

Gamete Biology: perspectives for tuna aquaculture

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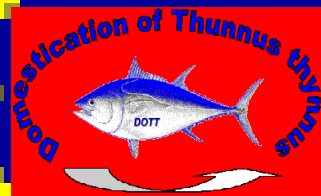
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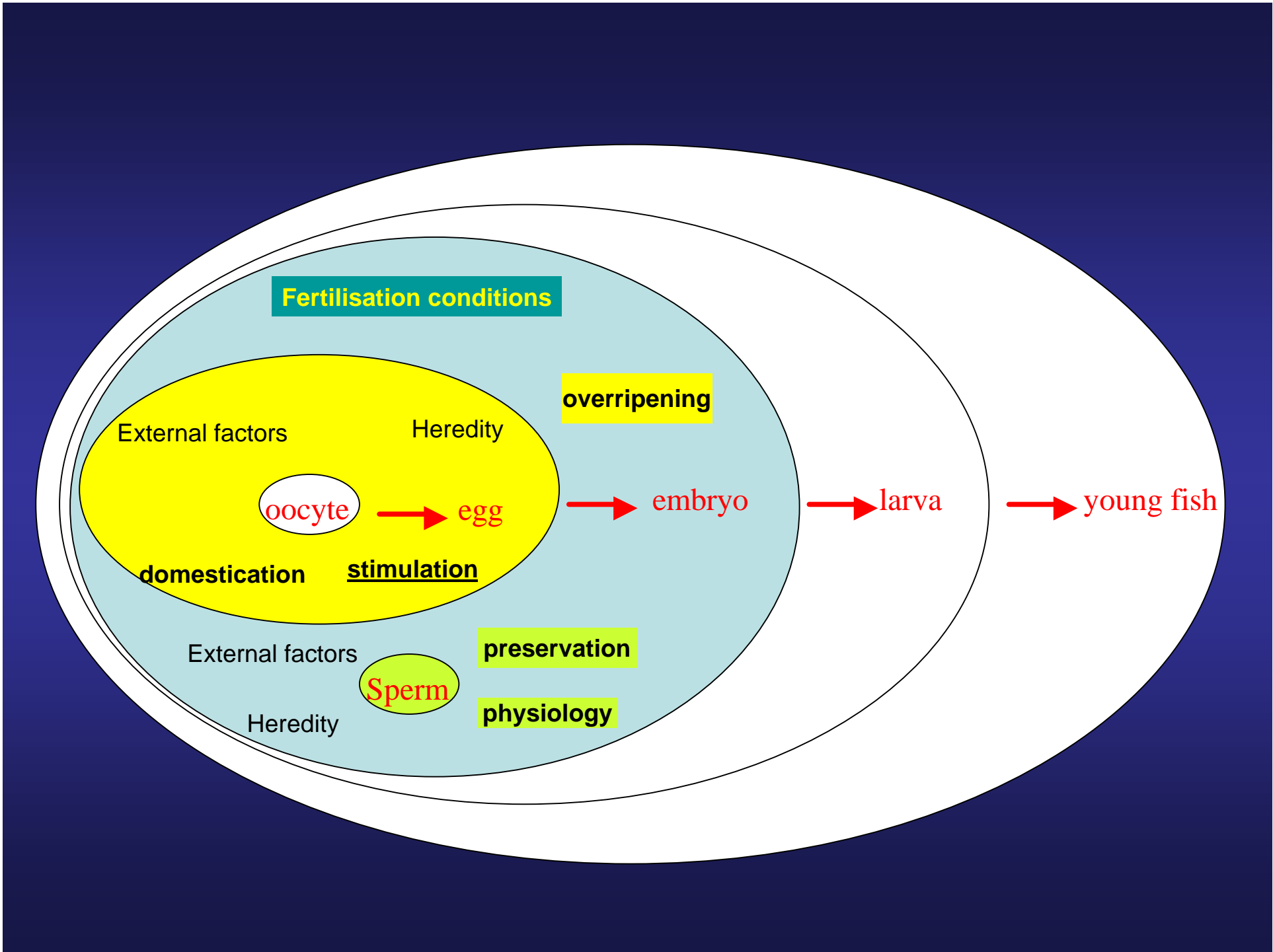
Ifremer



 **Kinki
University**



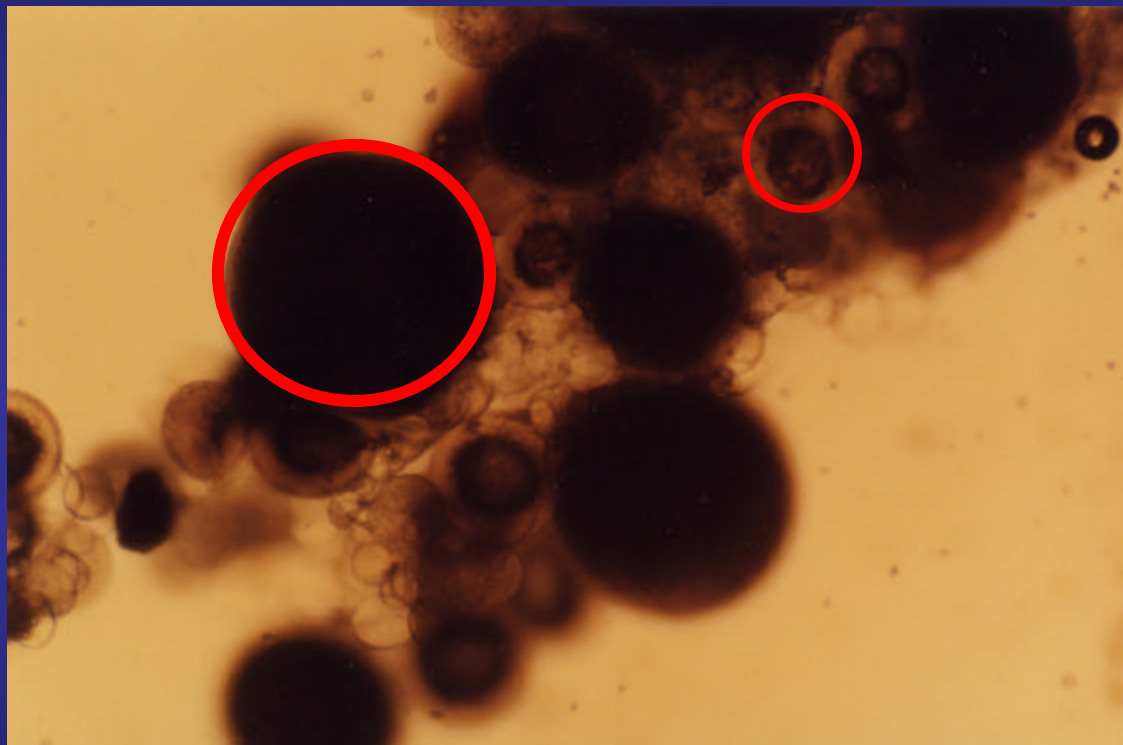
*Center of Aquaculture Science and Technology
for Bluefin Tuna and Other Cultivated Fish*
21st Century COE Program



oocytes (biopsy)

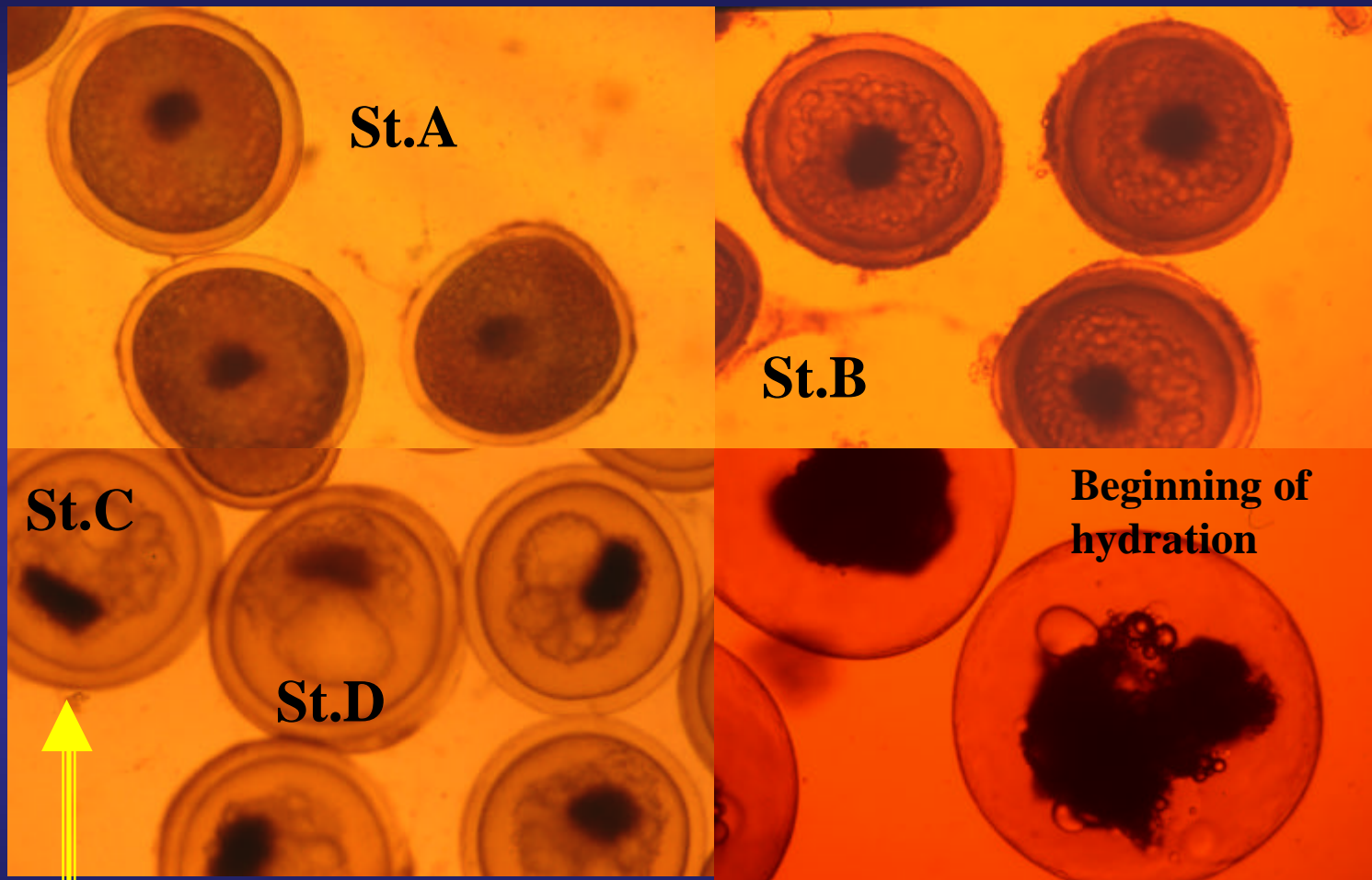
Vitellogenesis

250 μ m



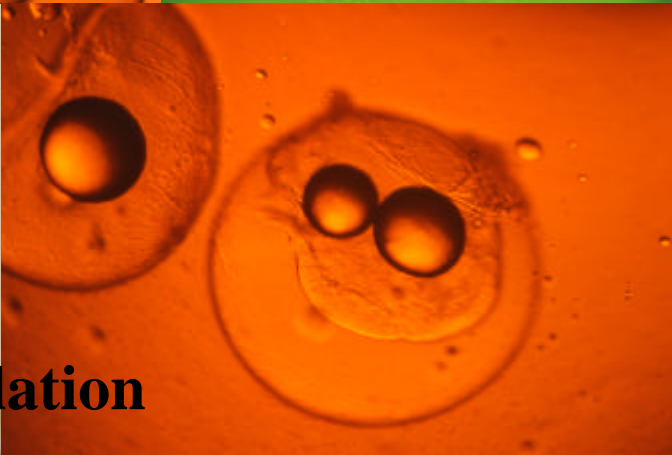
800 μ m

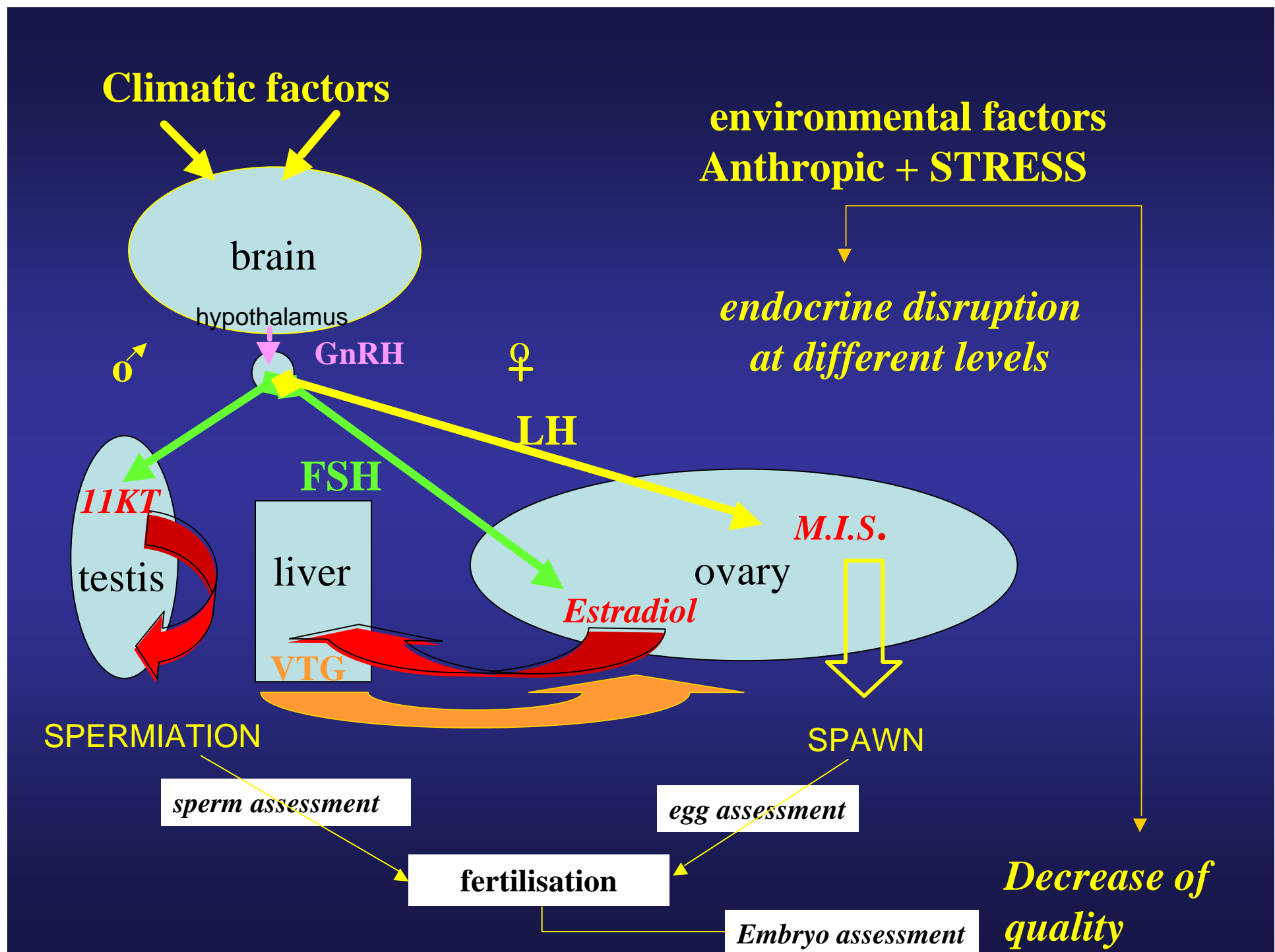
Postvitellogenesis (after clearing)



Best stage for hormonal stimulation of spawn

Hydration-ovulation

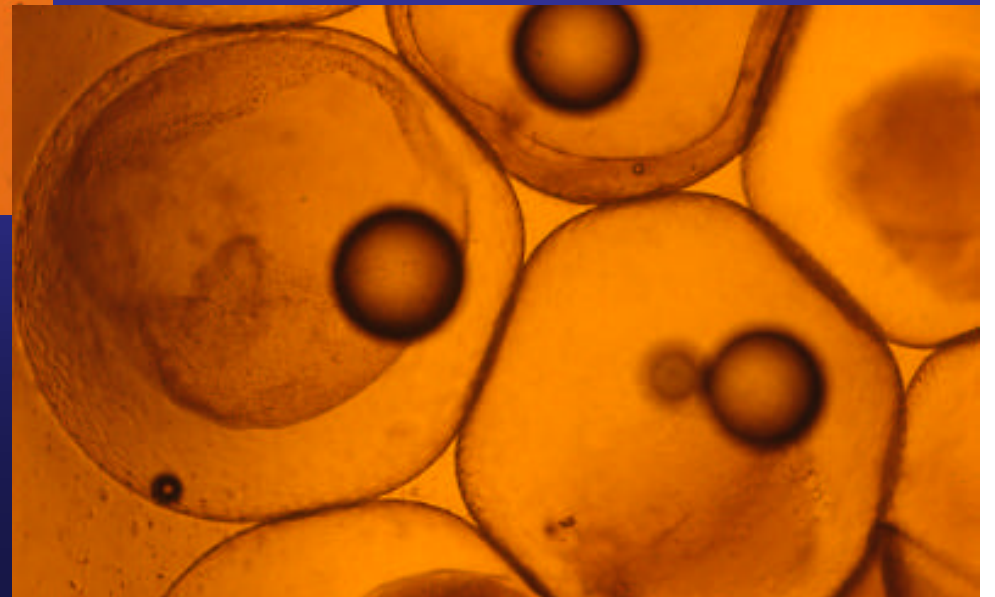
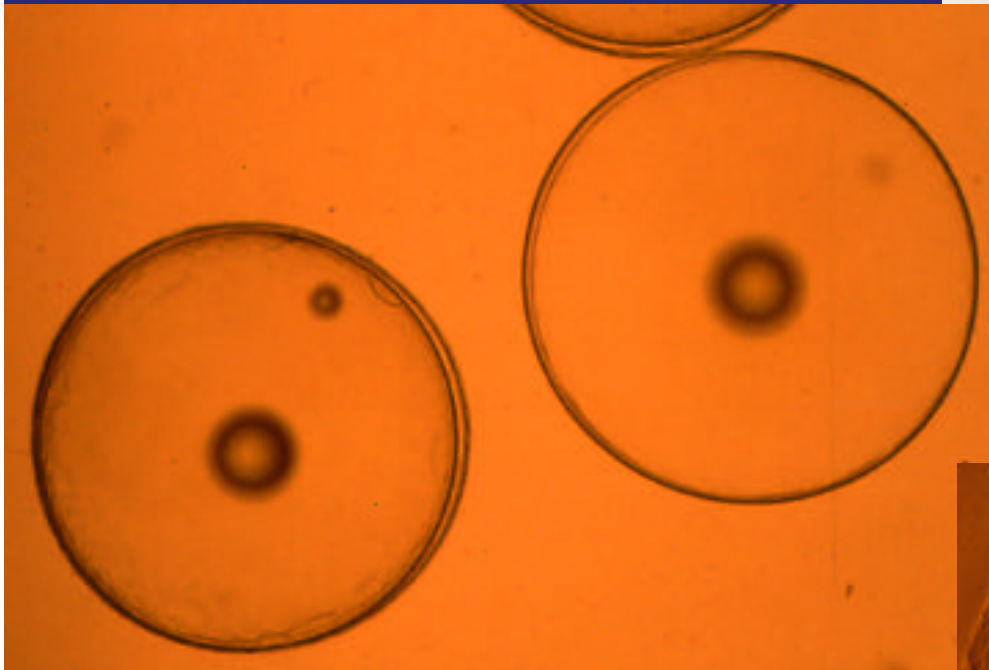




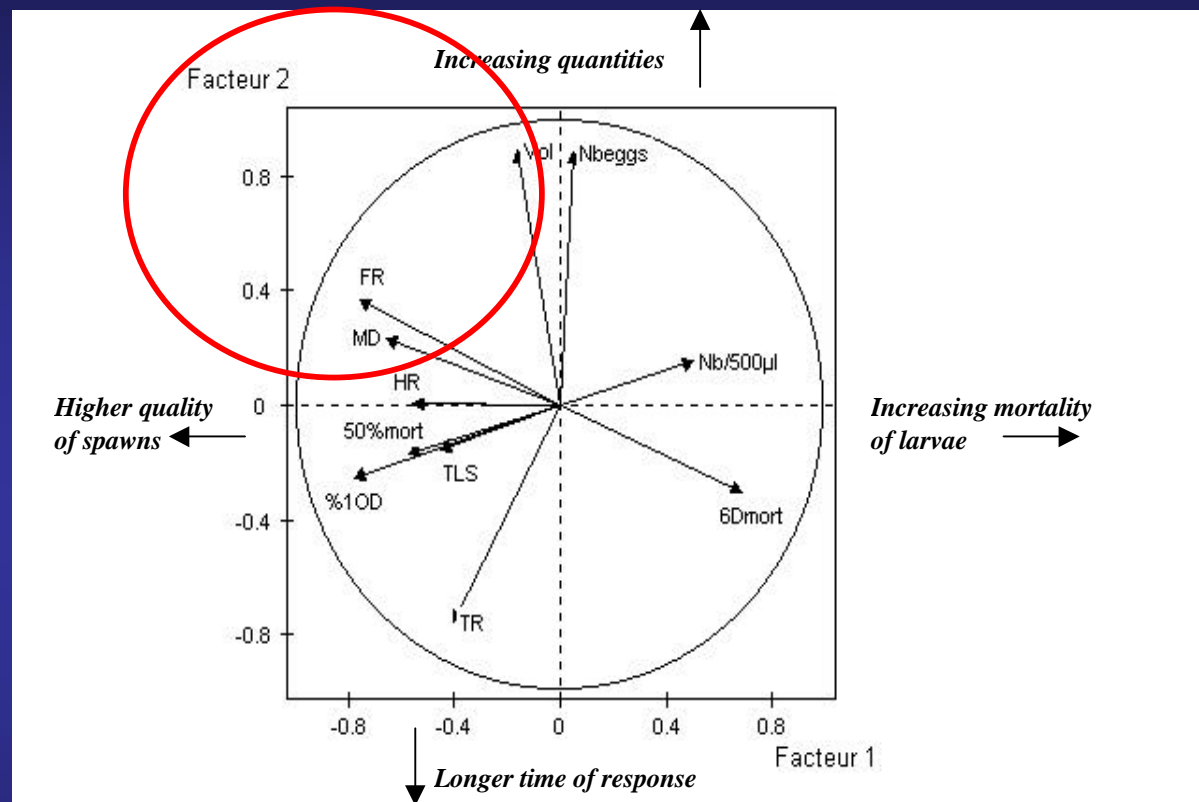
Egg collection



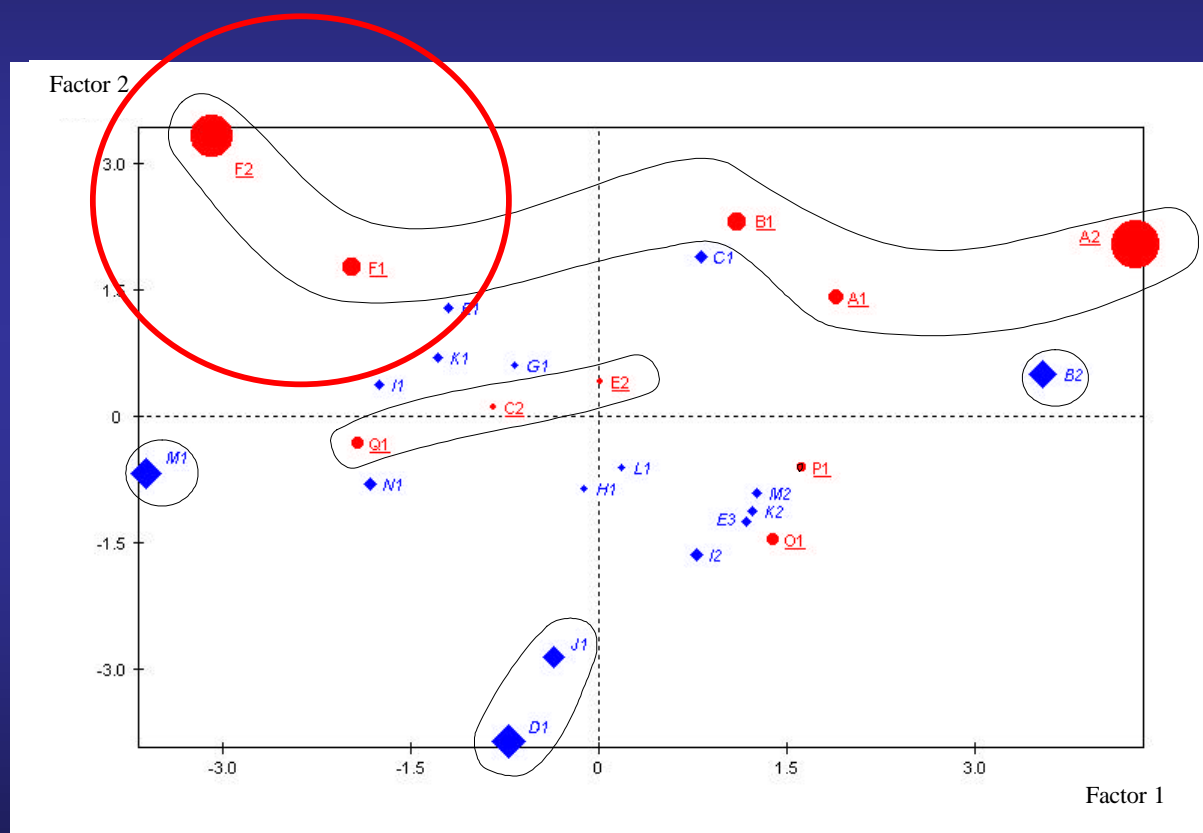
Egg quality?



Heterologous Hormonal Supply

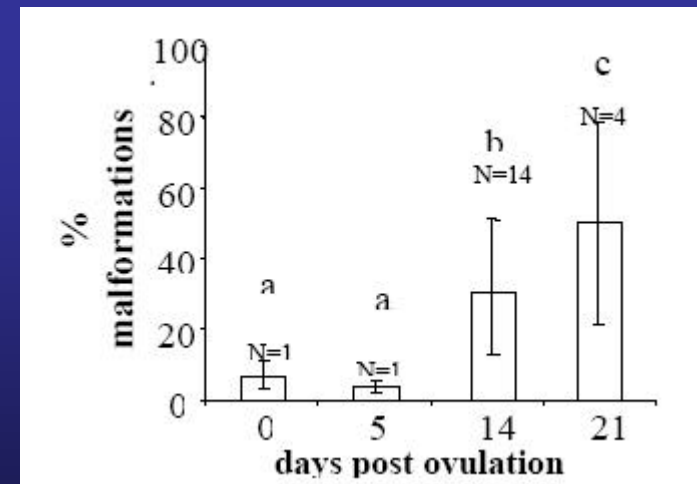
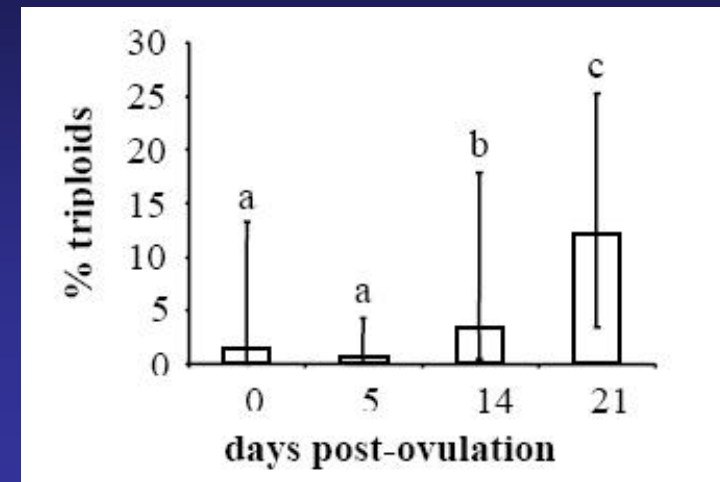
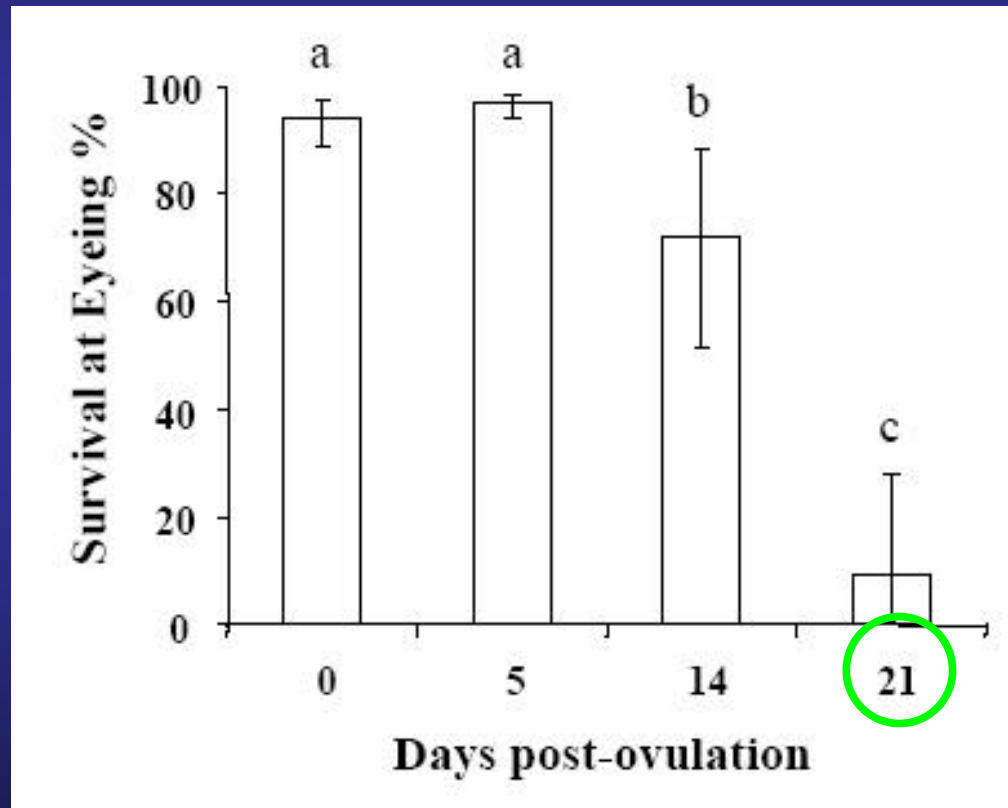


Dose effect : $20 \mu\text{g LHRH}$ (◊) vs $50 \mu\text{g.Kg}^{-1}$ (●)



Stage at stimulation: B (◆) vs C (●)

Overripening in rainbow trout



Aegerter 2004

Sperm collection

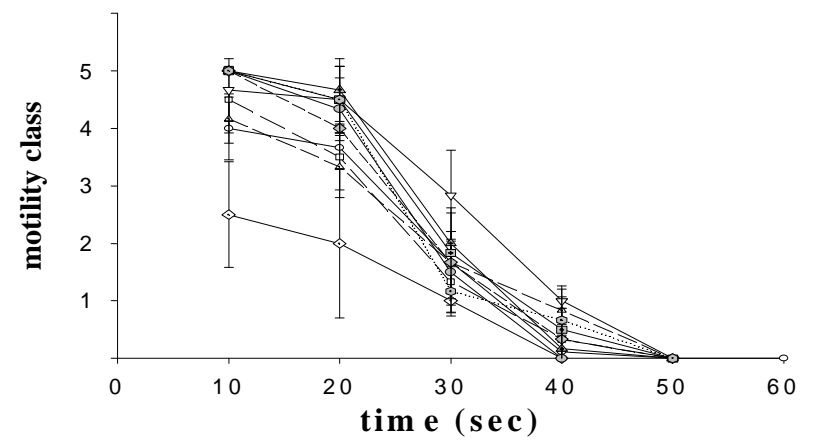


Sperm characterisation

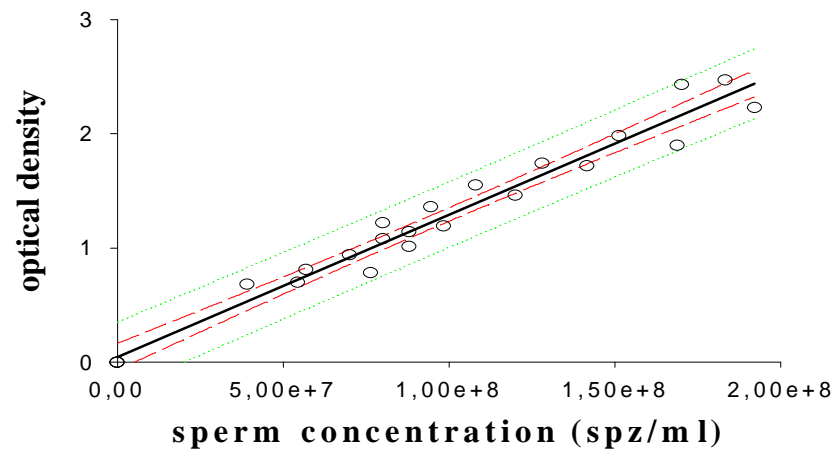
Volume



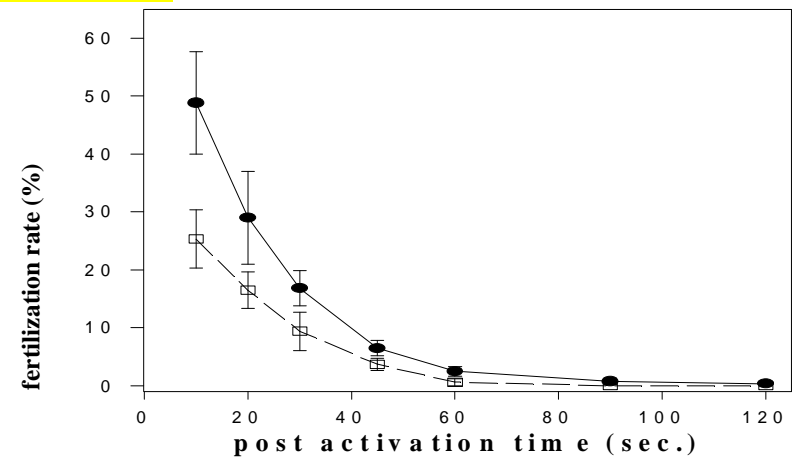
Motility



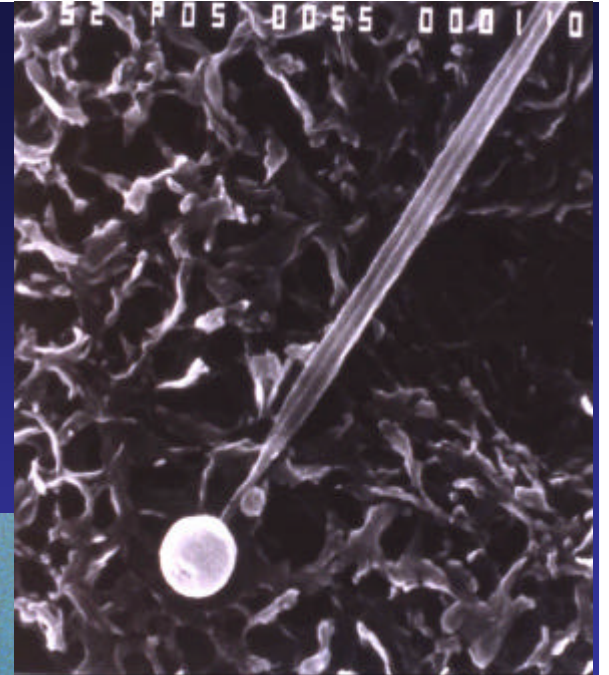
Concentration



Fertility



fertilisation

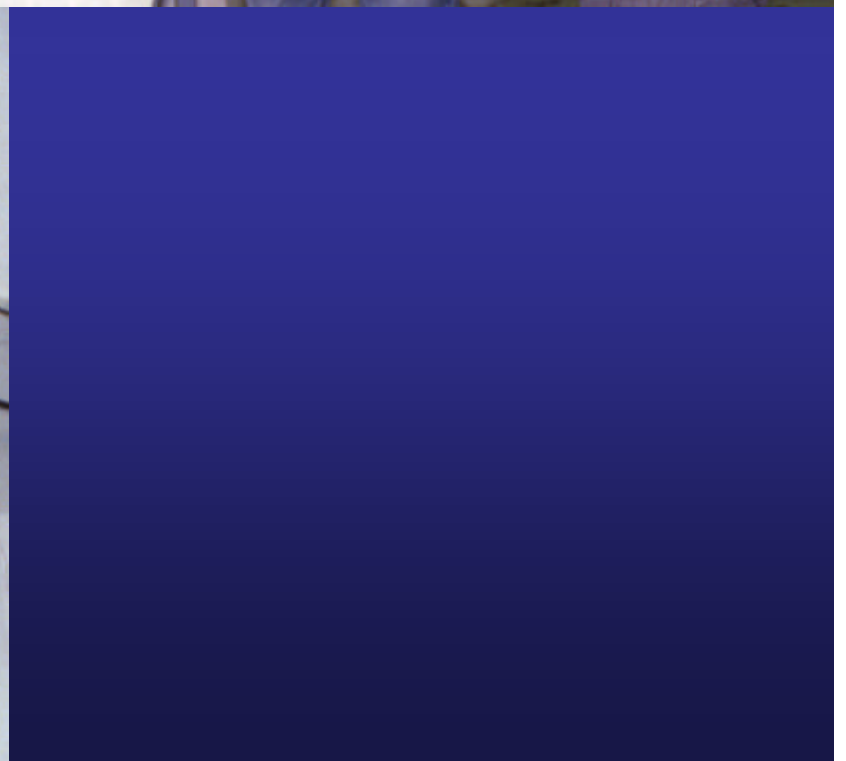


insemination

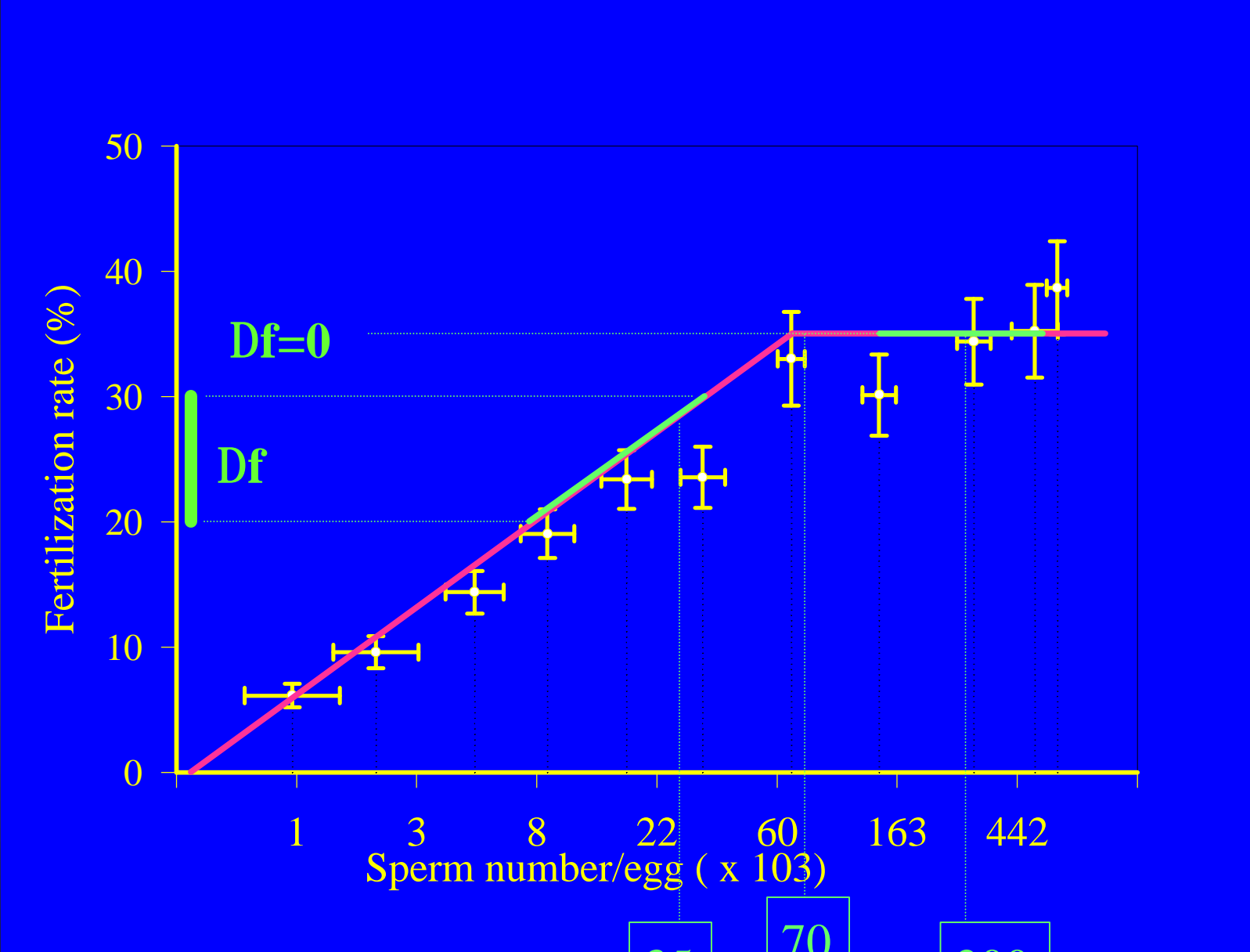
1 vol egg + 0.05 vol sperm

+

0.5 vol sw



Number of sperm cell per egg



experimental

35

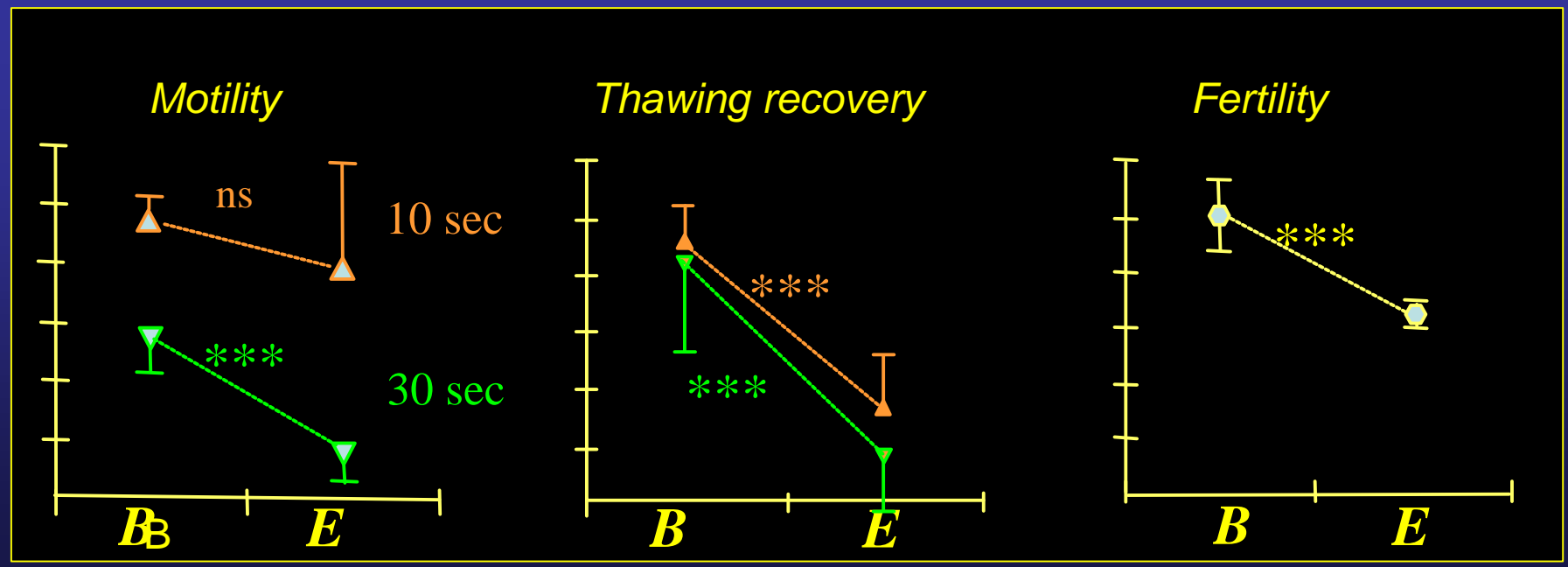
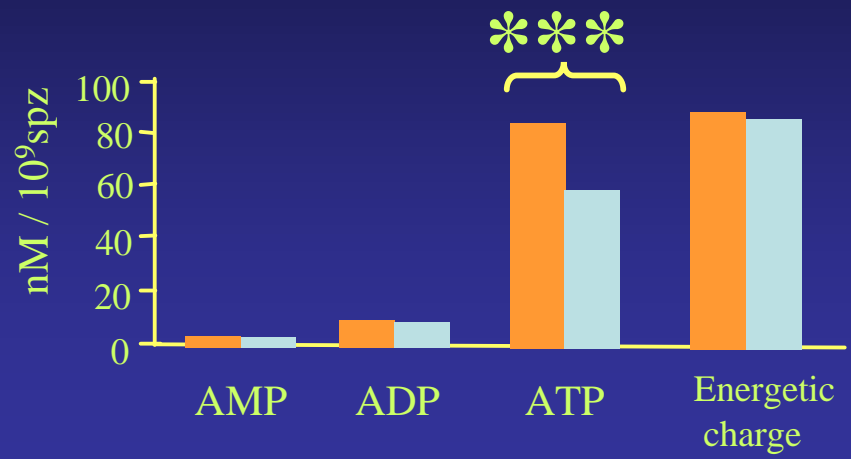
optimised

70

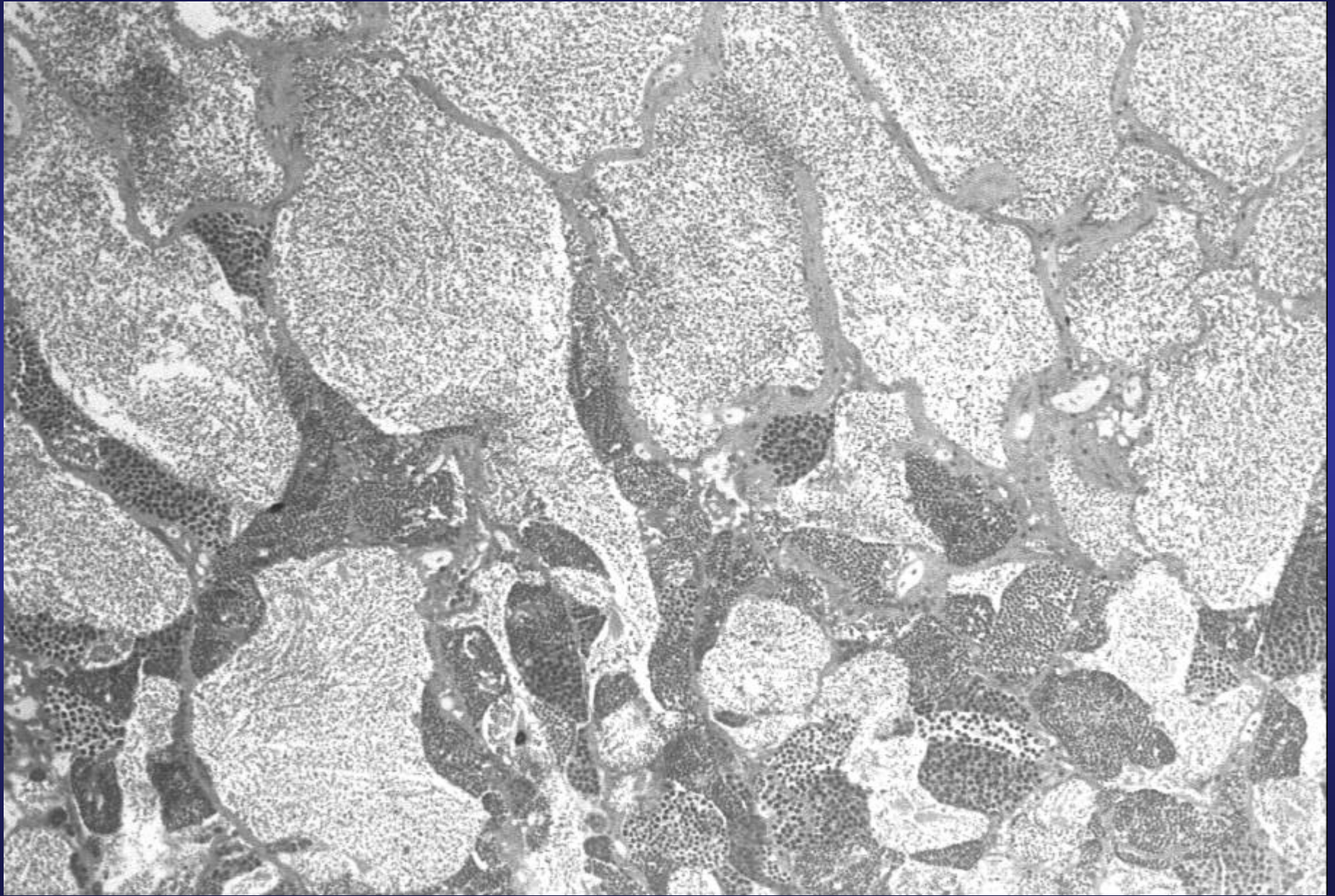
secured

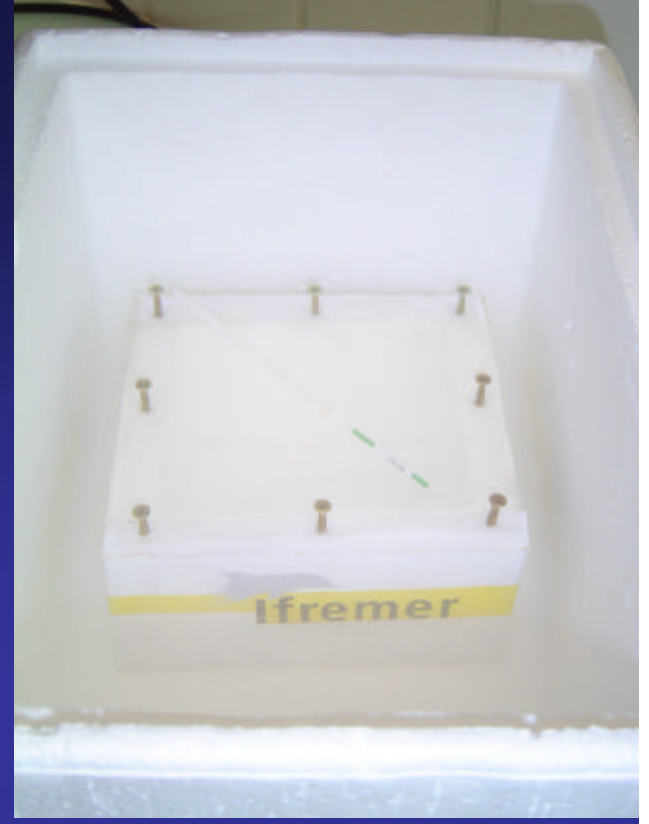
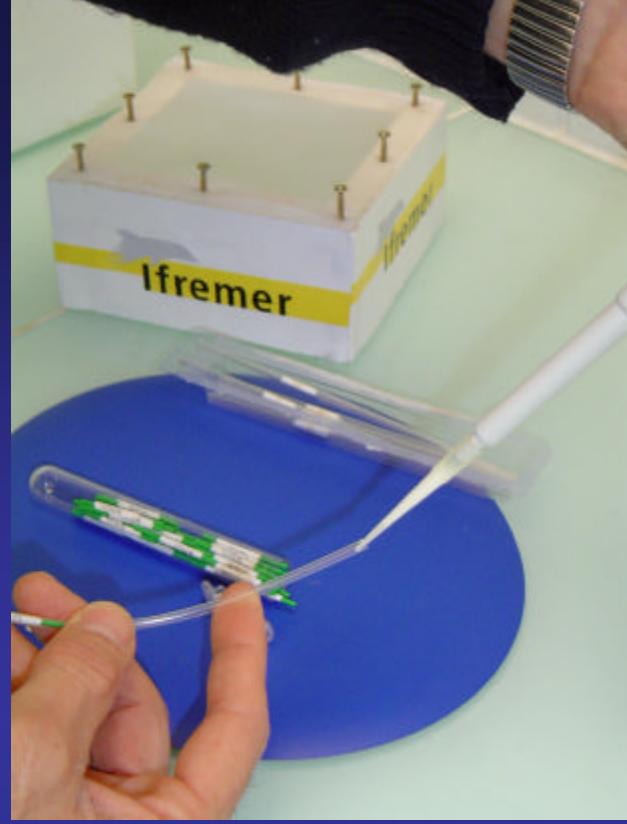
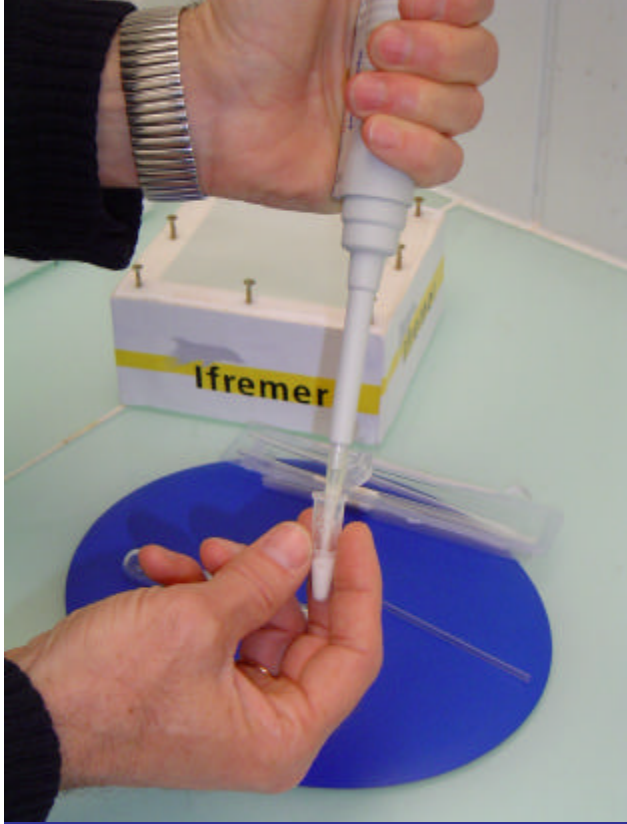
200

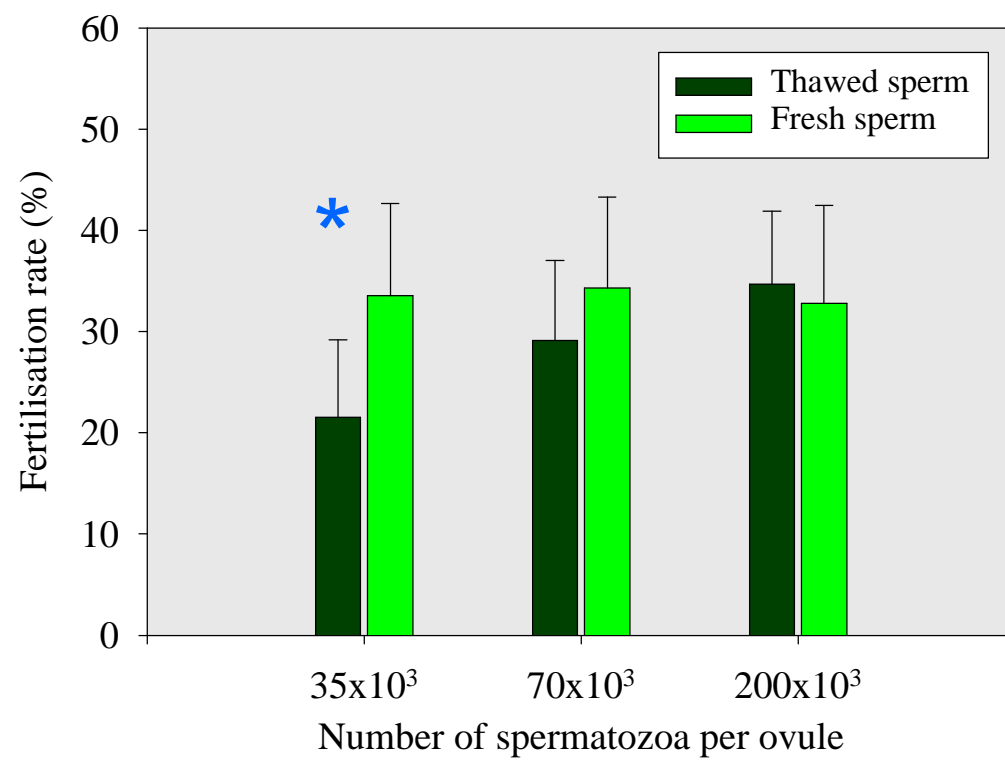
Sperm ageing



Male gametogenesis







	<i>mâle1</i>	<i>mâle2</i>	<i>mâle3</i>	<i>mâle4</i>	<i>mâle5</i>	<i>mâle6</i>
35*103spz/w	* p<0,05	**p<0,01	* p<0,05	p=0,21	p=0,15	p=0,74
70*103spz/w	* p<0,05	p=0,25	p=0,19	p=0,82	p=0,47	p=0,63
200*103spz/w	p=0,4	p=0,25	p=0,45	p=0,34	p=0,11	p=0,16

	FRESH			FROZEN/THAWED		
	MALE C	MALE D	MALE E	MALE C	MALE D	MALE E
Fertilisation rate (%)	71.3	71.6	73.3	66.3	67.0	46.0
Hatching rate (%)	84.1	79.5	81.8	76.8	67.1	64.5
Loss compared to fresh (%)				15.0	21.0	50.5
Number of larvae from 120,000 eggs	72000	68400	72000	61200	54000	35700
Total number of larvae from 360000 eggs	212400			150900		

What about reproduction of captive tuna in Europe?



ReproDOTT Workpackage 9



- ✓ Gamete description
- ✓ Gamete Quality assessment
- ✓ *In vitro* ovulation
- Set up of artificial insemination

3 Captive males (Spain 2003)

Only one male with small amount of testicular sperm

low GSI

Sperm:

- Very viscous
- Low concentration: $1.97 \cdot 10^{10}$ spz ml⁻¹
- Motility: 4 min after activation
- Cryopreservation: OK (same motility at thawing as fresh)

6 Wild males in Italy, 1980
(Doi et al, 1982)

Spermiating without any pressure

GSI ?

Sperm:

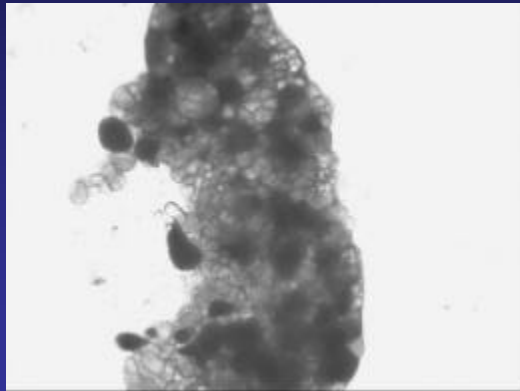
- Very viscous
- High concentration: $4.96 \cdot 10^{10}$ spz ml⁻¹
- Motility: 14 min after activation
- Short term storage : 3 days OK
- Cryopreservation: OK (good motility at thawing)

- **Short captivity allows spermatogenesis but prevents spermiation in our cage conditions**
- **Long captivity allows spermatogenesis and spermiation in our cage conditions**

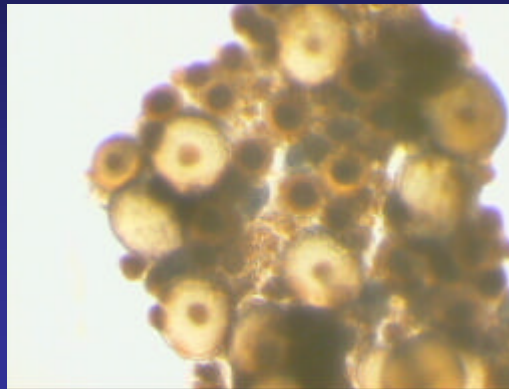
Tuna captive female

June 2004: 3 cases

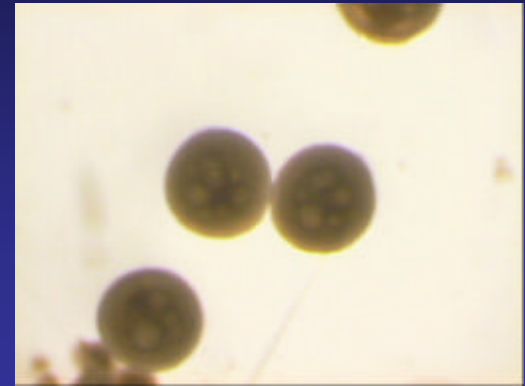
Aborted vitellogenesis



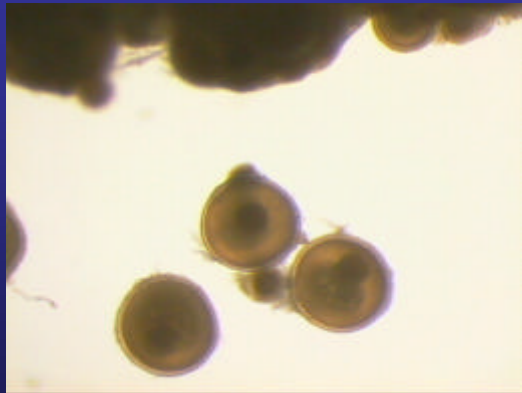
Full vitellogenesis



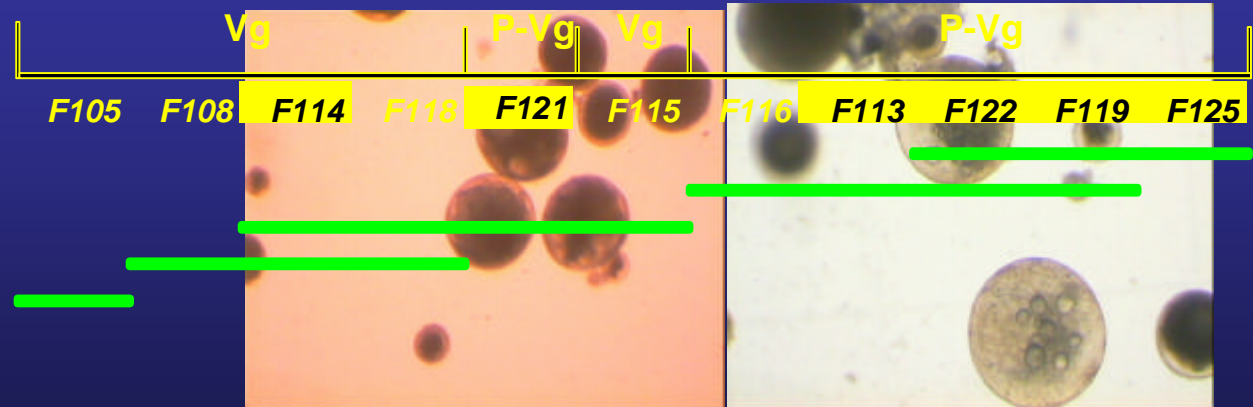
post vitellogenesis



+LHRH: Nucleus migration



Tissue incubation: Hydration or alteration?



F105 : Control

F114 : Treated

Perspectives

Sustainability of aquaculture requires trusty production of fry

This is obtained in salmonid carp, bream, bass ... all domesticated species

Tuna offers interesting possibilities :

- Reproduction in captivity observed in Japan
- Good indications for success in Europe

But...

High mortality due to handling prevent


- Monitoring of gametogenesis
- Programmation of spawning or stimulation

Solutions:

- Develop research programs for less stressful handling
- Use of less stress-susceptible fish (domesticated)

Closing the life cycle in captivity (Kinki University, 2002) is hopeful since it provides fish subjected to first rearing selection

Such fishes should be of great interest for the development of domestication process and reproduction control.



ども ありがとう

ございました