Broodstock management and larvi-culture in marine species reared in Polynesia and New Caledonia : genetic and health approach

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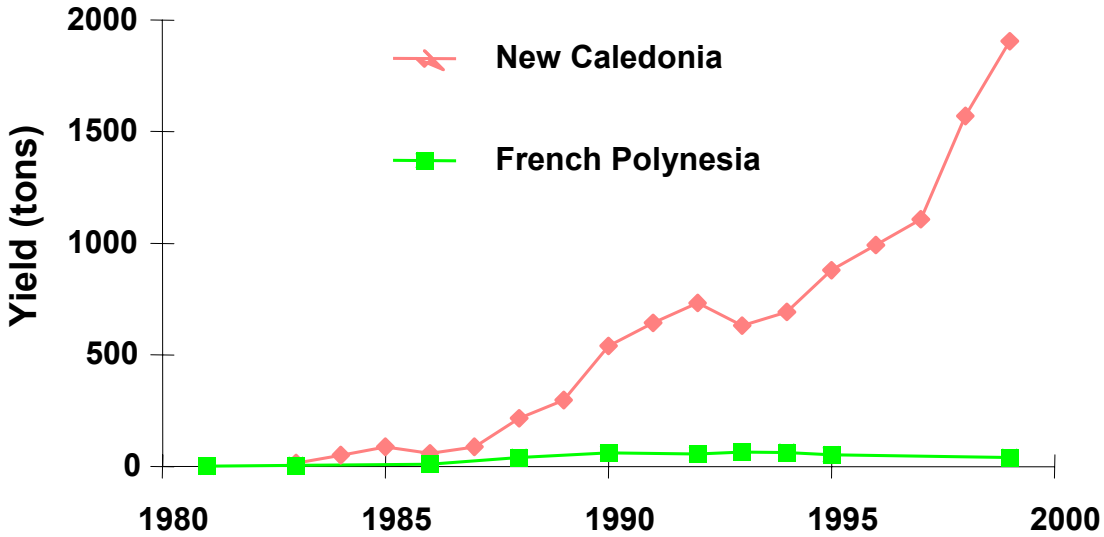
Aquaculture productions

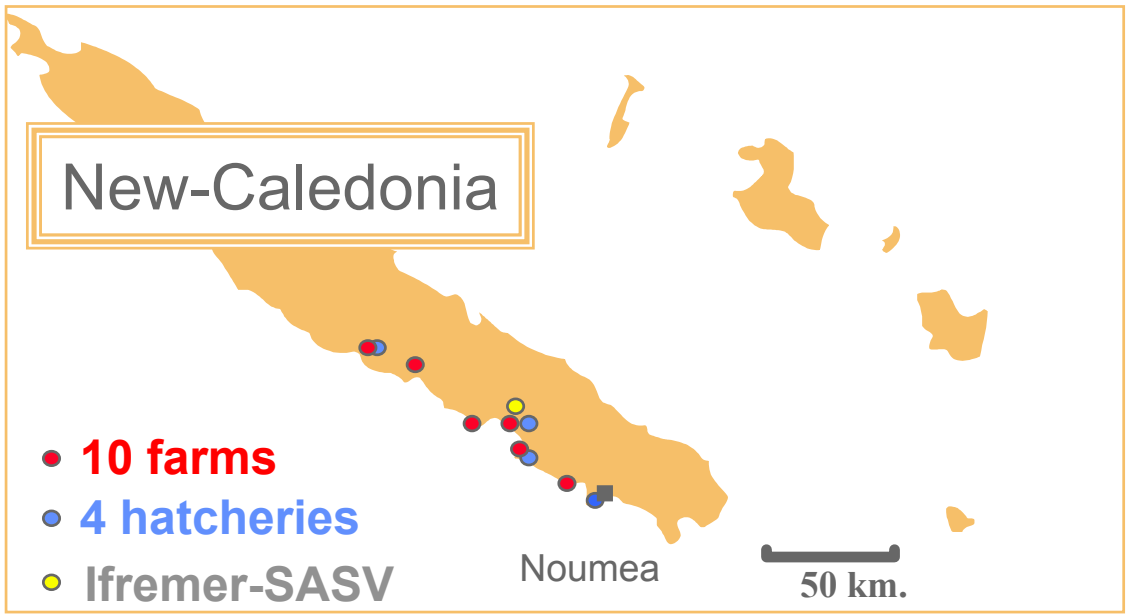
- Shrimp : *Penaeus stylirostris*
  - New Caledonia and French Polynesia
- Pearl oyster : *Pinctada margaritifera*
  - French Polynesia,
- Fish : *Lates calcarifer*
  - French Polynesia

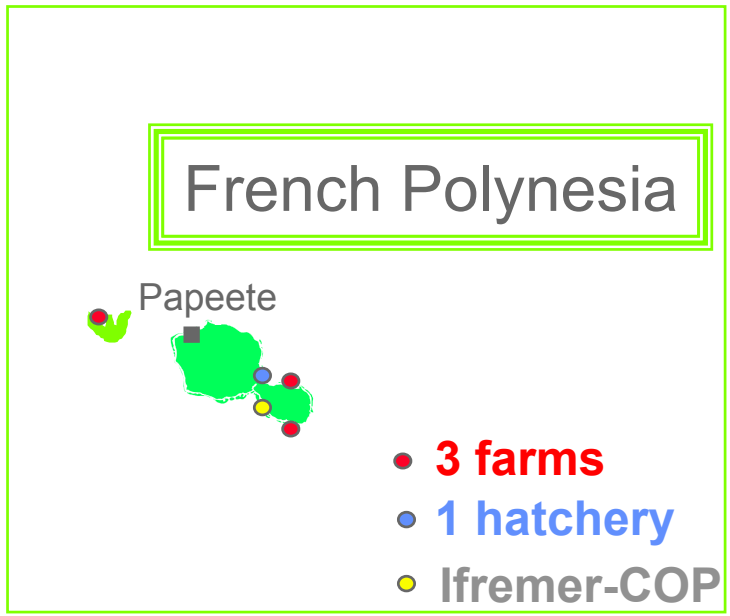
Shrimp : Historic

- 1970-1980
  - Selection of species for their reproductive ability in captivity and growth performances.
- 1980-1990
  - Broodstock maturation : nutritional improvement
  - Larval rearing :
    - replacement of live food by microparticulate diet,
    - water management (biological filter)
  - Sanitary status : genetic resistance to IHHNV (1987)
- 1990-2000 :
  - Starting of a genetic program
    - characterisation and improvement of strains domesticated in Polynesia and Caledonia
    - development of tools : molecular markers and method of gametes conservation

Main French shrimp farming areas







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P. stylirostris shrimp : Broodstock management

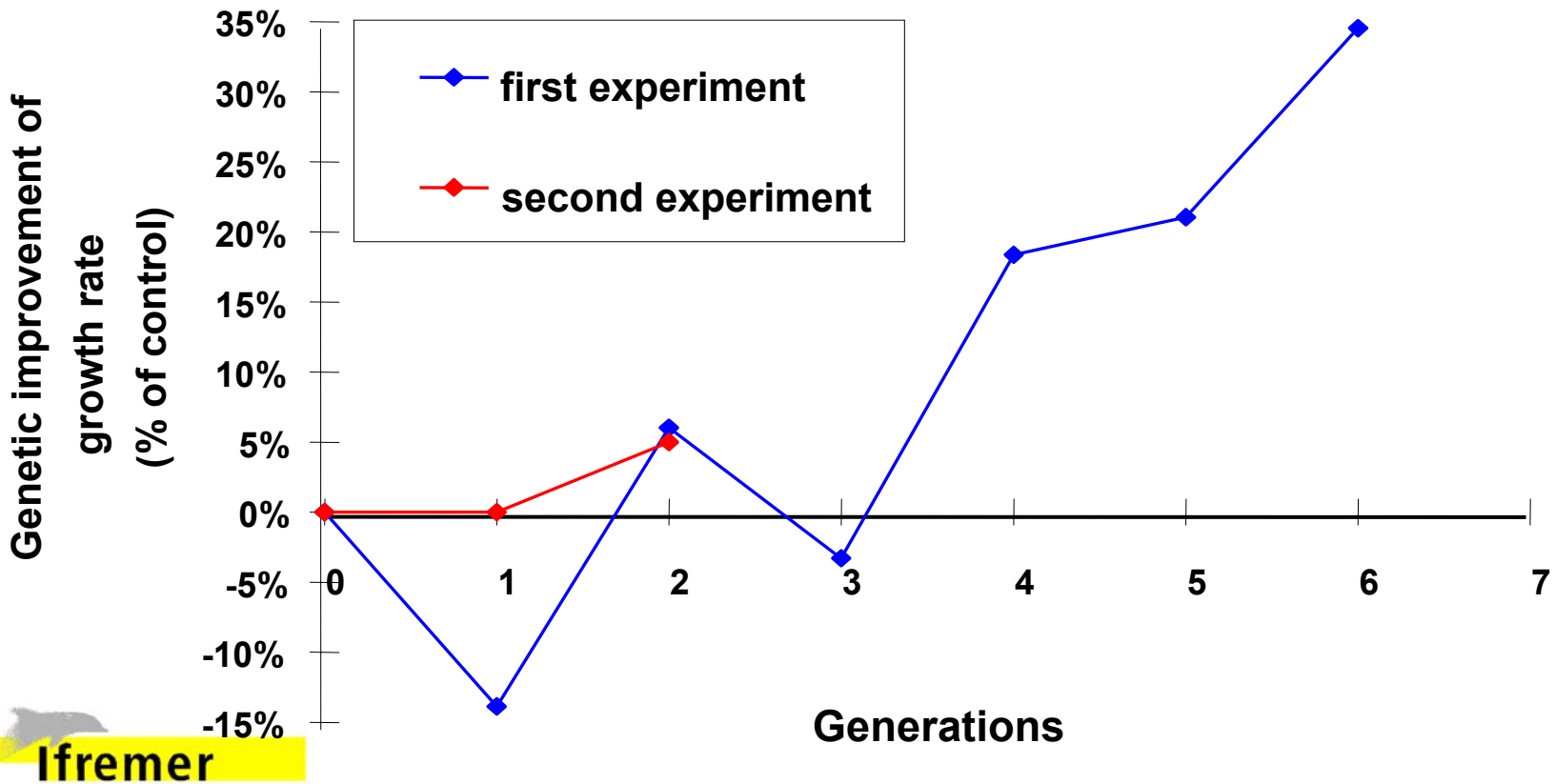
- **Maturation**
  - **unilateral eyestalk ablation in females**
  - **males kept at 25-26°C**
  - **improved feeding**
  - **artificial insemination (2 males X 1 female)**

- **Gamete preservation I**
  - **refrigeration at 4°C in conservation medium at pH 7 containing antibiotic allow to use sperm during 6 days**
  - **transfer from Ecuador to Tahiti (2 to 6 days from collection to mating), 165 wild males X 87 domesticated females (17% success)**

- **Gamete preservation II**
  - **cryo-preservation in liquid nitrogen : *P. vannamei* nauplii obtained in 1991 and 1996, non reproducible probably due to breeder and gamete of insufficient quality, no results obtained with *P. stylirostris***

P. stylirostris shrimp : Genetic improvement

- Resistance to disease
  - spontaneous resistance to IHHNV
- Selection for growth
  - an improvement of 34% of weight at the sixth generation



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P. stylirostris shrimp : Genetic resources

- **Micro-satellite and intronic markers have allowed to characterise the strain and found :**
  - **a positive relation between heterozygocity and growth**
  - **the loss of genetic variability during the domestication process (20 generations) compared to the wild stock from Ecuador**
- **These markers allow to check shrimp pedigree and now to implement a strategy to preserve residual variability**
  - **maintenance of several independent populations**
  - **structuring of each population into 2 sub-populations**

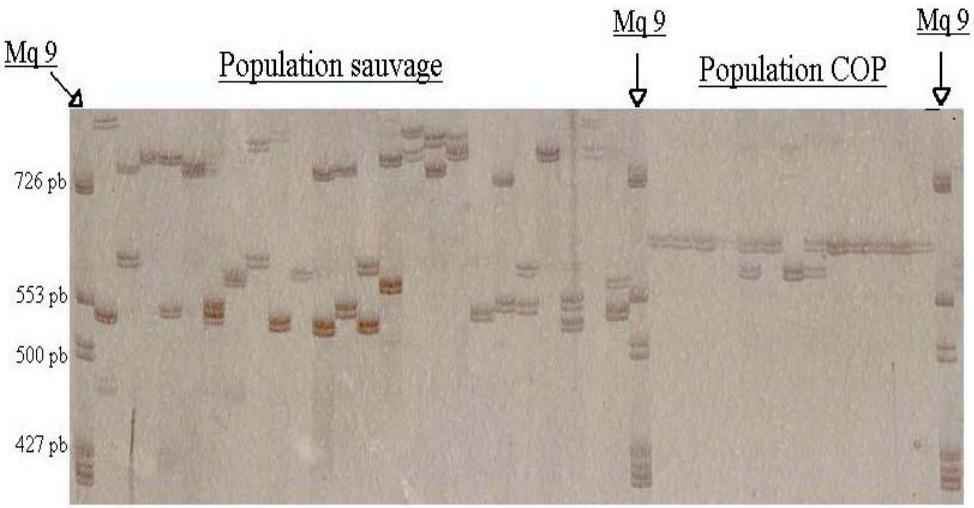
- **Importation of genetic variability**
  - **Using refrigerated spermatophores :15 F1 families were produced in quarantine facility**

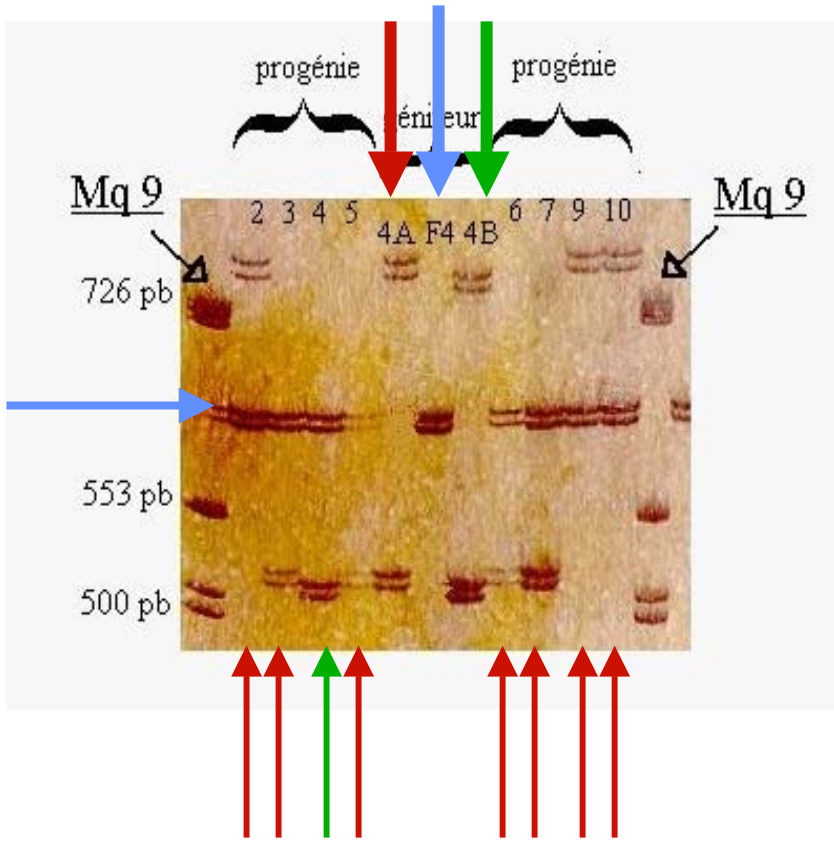
P. stylirostris shrimp : Genetic resources

Checking of pedigrees

Evidence of genetic variability loss during the domestication process (20 generations) compare to the wild stock from Ecuador

P. stylirostris strains	Allelic number / locus	Heterozygocity level
Tahiti	1-3	0.1
New Caledonia		0.2
Wild Ecuador	14-25	0.9





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P. stylirostris shrimp : Sanitary management

Sanitary status of breeders

- Except the presence of IHHNV, Polynesia and Caledonia territories keep a favourable sanitary status probably related to the use of closed broodstock
- perspectives
  - Health state management (SPF)
  - Immune criteria for health status determination in relation with environmental, nutritional and genetic state

Control of disease in larval rearing

- antibiotherapy : erythromycine, oxytetracycline (furazolidone replacement)
- alternative : probiotic (*Pseudoalteromonas piscicida*)

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Pearl oyster industry in French Polynesia

Historic

- First episode : mother-of-pearl era in XIXth century

- mother-of-pearl was exploited to supply the button industry
- appearance of synthetic button is the death of this industry in the middle of XXth century

- Second episode : fast-growing of pearl oyster industry

- 1960 : research on pearl production with the help of Japanese experts especially on spat collection and graft trials

- 1980 : start of pearl oyster industry

- 1999 : 6 metric tons of pearl, 200 million US$, 1076 farms-7000 employment

- 2000 : difficulties due to overproduction,

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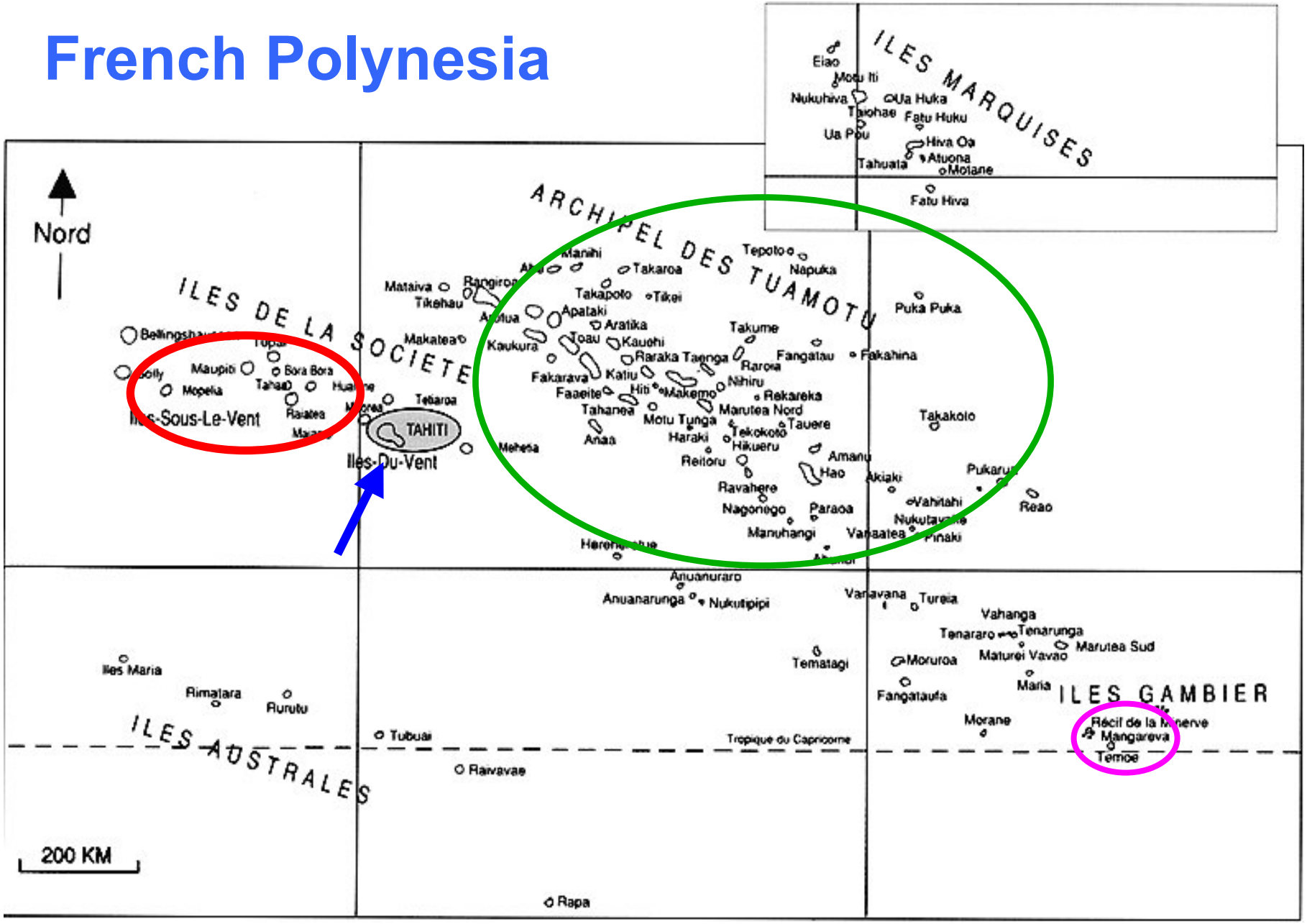
French Polynesia



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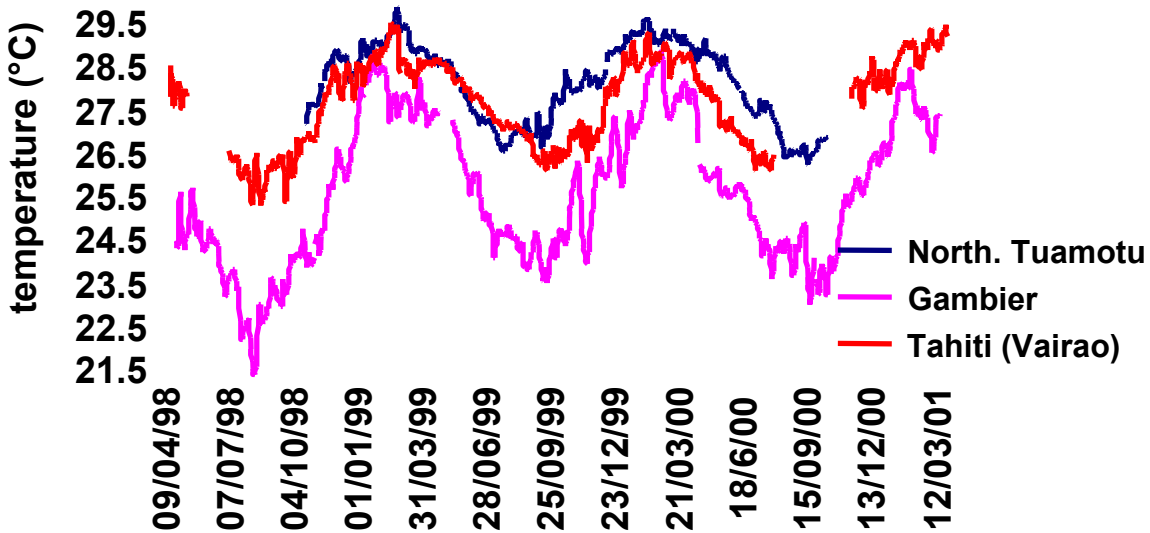
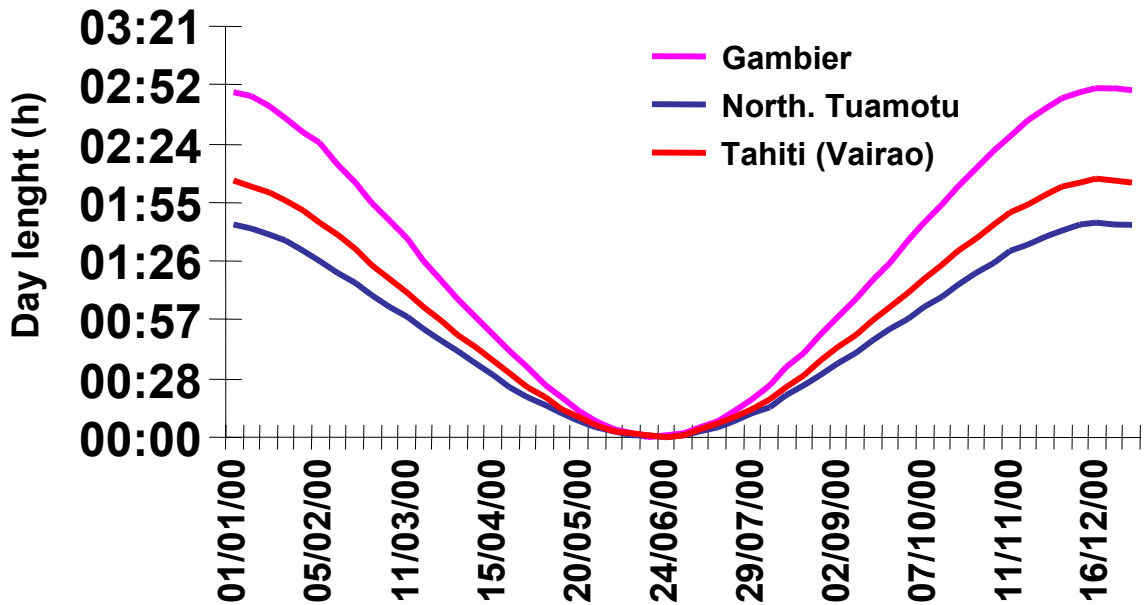
Annual environmental change in French Polynesia lagoons according to the latitude





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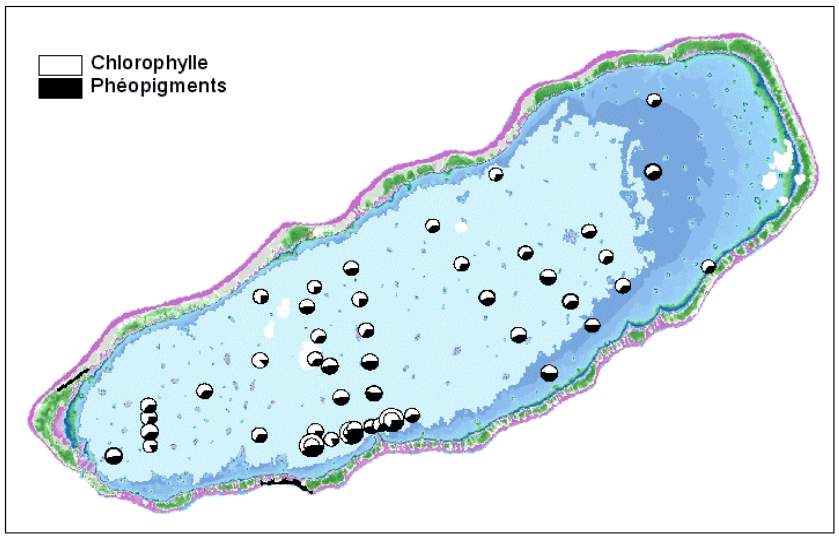
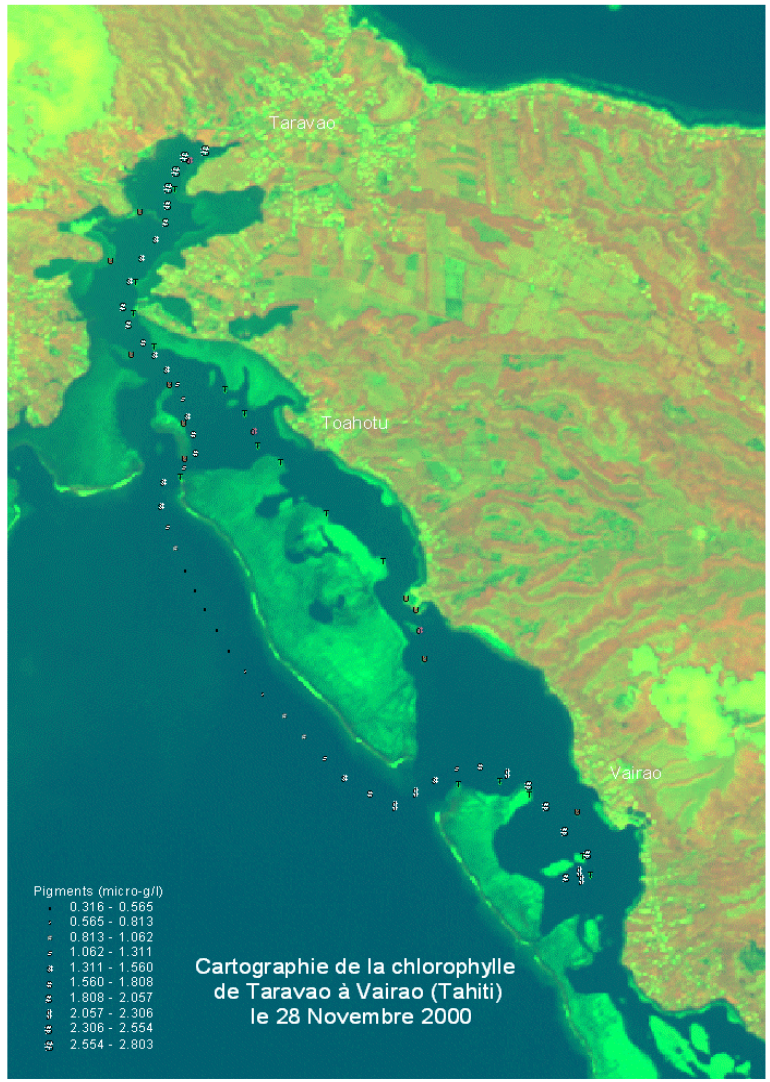
Hydrobiological characterisation of lagoons

	Organic matter ($mg.L^{-1}$)	Chlorophyll *a* ($\mu g.L^{-1}$)
Makemo	0.35	0.21
Manihi	0.50	0.46
Takaroa	0.48	0.30
Takapoto	0.35	0.40
Raiatea	0.42	0.54
Vairao	0.50	0.30

Distribution spatiale des pigments à Manihi
Mission du 25 au 28 Juillet 2000





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Pearl oyster : sex and reproductive cycle

Sex and reproductive cycle are evaluated with

Spawning (induced with thermal shock)

biopsy

histology

gonadal weight changes

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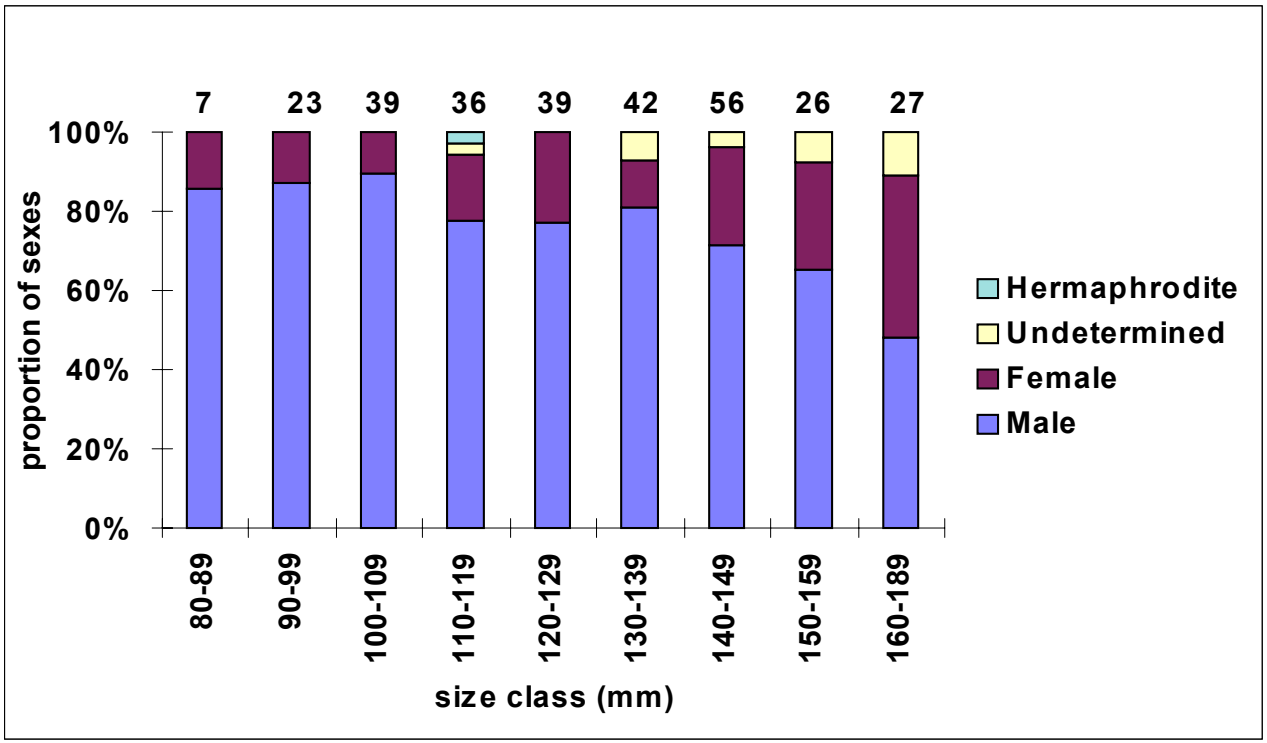
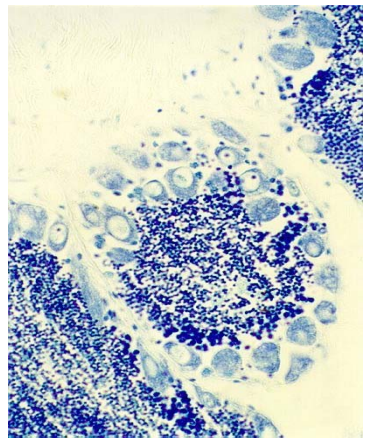
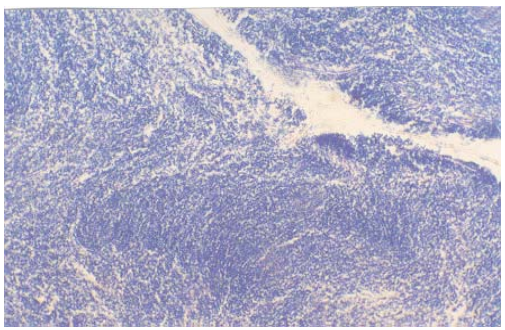
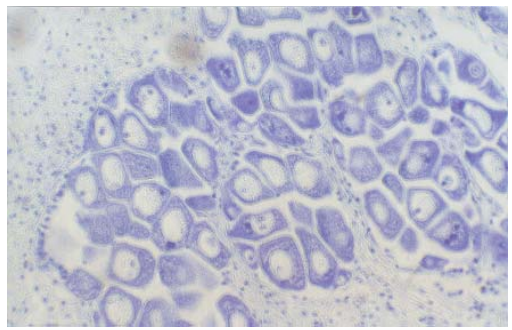
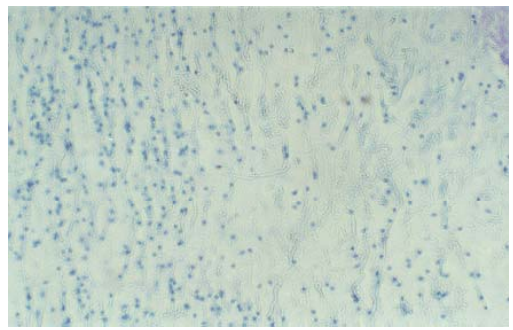
Sex ratio and gametogenesis











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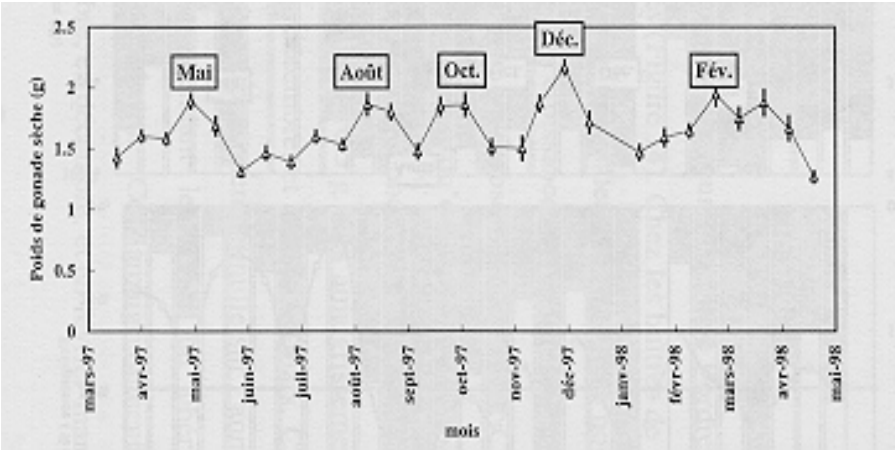
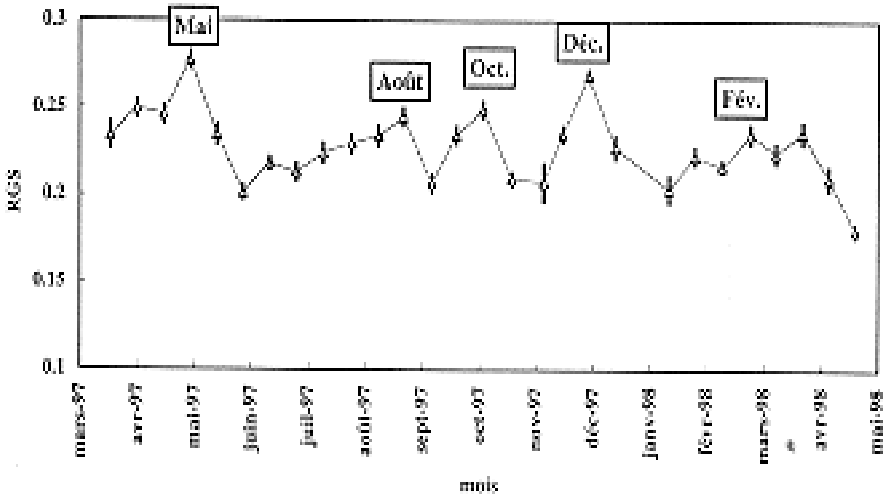
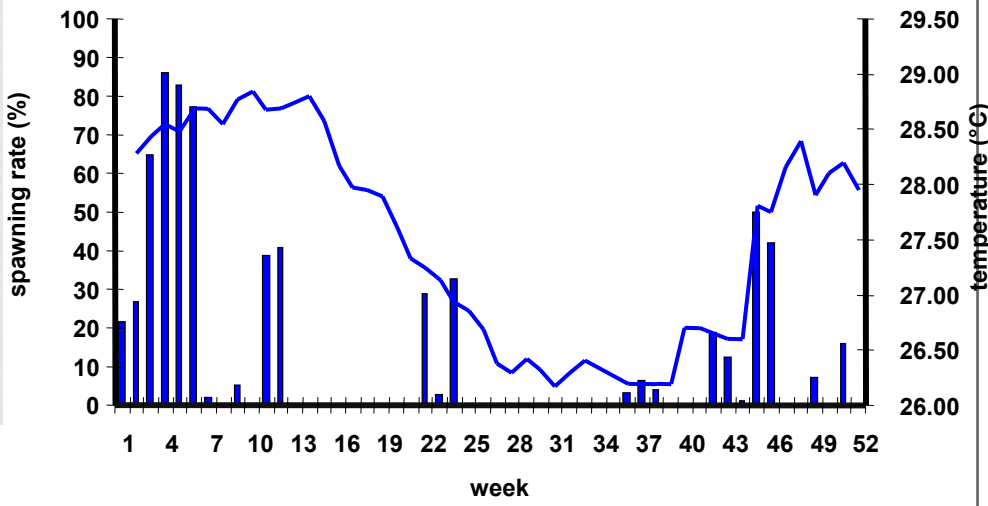
Gametogenesis *in situ*

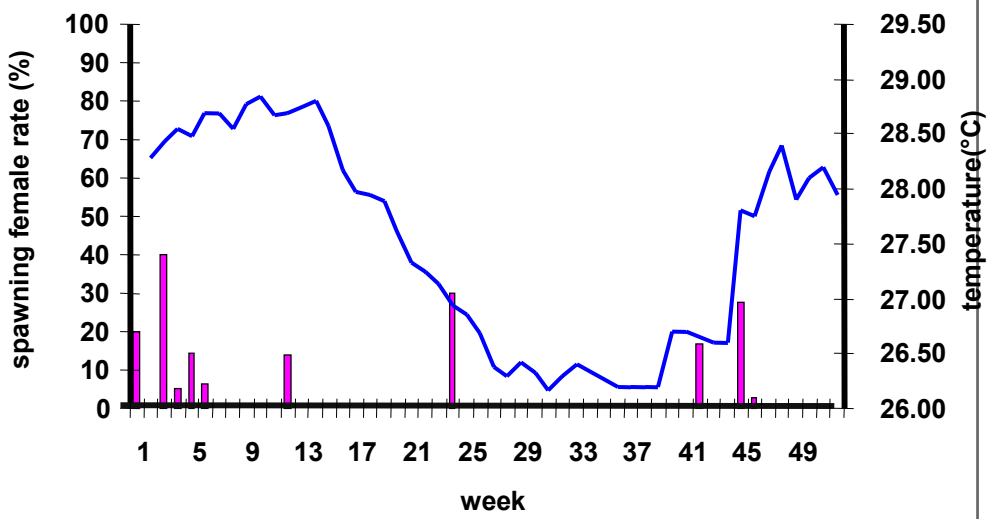
Gonadal weight and gonadal index (northern Tuamotu) (Pouvreau, 1999)





Spawning rate related to the temperature change (Vairao)





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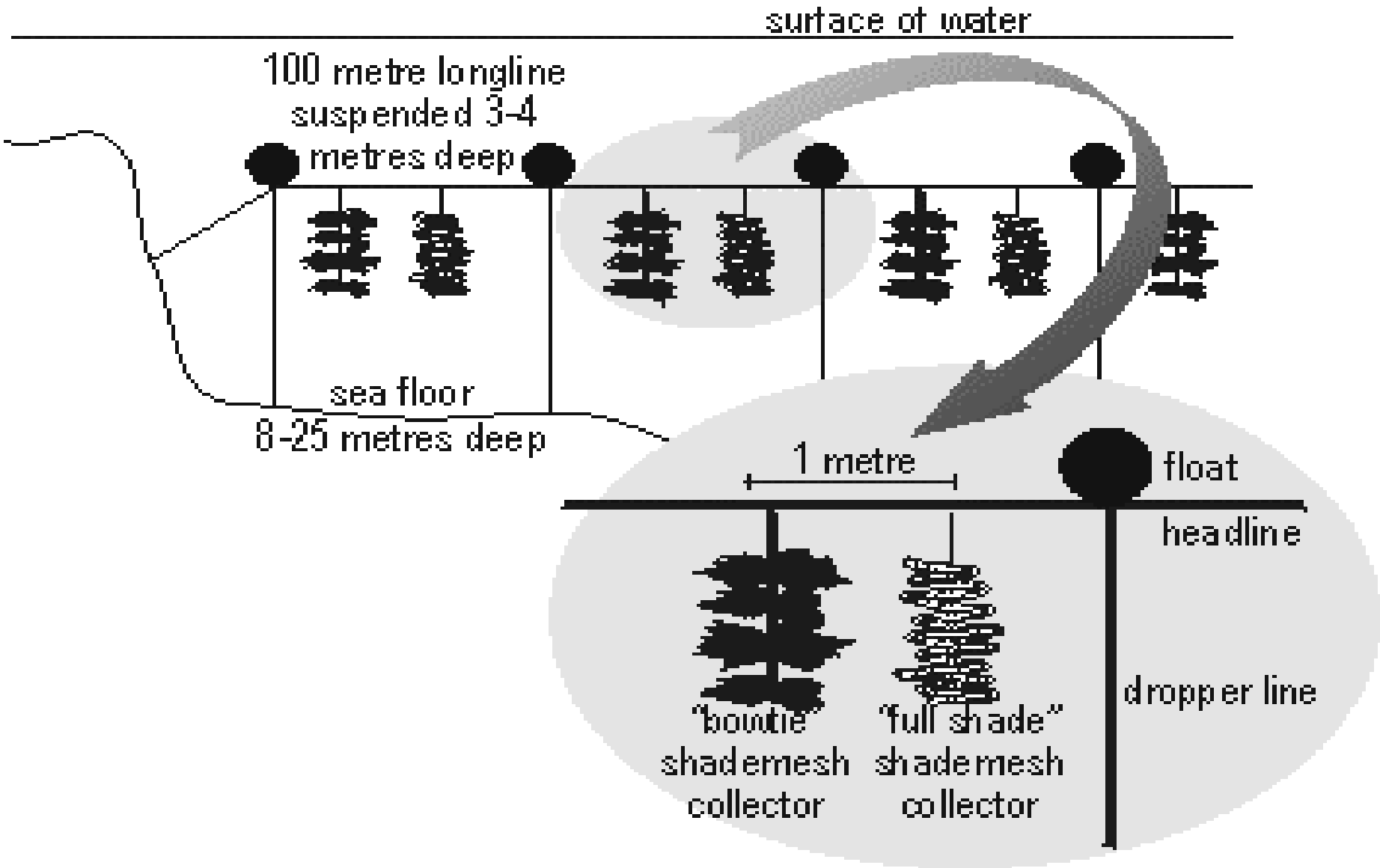
Spat collection



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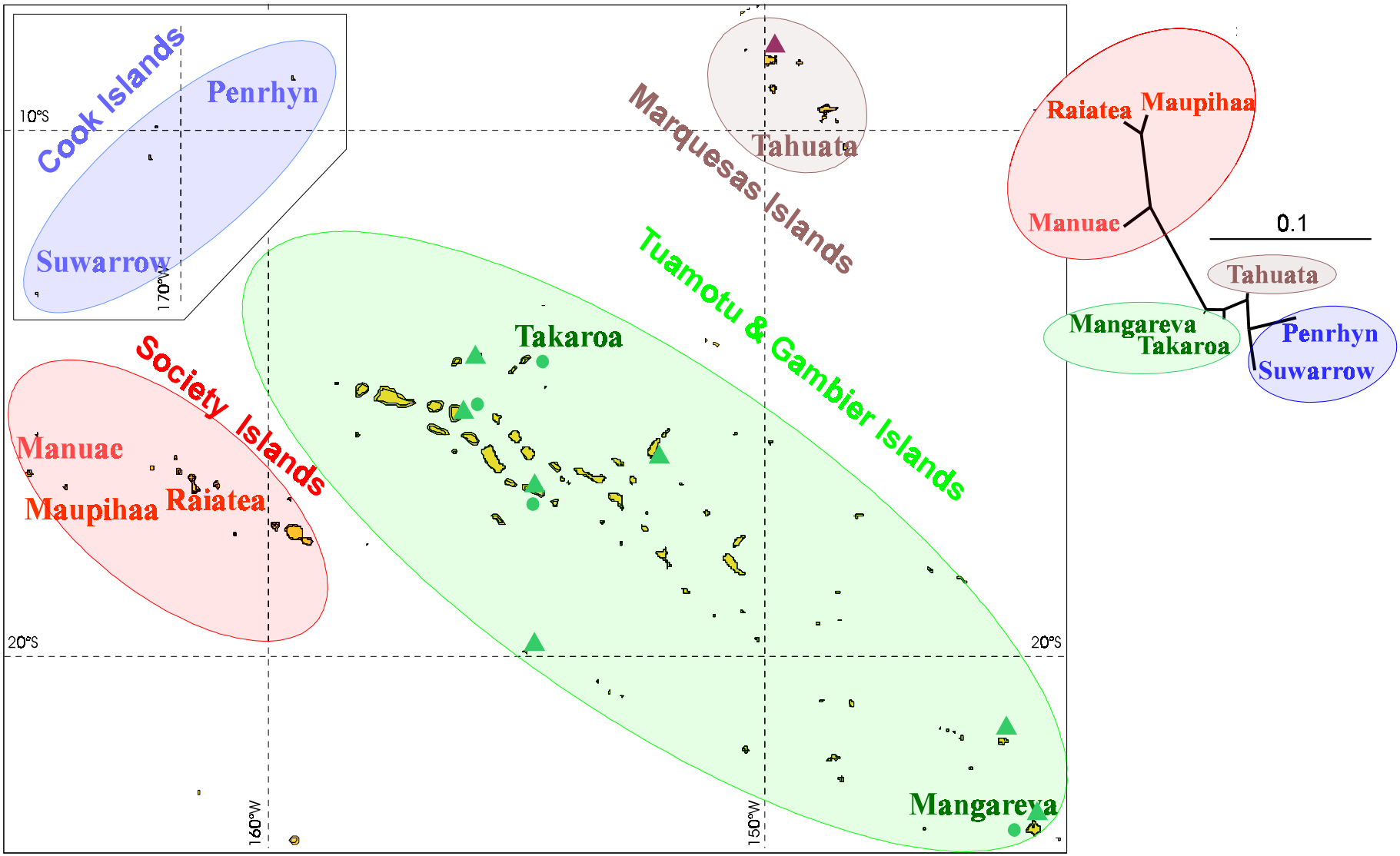
Genetic resources of *P. margaritifera*



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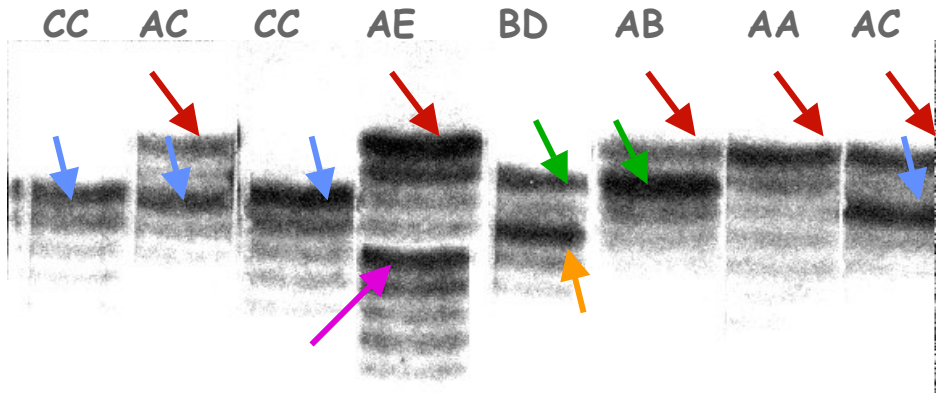
Genetic resources of *P. margaritifera*

Genetic markers

DALP Method (Direct Amplification of Length Polymorphism)

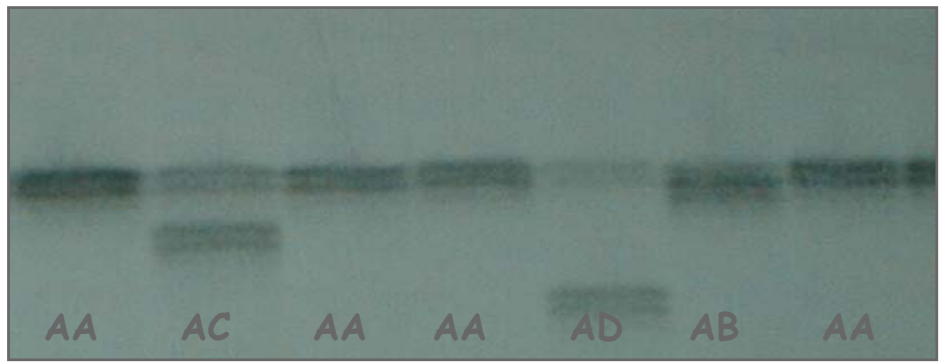
3 markers :
- pin 1 : 6 alleles
- pin 2 : 5 alleles : A, B, C, D, E
- pin 3 : 5 alleles



EPIC Method (Exon Primers Intron Crossing)

use of primers defined in exonic sequence (amylase, aldolase) Amplification of intronic sequence

2 markers:
- U4 : 6 alleles
- aldo : 5 alleles



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Development of hatchery

Objective

Supply spat when the collecting is insufficient

Produce genetically improved spat
(triploïd, selected for growth, pearl colour)

Supply spat to restock some atolls by respecting
the genetic structure of the natural populations

Method

Broodstock conditioning

- synchronism of breeders to control crossbreeding
- control of sex ratio (increase female percentage)
- improve larval quality with breeder feeding

Larval rearing

- feeding
- water management
- disease control

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Sanitary rules
and zoo-sanitary surveillance network

- 1985 : mass mortality (60% of oysters)
- Context still favourable to the emergence of disease (uncontrolled transfert)
- outbreak in Japan (Akoya virus)

- Zoo-sanitary regulations
  - banning of importation of *P. margaritifera* from other areas and new pearl oyster species
  - surveillance network to prevent outbreak of disease

Conclusion

The industrial rearing of marine species in French Polynesia and New Caledonia are the subject of genetic and sanitary follow-up

Preservation of the genetic resources

Genetic improvement and creation of selected lines

Saving of the sanitary state with the enforcement of zoo-sanitary regulations

Suppression of antibiotic use

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