



# Reproductive cycle of wreckfish *Polyprion americanus* in captivity

Constantinos C. Mylonas<sup>1</sup>, Maria Papadaki<sup>1</sup>, Ioannis Fakriadis<sup>1</sup>, Jose Benito Peleteiro<sup>2</sup>, Blanca Alvarez-Blázquez<sup>2</sup>, J.L. Rodríguez Villanueva<sup>3</sup>, Fatima Linares<sup>3</sup>, Antonio Vilar<sup>4</sup>, Evaristo Pérez Rial<sup>2</sup>, Nuria Lluch<sup>2</sup>

<sup>1</sup>Hellenic Center for Marine Research, Crete, Greece

<sup>2</sup>Instituto Español de Oceanografía, Spain

<sup>3</sup>Xunta de Galicia, Spain

<sup>4</sup>Aquarium Finisterrae, A Coruña, Spain



## Wreckfish reproduction (2013)

- ✓ **Poor knowledge available on the process of gametogenesis, either in wild or captivity**
- ✓ **Spawning rare and limited egg production with strip-spawning (*in vitro* fertilization)**

### Description of the reproductive cycle

- ✓ **Recognize possible reproductive dysfunctions**
- ✓ **Establish broodstock management procedures**
- ✓ **Implement spawning induction protocols**

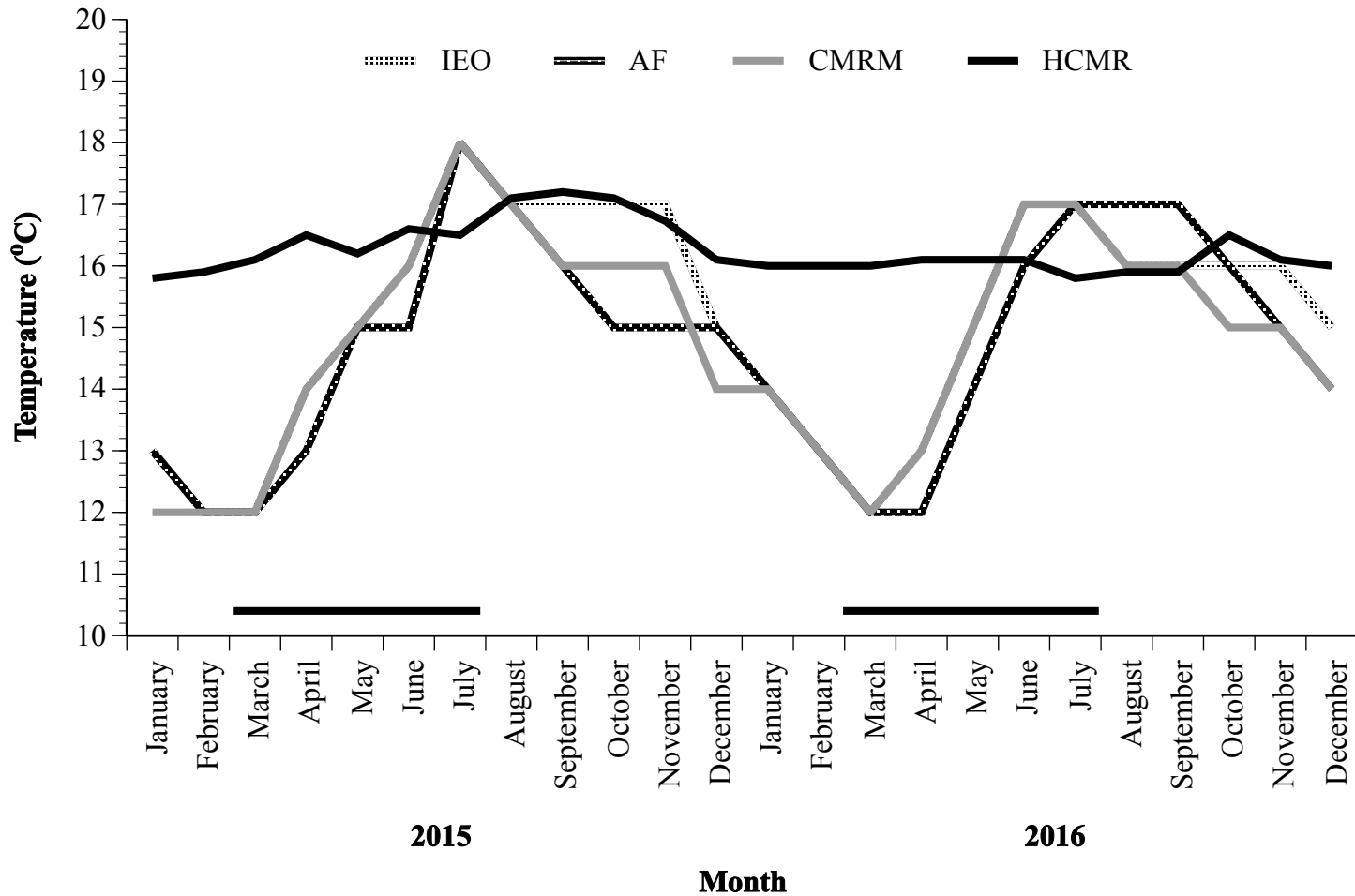
# Wreckfish broodstocks



# Wreckfish broodstocks

Broodstocks	IEO	CMRM	MC2	HCMR
Number of fish	13	21	11	3
Females	10	12	8	1
Males	3	9	3	2
Tank	110 m <sup>3</sup>	35 m <sup>3</sup>	180 m <sup>3</sup>	15 m <sup>3</sup>
Photoperiod	Natural	Simulated natural	Natural	Simulated natural
Temperature	Natural	Natural	Natural	16° C

# Wreckfish broodstocks - temperature



## Biopsy collection



## Sperm collection



## Blood collection



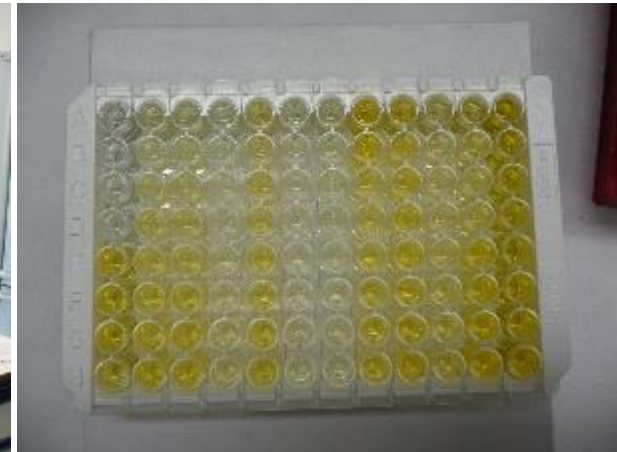
- **Bimonthly from August until January**
- **Monthly from February until July**



## Histological analysis

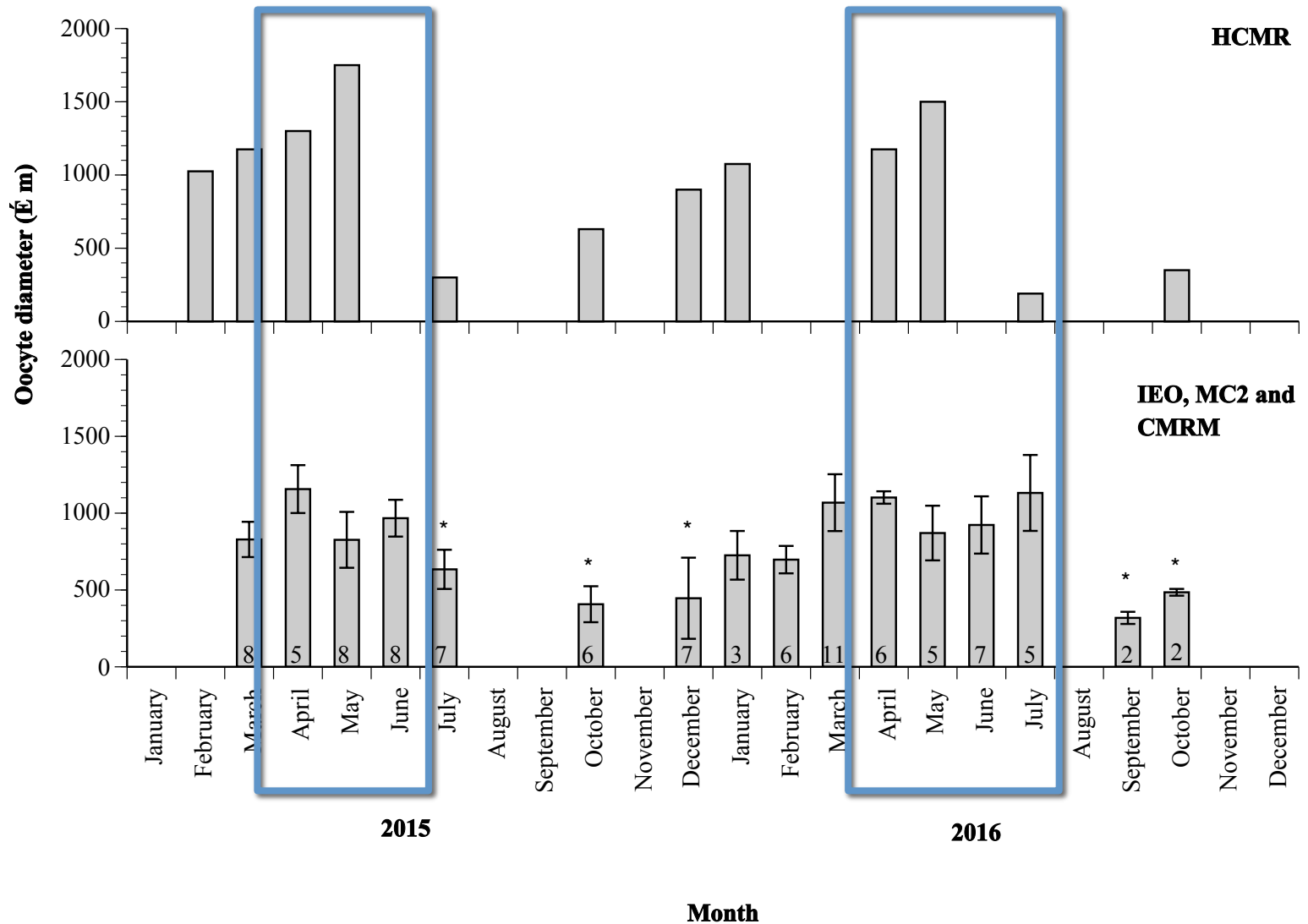


## Sperm evaluation



## Sex steroid hormones

# Reproductive cycle – max oocyte diameters



# Reproductive cycle – oogenesis

**Ld-lipid droplets**

**eVg-early vitellogenesis**

**aVg-advanced vitellogenesis**

**IVg-late vitellogenesis**

**eOM-early oocyte maturation**

**FOM-final stages of OM**

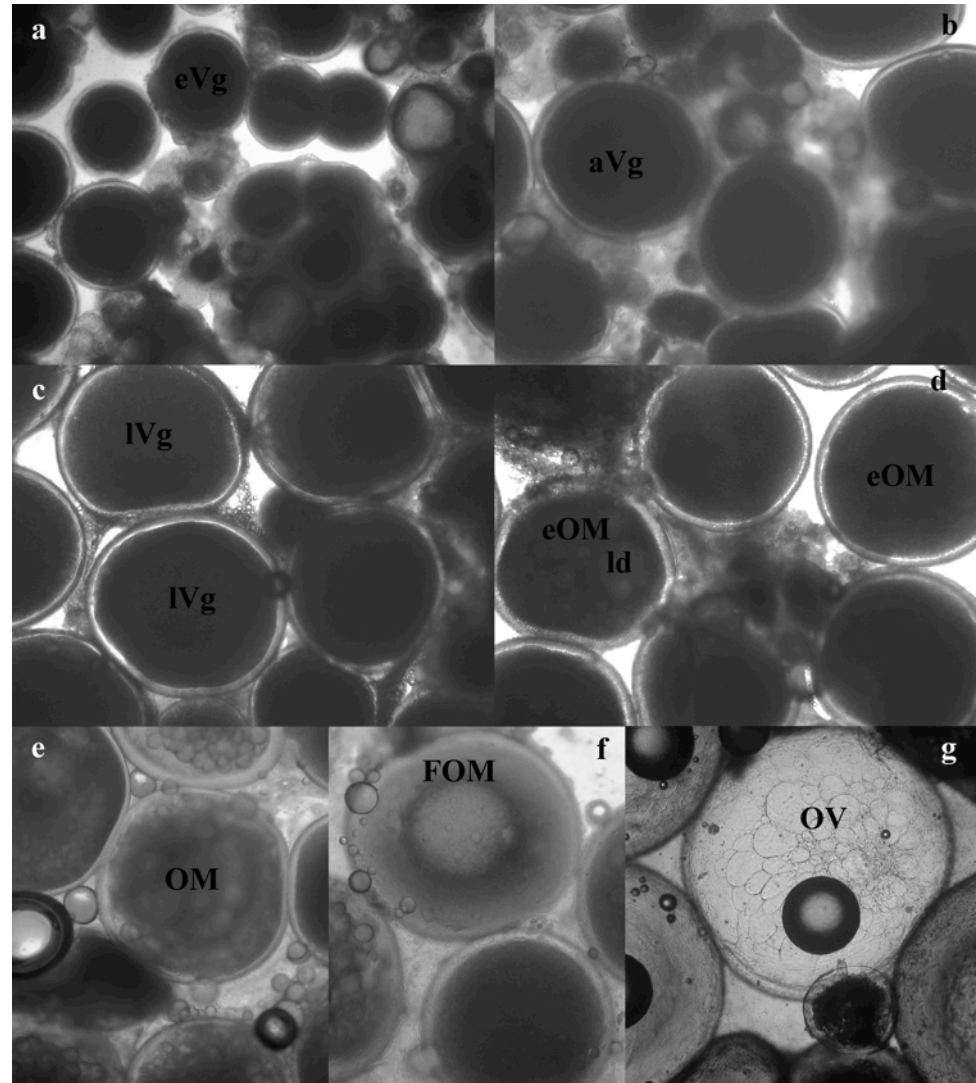
**OV-ovulation**

**Vitellogenesis in Winter**

**Maturation in Spring**

**Vg oocytes 1400 µm**

**Eggs 2000 µm**

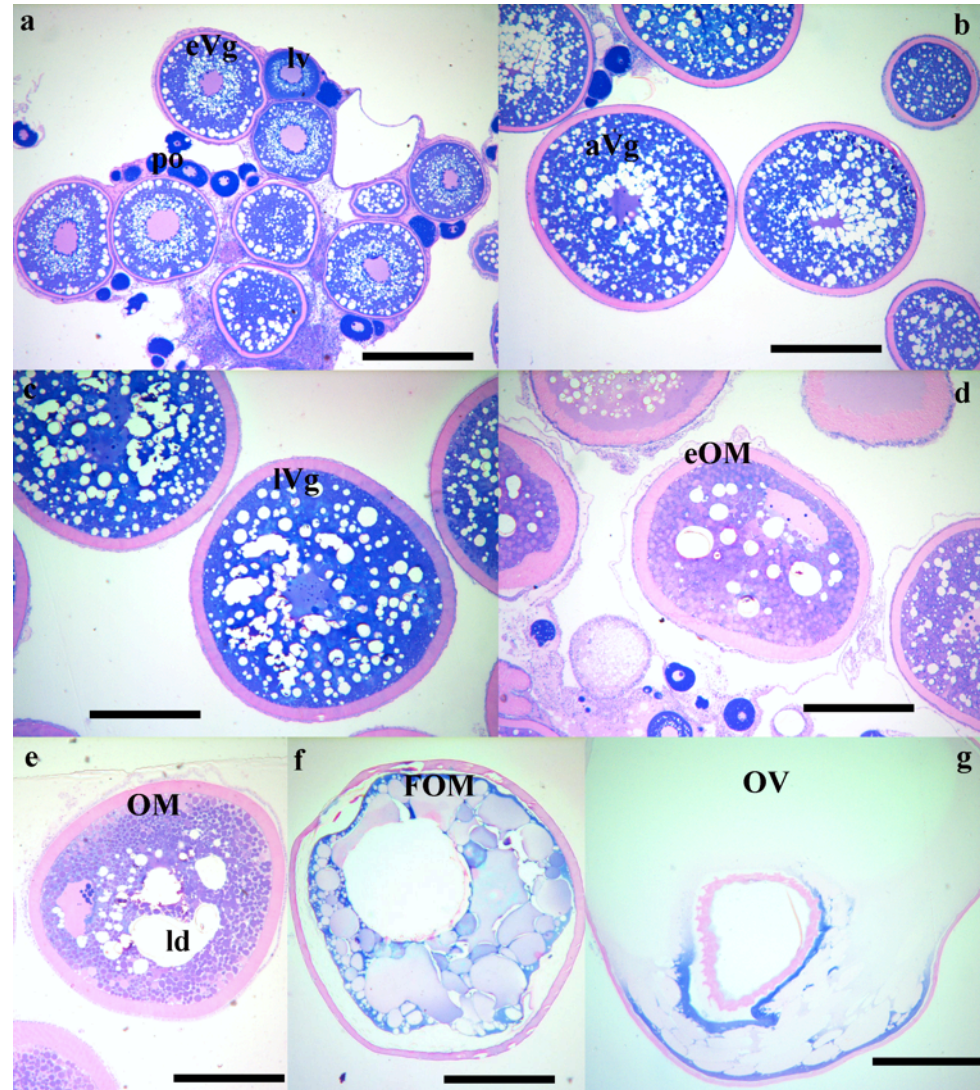




# Reproductive cycle – oogenesis

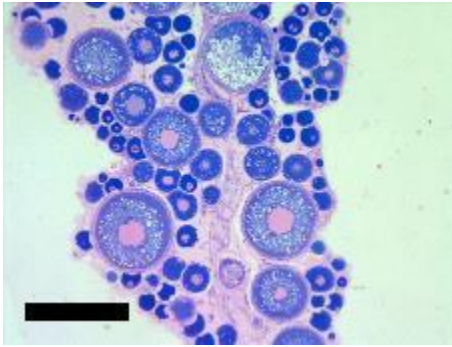
Bar: 500  $\mu$ m

po-primary oocytes  
ca-cortical alveoli  
Ld-lipid droplets  
eVg-early vitellogenesis  
aVg-advanced vitellogenesis  
IVg-late vitellogenesis  
eOM-early oocyte maturation  
FOM-final stages of OM  
OV-ovulation

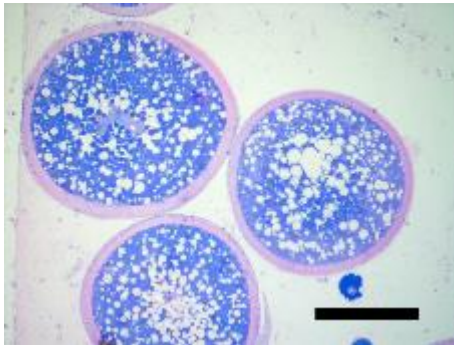


**Vitellogenesis in Winter**  
**Maturation in Spring**  
**Vg oocytes 1400  $\mu$ m**  
**Eggs 2000  $\mu$ m**

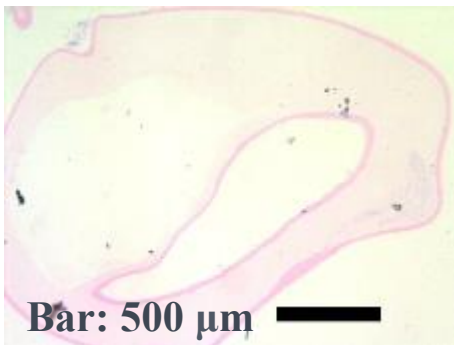
# Reproductive cycle – oogenesis



- 19% of the females with arrested oocyte development at the cortical alveoli stage (350  $\mu\text{m}$ )

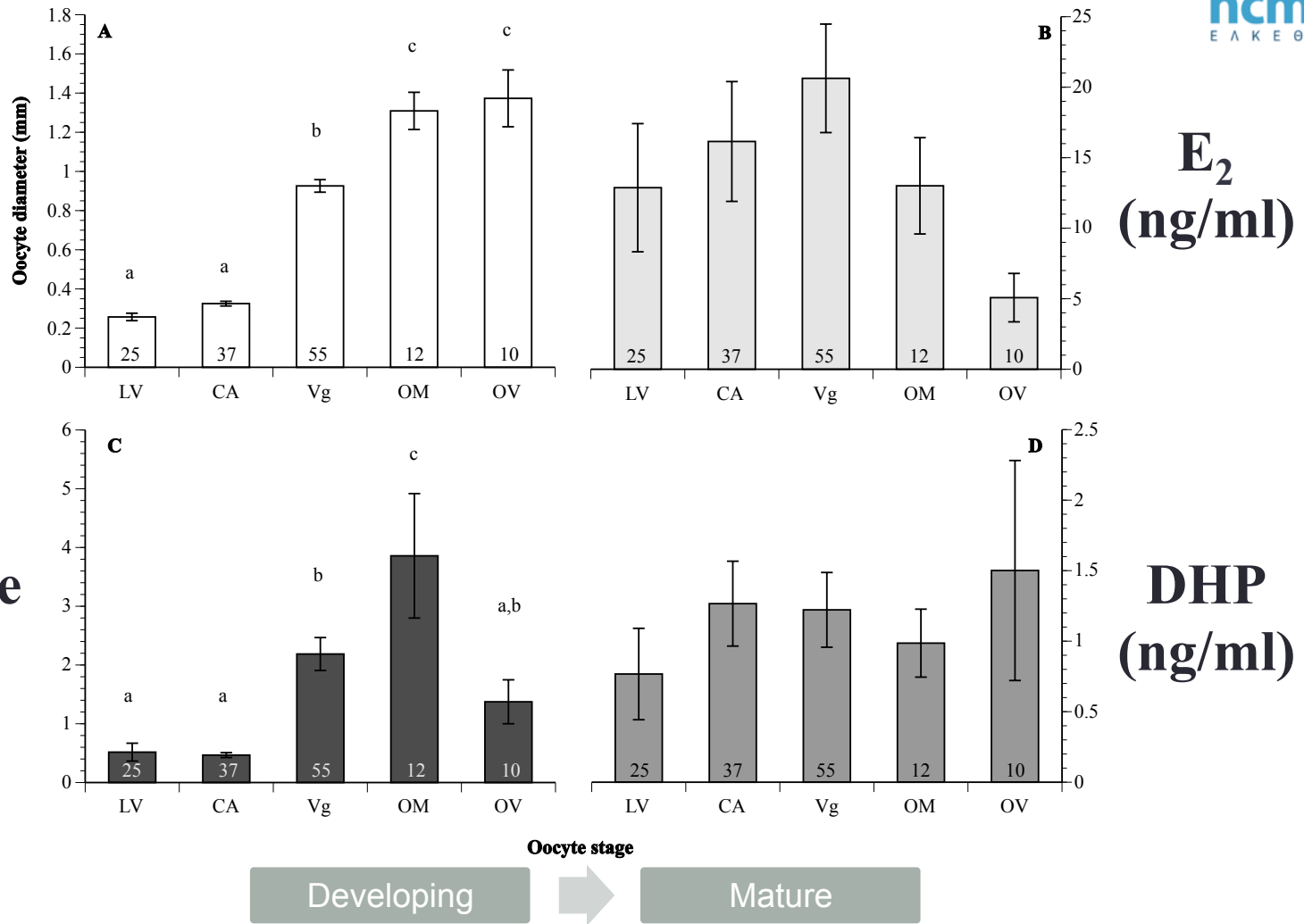


- 65% of the females with complete vitellogenesis, but no maturation (~1200  $\mu\text{m}$ )



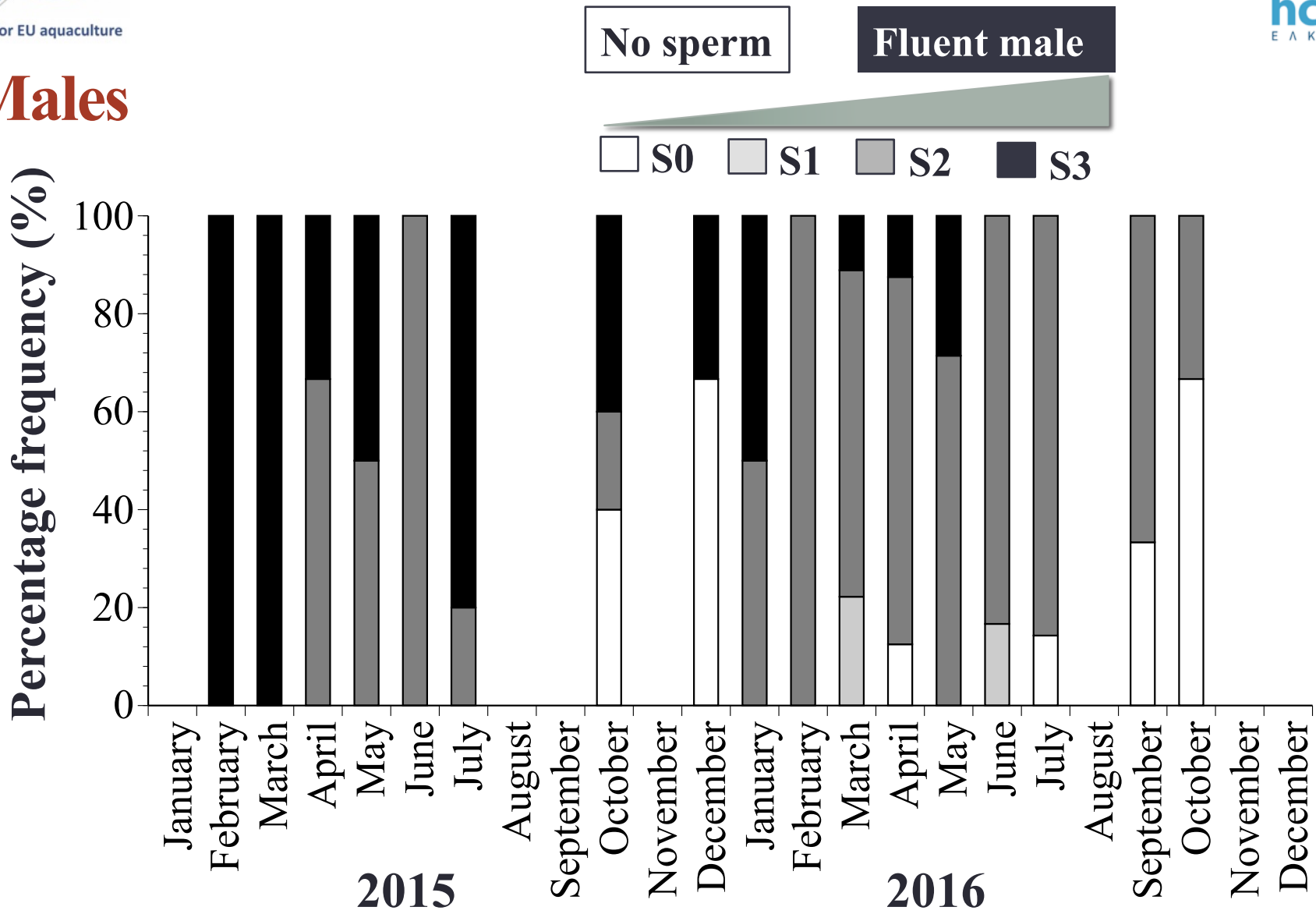
- 16% of the females spawned spontaneously (2000  $\mu\text{m}$ )

# Females



- Estradiol high during vitellogenesis
- Testosterone peaked at maturation
- 17α,20β-DHP low and unchanged (irrelevant as MIS?)

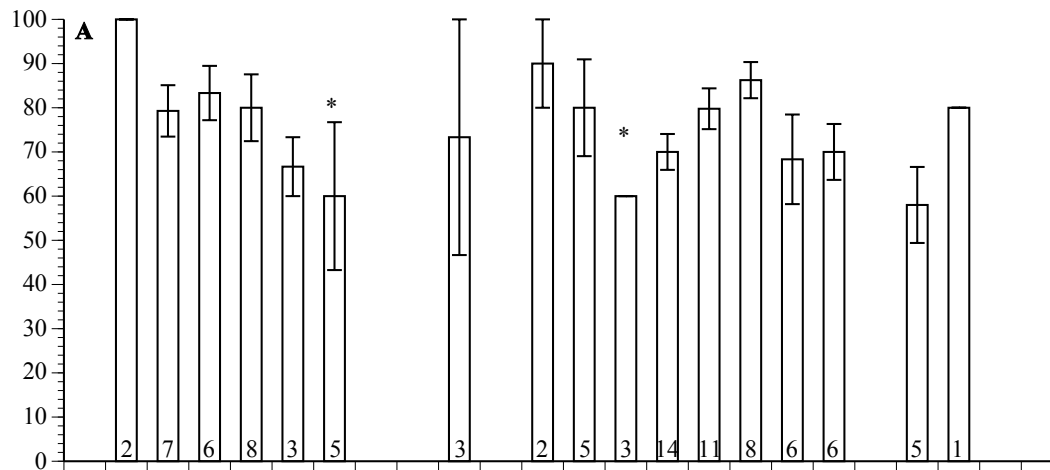
# Males



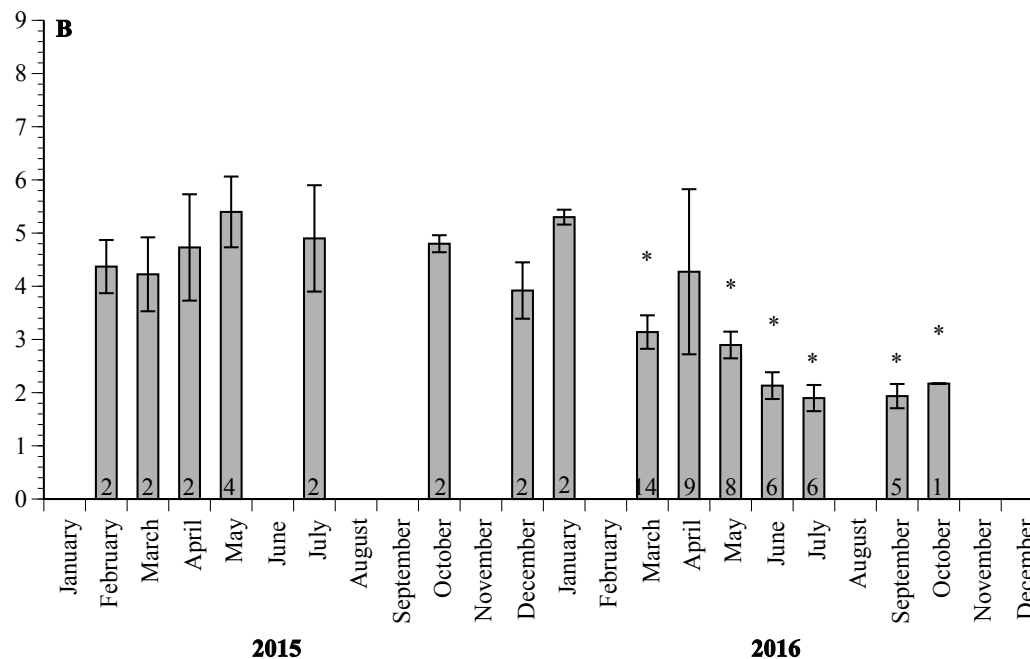
➤ Sperm was produced almost all year

# Sperm quality

## Sperm motility (%)



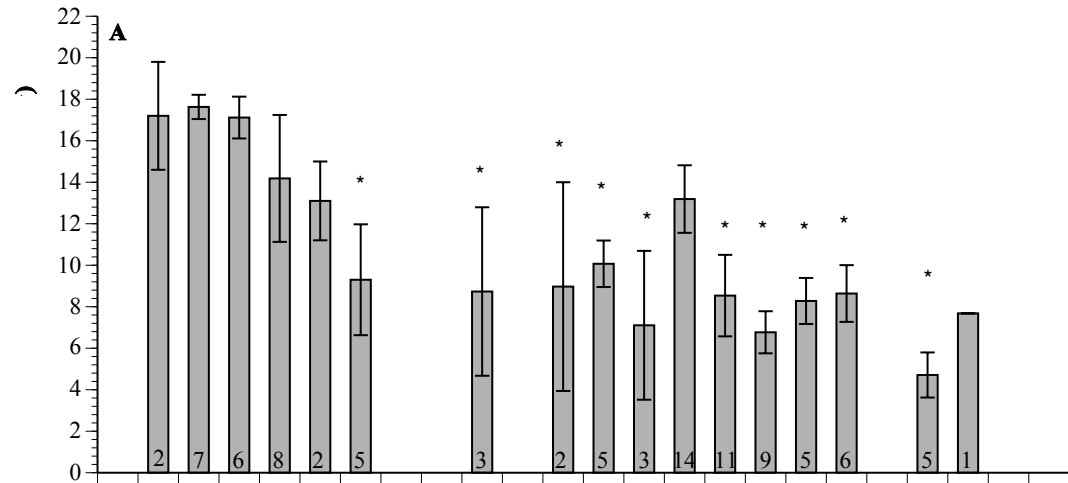
## Duration of motility (min)



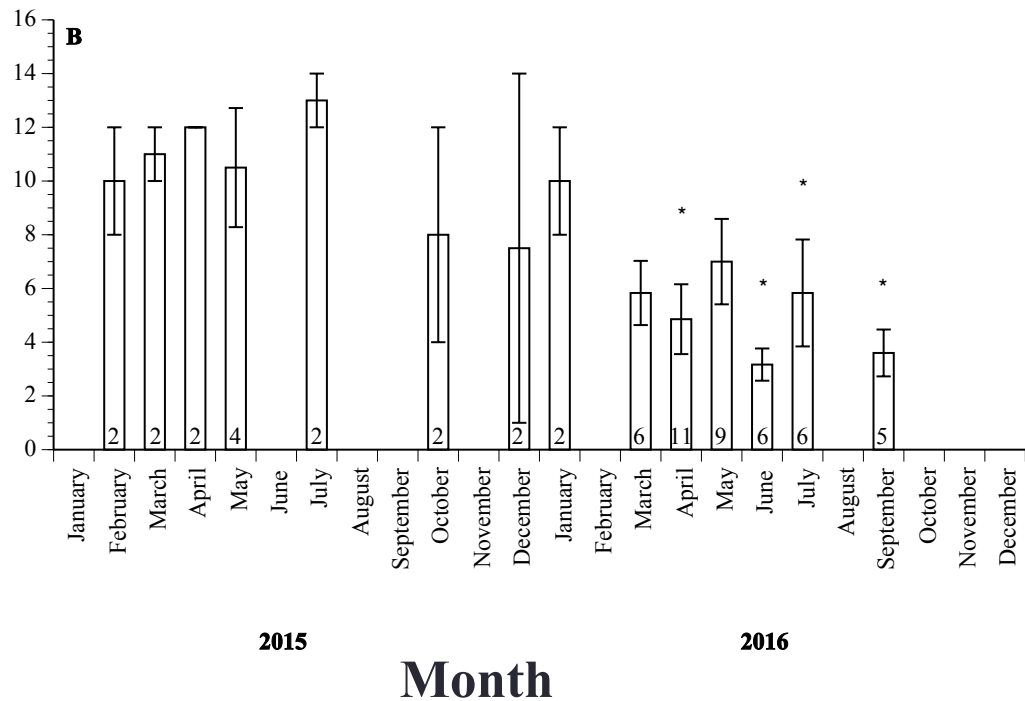
## Month

# Sperm quality

Szoa density  
( $\times 10^9 \text{ ml}^{-1}$ )

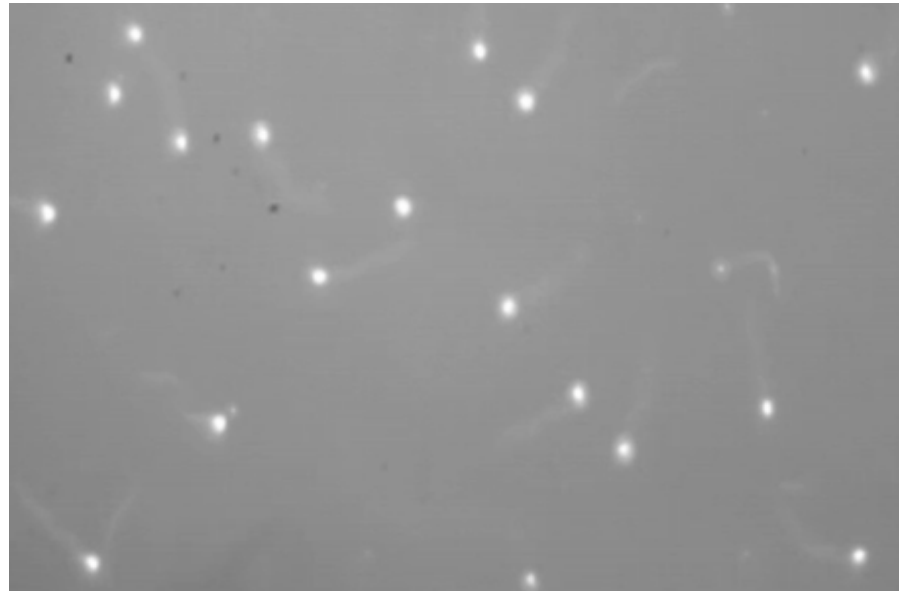


Sperm survival  
(days)

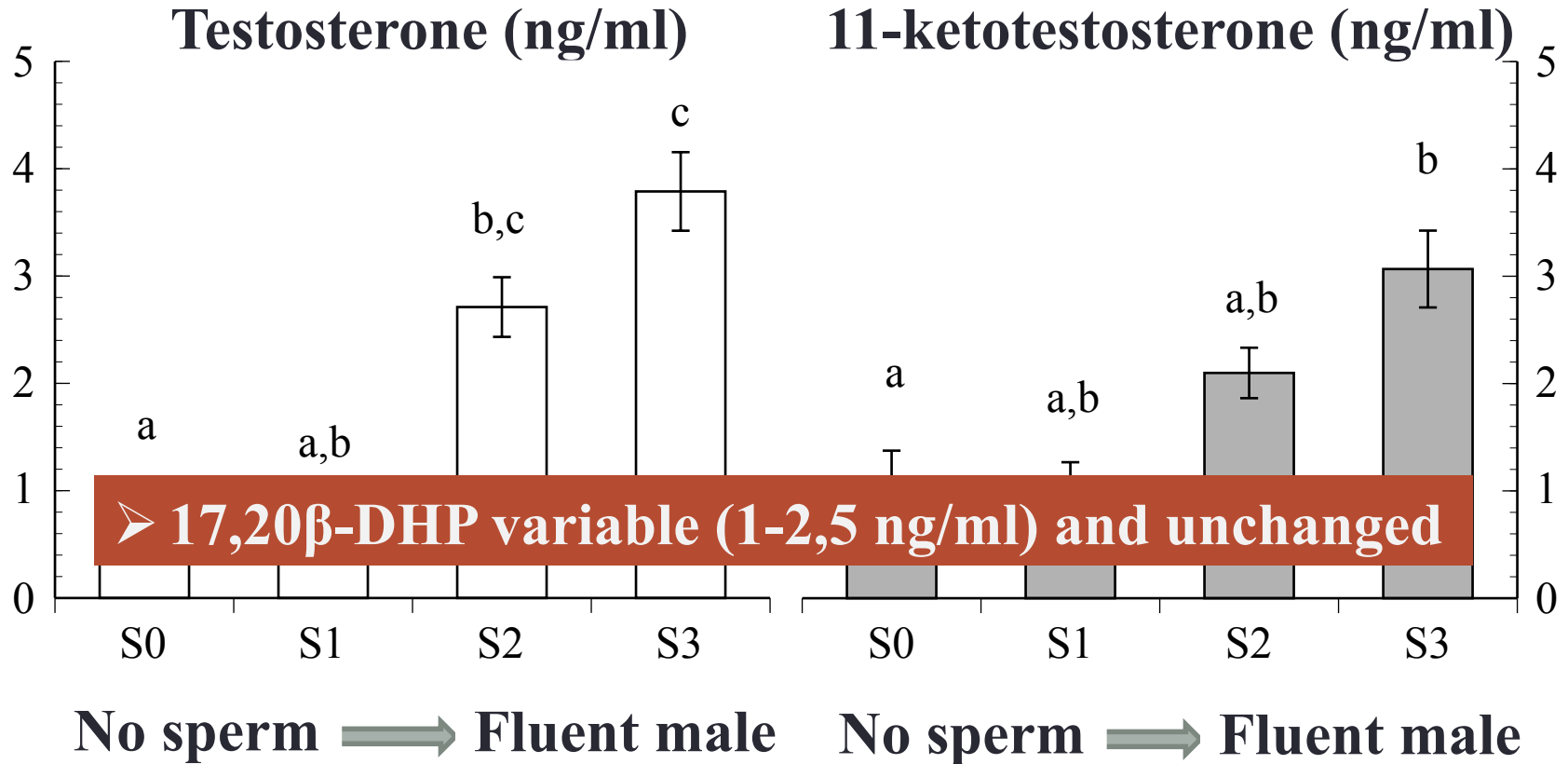


## Sperm quality - summary

- sperm density between  $4.5 - 11.5 \times 10^9$  sperm ml<sup>-1</sup>
- sperm motility always >60%
- motility duration 1.5 - 6 min
- survival of sperm at (4°C) for 3 - 10 days



# Males



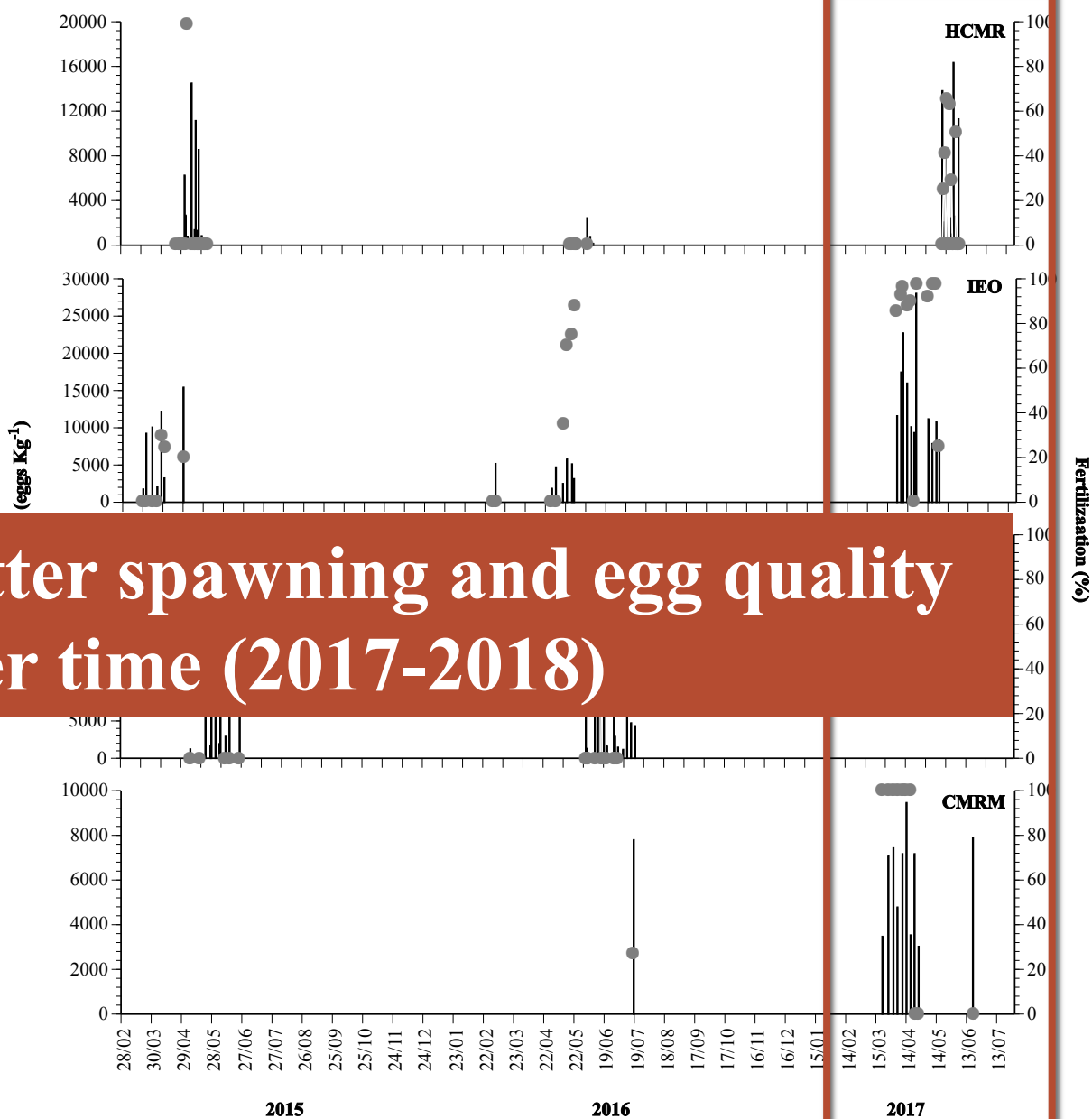
- Testosterone and 11-ketotestosterone increased with increased sperm production





Relative fecundity (eggs kg female<sup>-1</sup>)

Fertilization (%)



Better spawning and egg quality over time (2017-2018)

Year

## **Reproductive cycle in captivity – Females**

- 1. Undergo gametogenesis, but maturation and egg production may take some years to reach adequate levels**
- 2. Reproduction under fluctuating or constant low temperature**
- 3. Plasma T and E<sub>2</sub> followed the proper fluctuations during oogenesis, but 17,20β-DHP was not correlated to reproductive stage (not the MIS?, sample timing?)**

**Better understanding of the required environmental conditions, to ensure that more fish complete gametogenesis and mature/spawn.**

## **Reproductive cycle in captivity – Males**

- 1. Produce sperm all-year round (natural or captivity-induced?)**
- 2. Plasma T and 11-KT followed the proper fluctuations during spermatogenesis, but 17,20 $\beta$ -DHP was not correlated to reproductive stage (not the MIS?)**
- 3. Sperm was of good quantity and quality**

**Sperm availability and quality is not limiting to reproduction in captivity, but breeding behavior may not be fully expressed. More focus on environmental requirements for successful spawning**

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## Many thanks to:

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**Maria Papadaki<sup>1</sup>, Ioannis Fakriadis<sup>1</sup>, Jose Benito Peleteiro<sup>2</sup>, Blanca Alvarez-Blázquez<sup>2</sup>, J.L. Rodríguez Villanueva<sup>3</sup>, Fatima Linares<sup>3</sup>, Antonio Vilar<sup>4</sup>, Evaristo Pérez Rial<sup>2</sup>, Nuria Lluch<sup>2</sup>**

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