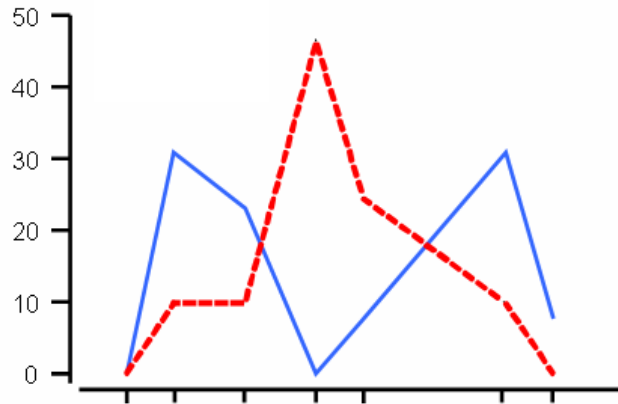
In 1 or 2 mass spawning



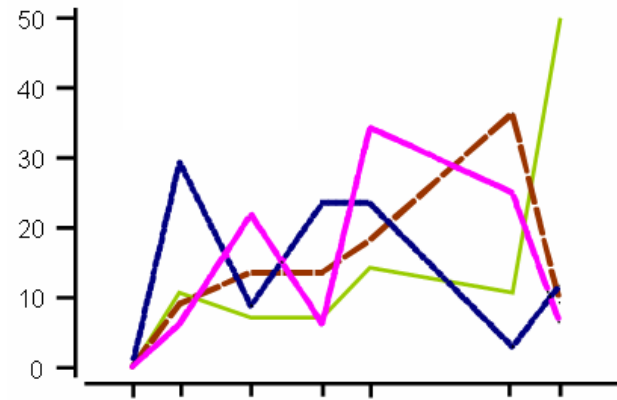
In 3 mass spawning



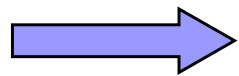
In 4 mass spawning



In 5 mass spawning

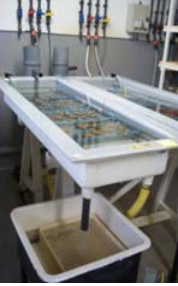


In 6 mass spawning



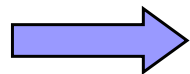
More complex patterns

Implications / Conclusions



- Better knowledge of reproductive biology of *O. edulis*
 - Ability to change of sex very quickly (maturation of 2 types of gametes simultaneous)
 - Spawning of an individual does not stimulate all the others
 - High variability in the temporal dynamics of emission of gametes and larvae

- Relative variance in reproductive success in males and females:
 - For most females, spawning events are constituted by the progeny of several males (especially in our hatchery experiment)
 - Most males fertilize several females (hatchery experiment)
 - Effective reproductive period is longer for males than for females (hatchery experiment)



Higher variance in reproductive success in females than in males