



Meagre



INDUCED SPAWNING OF PAIRED MEAGRE (*ARGYRO SOMUS REGIUS*) WITH MALE ROTATION: AN APPROACH TO PRODUCE MULTIPLE FULL AND HALF-SIB FAMILIES FOR GENETIC BREEDING PROGRAMS

DIVERSIFY - EU PROJECT

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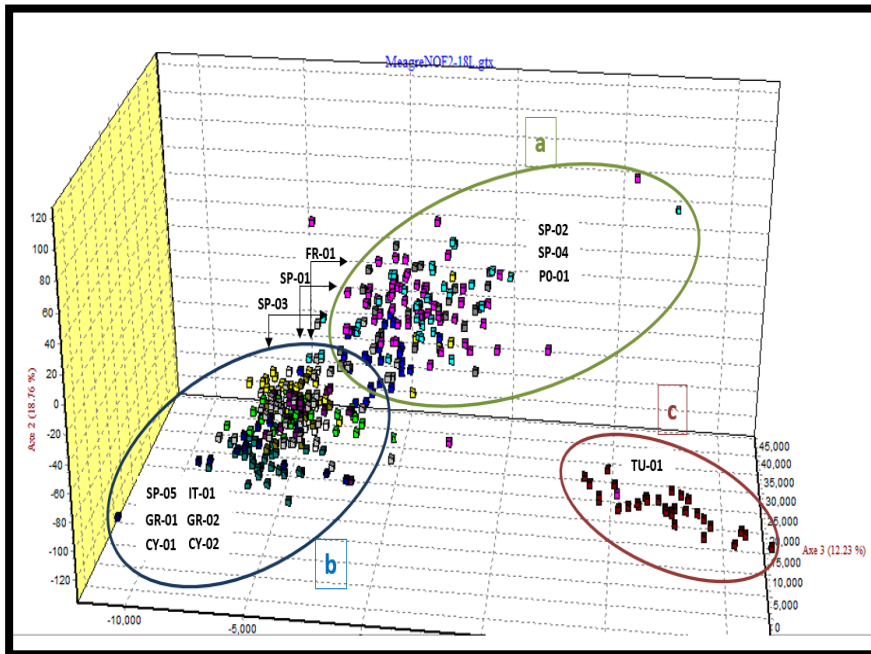
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Task 2.2 Development of protocols for paired crossing in spontaneous spawning
Deliverable 2.3 Protocol for paired spontaneous tank spawning of meagre
(Delivered in 2015)



Introduction: Bottlenecks

Need for breeding programs.



Broodstocks

1. Adequate genetic variation for a breeding program,
2. Low mean number of alleles
3. Enlarged broodstocks with new families
4. Reproductive control





Meagre

Diversify to examine 2 approaches:

- 1. Spawning of pairs to produce a desired family**
- 2. Strip spawning – *In vitro* fertilization**

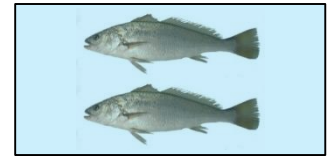
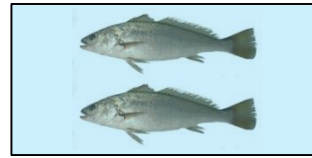
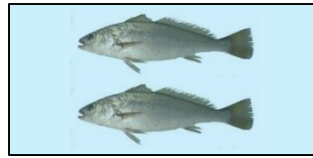
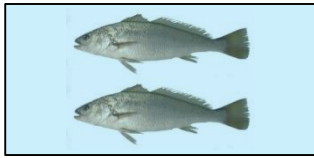
Objective:

Protocol for the induction of spontaneous tank spawning of pairs of meagre (one male and one female) to produce a maximum number of families.



Experiments completed:

An experiment that examined the maximum number of spawnings in response to weekly GnRHa injections (HCMR in 2014)



Four experiments to examine paired spawnings with male rotation on a weekly basis (IRTA in 2014 and 2015; HCMR in 2015)

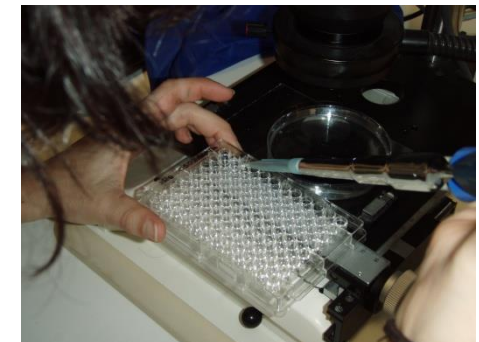


Common methods, induced spawning:

1. Females , > 500 μ m oocytes induced with single injection 15 μ g / kg GnRH α
2. Males with sperm, either:
 - single injection 15 μ g / kg GnRH α ,
 - single injection 7.5 μ g / kg GnRH α or
 - implant ~ 50 μ g / kg GnRH α

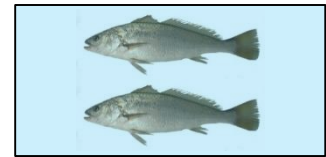
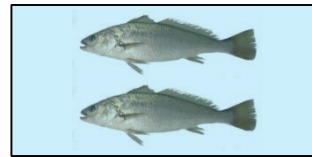
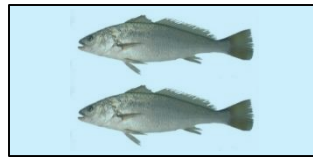
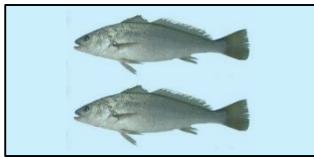
Common methods, egg quality:

1. Count number of eggs spawned, volumetric sub-samples
2. Fertilization, from egg (n>100) development when collected
3. Hatching and 5 day larval survival in 96 well plates, 2 plates / spawn

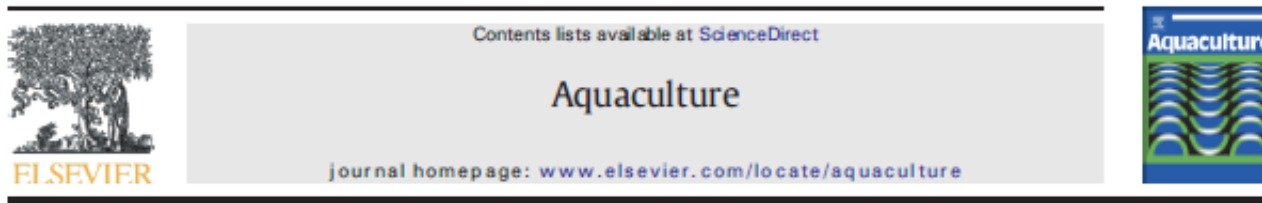


Experiments completed:

An experiment that examined the maximum number of spawnings in response to weekly GnRHa injections (HCMR in 2014)



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Enhancement of oogenesis/spermatogenesis in meagre *Argyrosomus regius* using a combination of temperature control and GnRHa treatments

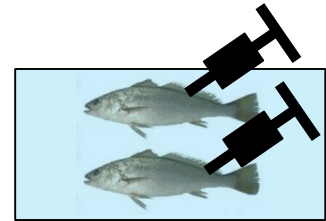
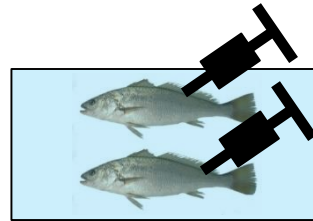
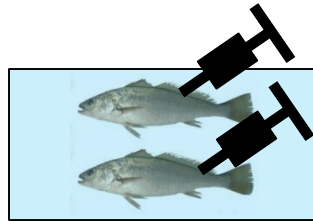
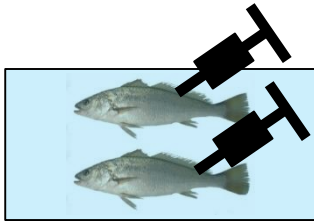


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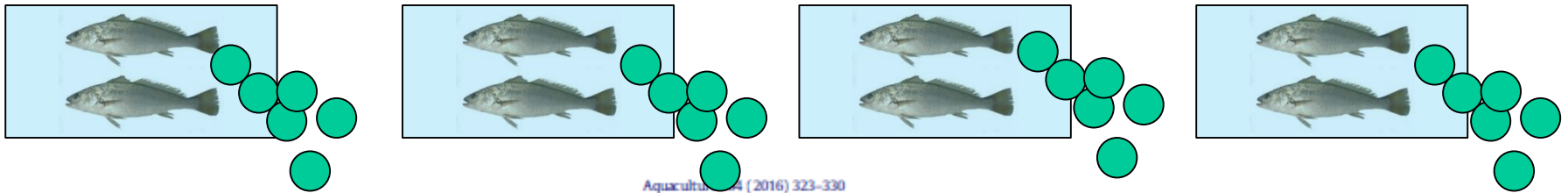


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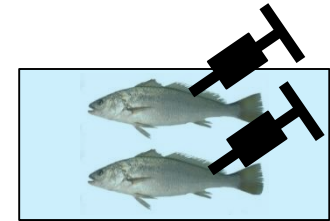
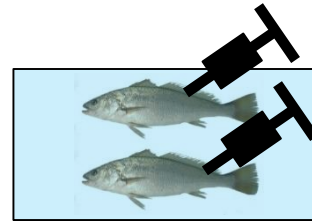
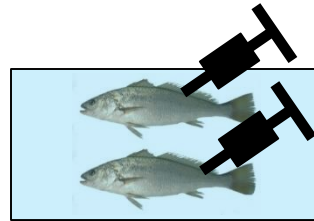
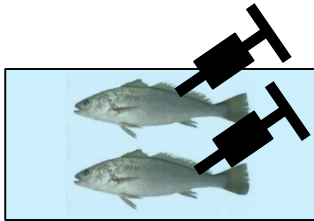


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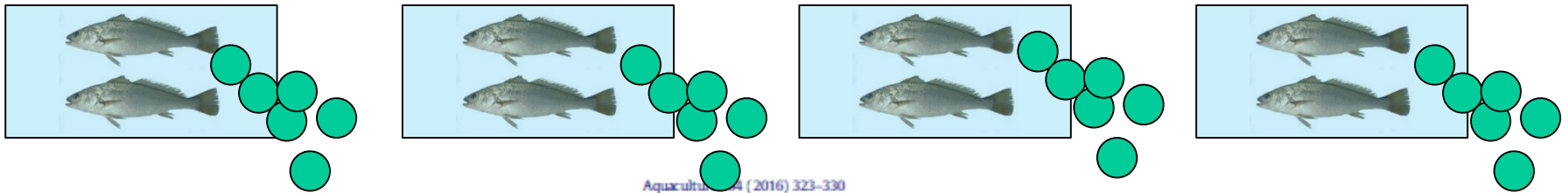


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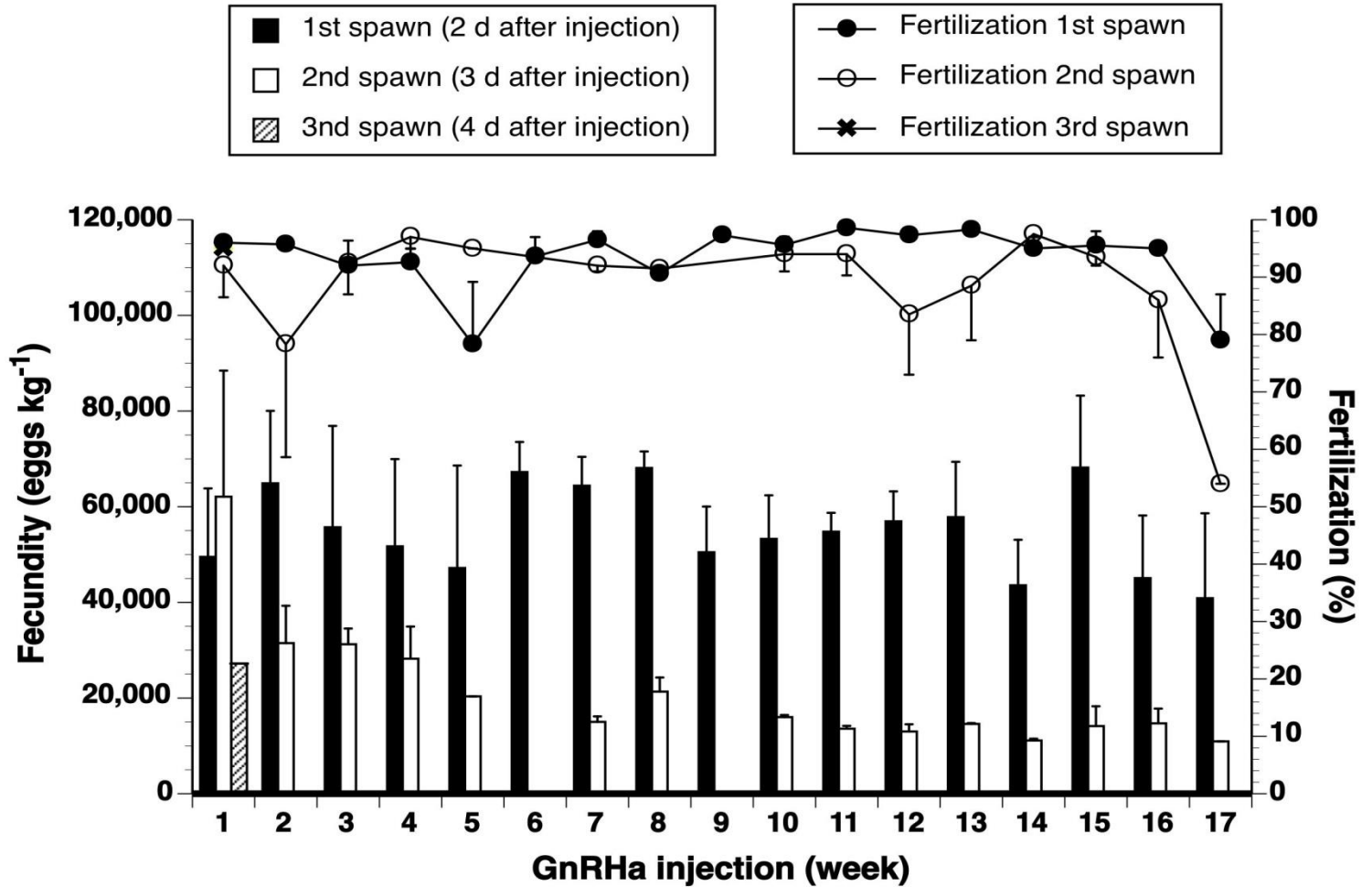
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Results: Fecundity



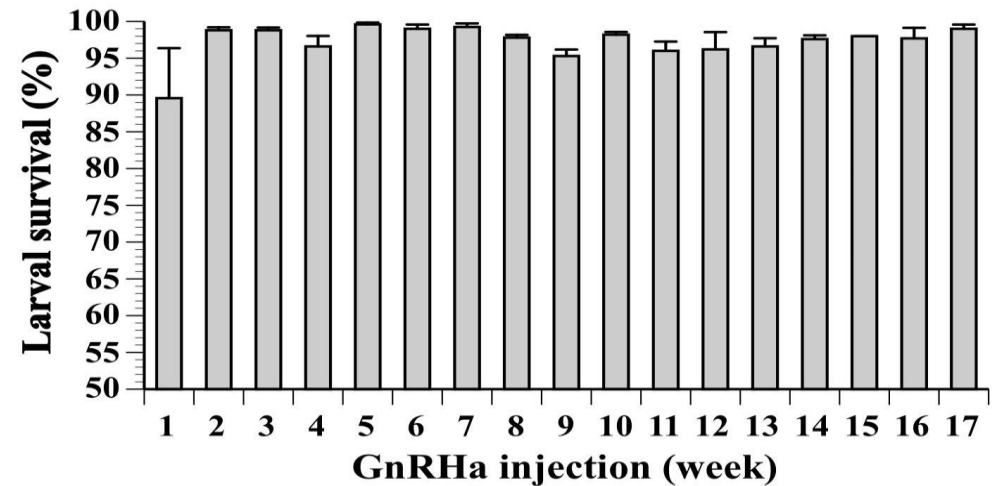
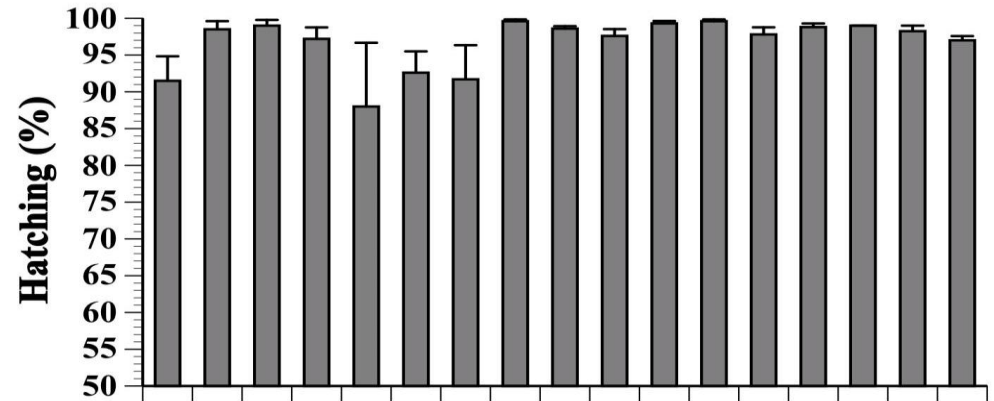
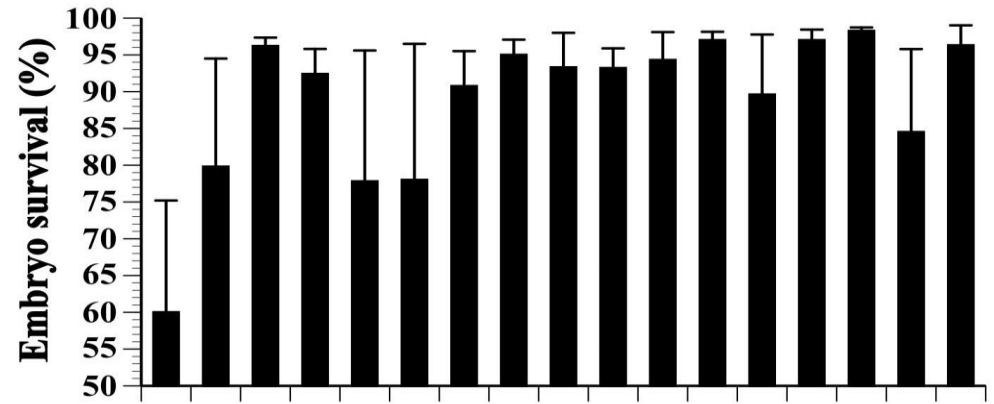


Results: Egg quality

High fertilization

High hatching %

High 5 day larval survival





Meagre

Conclusion

**Pairs of meagre
can be induced to spawn
high quality and quantity of eggs
on a weekly bases
for up to 17 weeks**



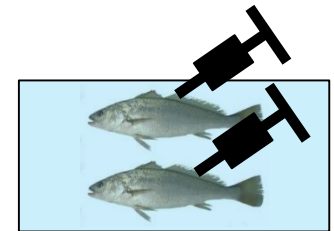
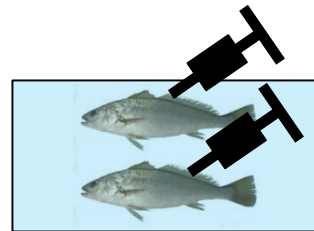
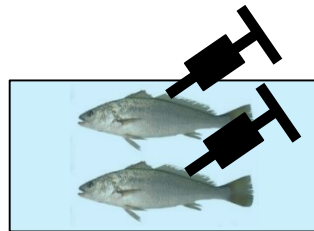
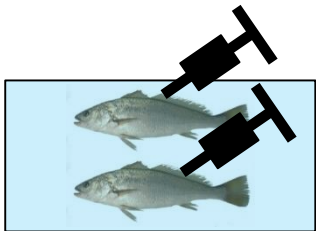
Experiments completed:

Four experiments to examine paired spawnings with male rotation on a weekly basis (IRTA in 2014 and 2015; HCMR in 2015)



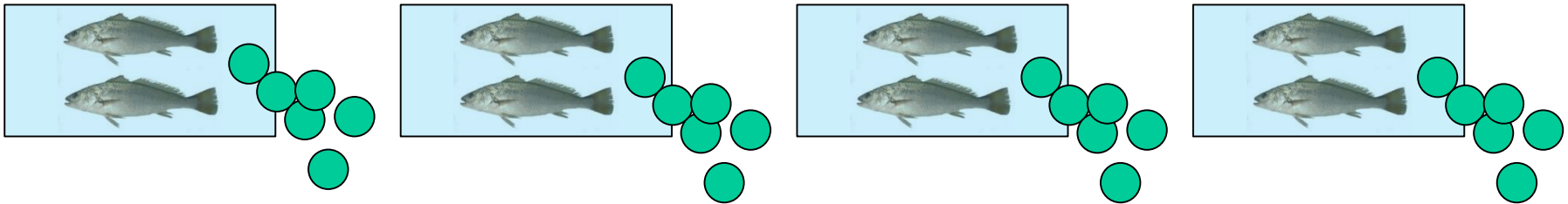
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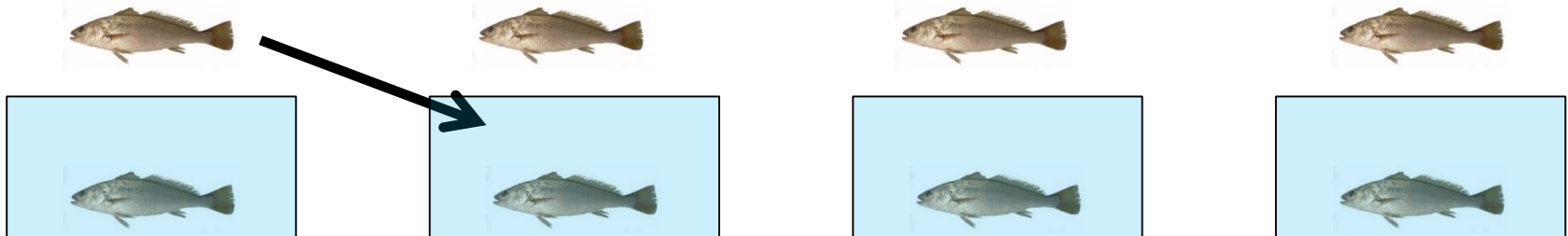
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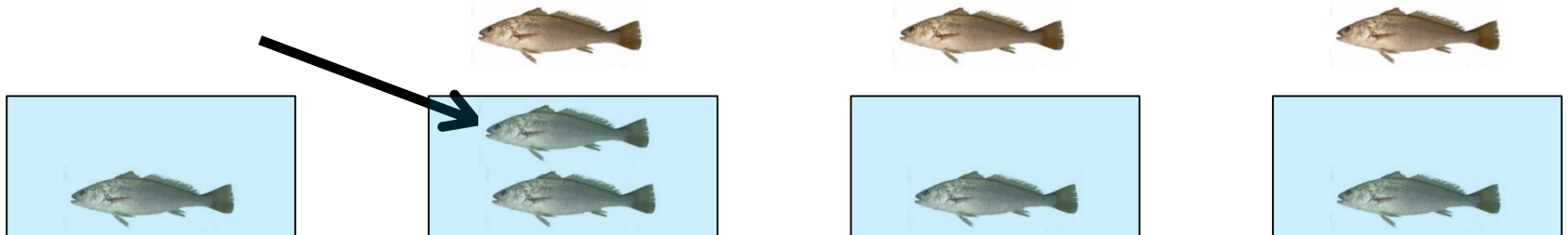
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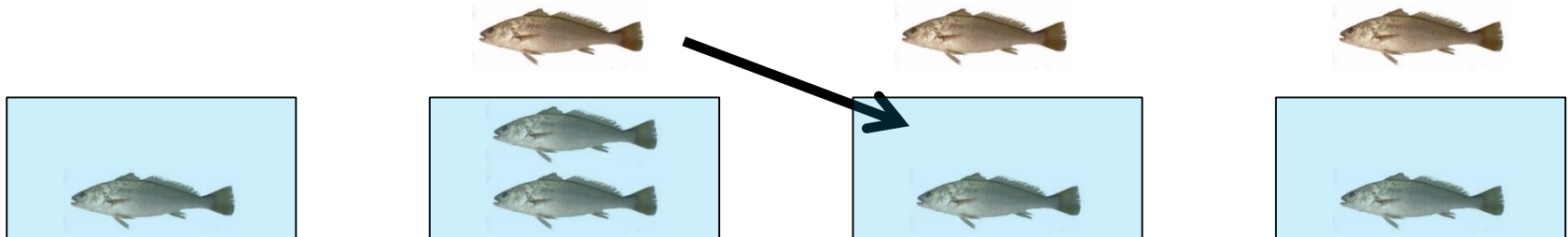
Experiments completed:

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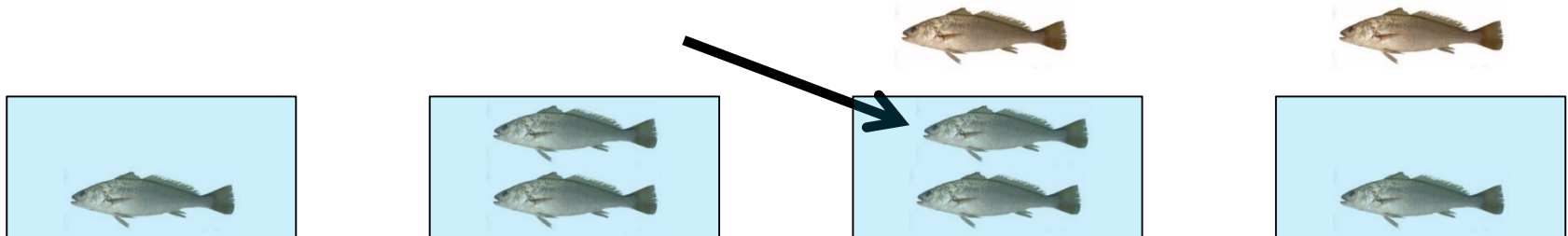
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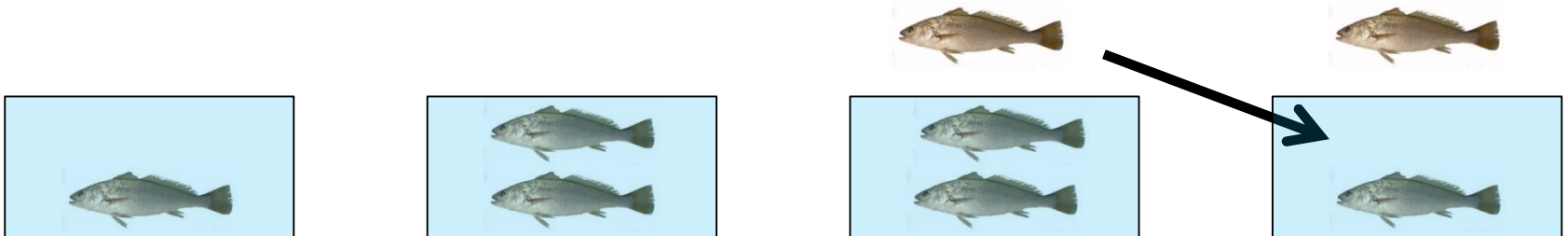
Experiments completed:

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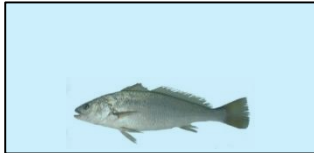
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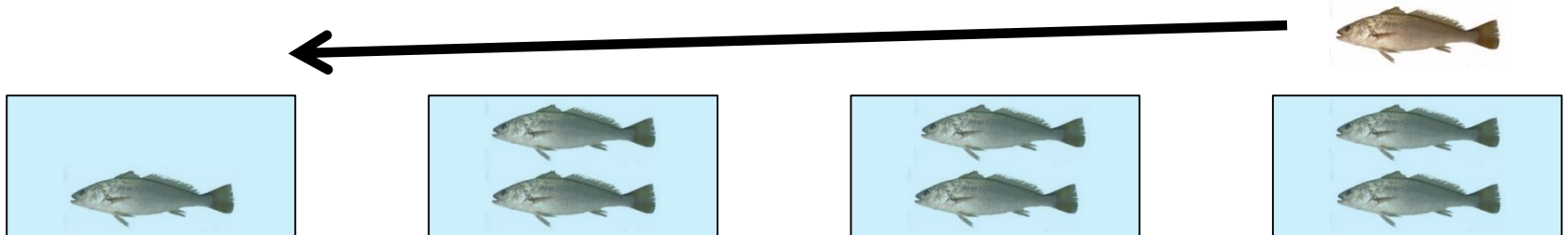
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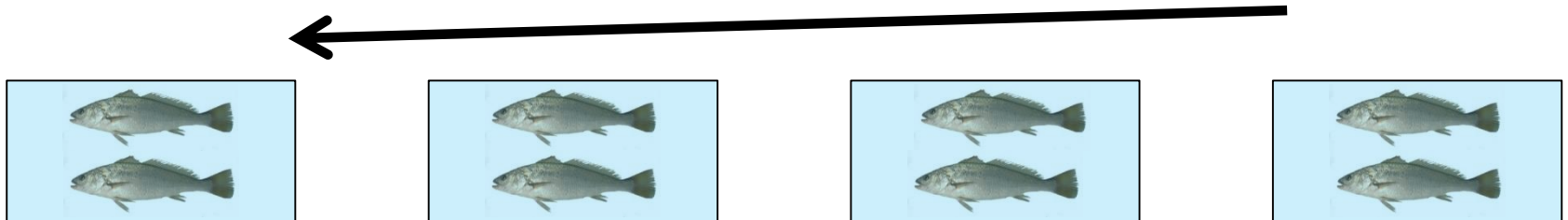
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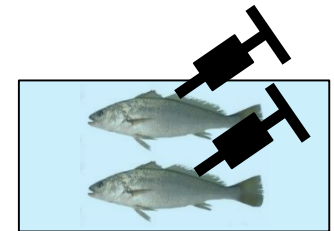
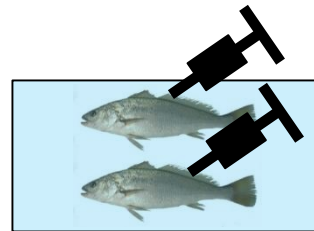
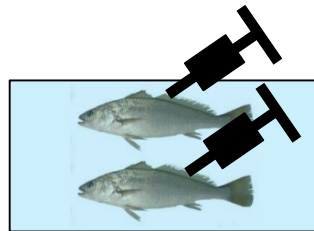
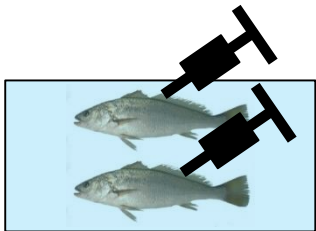
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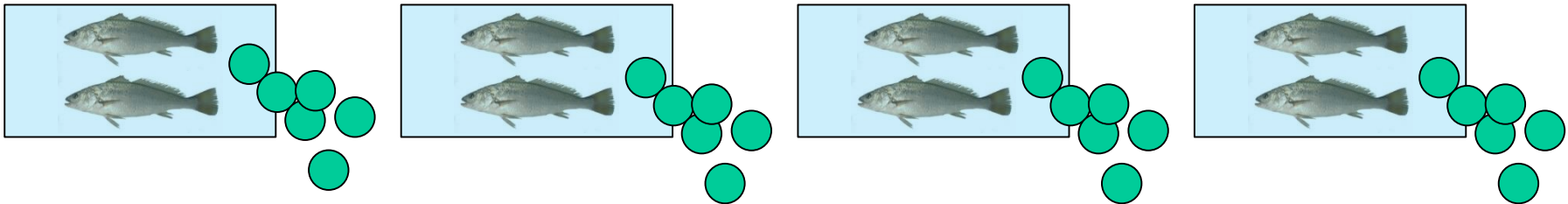
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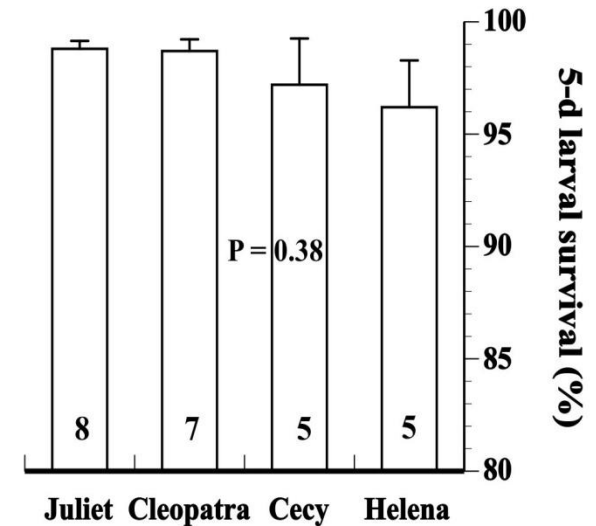
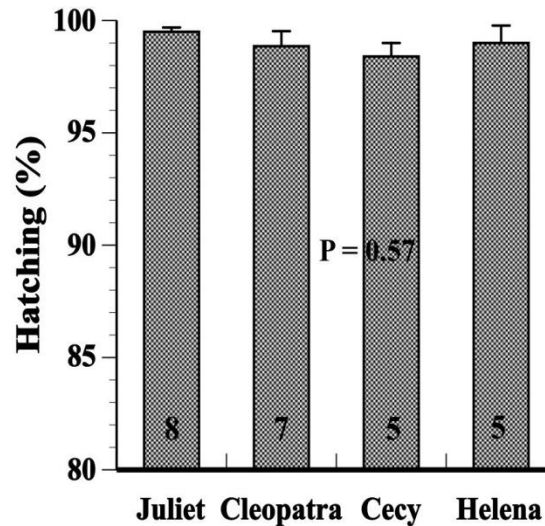
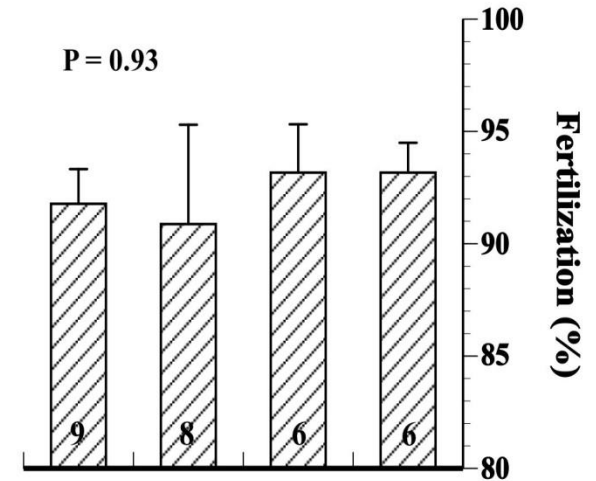


Results: Egg quality

High fertilization

High hatching %

High 5 day larval survival



Female-specific egg production and quality

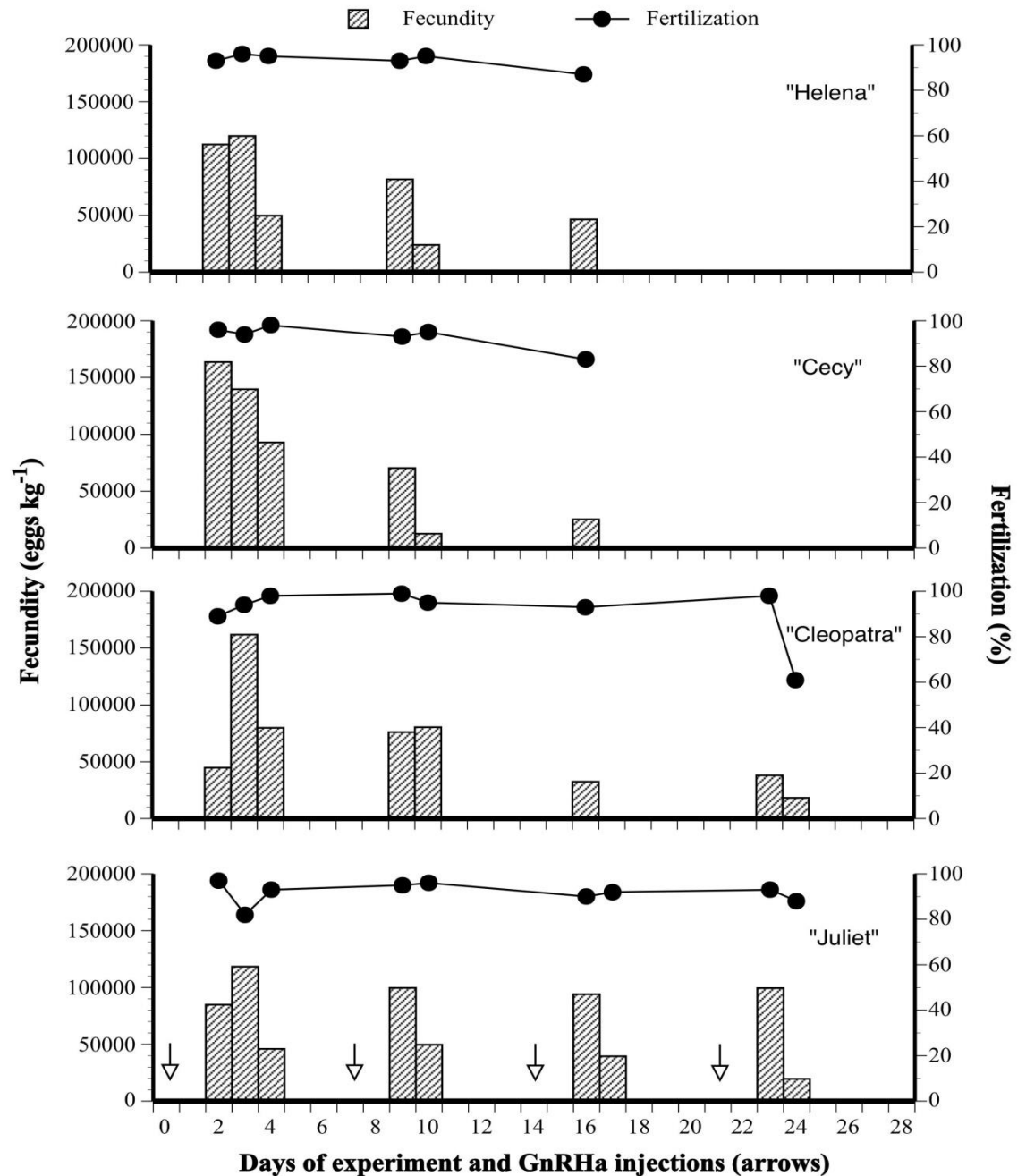




Results: Fecundity

Two pairs did not spawn after 4th injection of GnRH_a

Apparent decline in fecundity



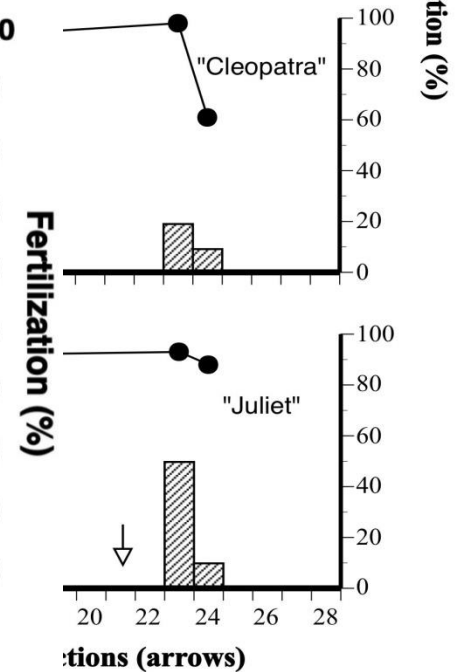
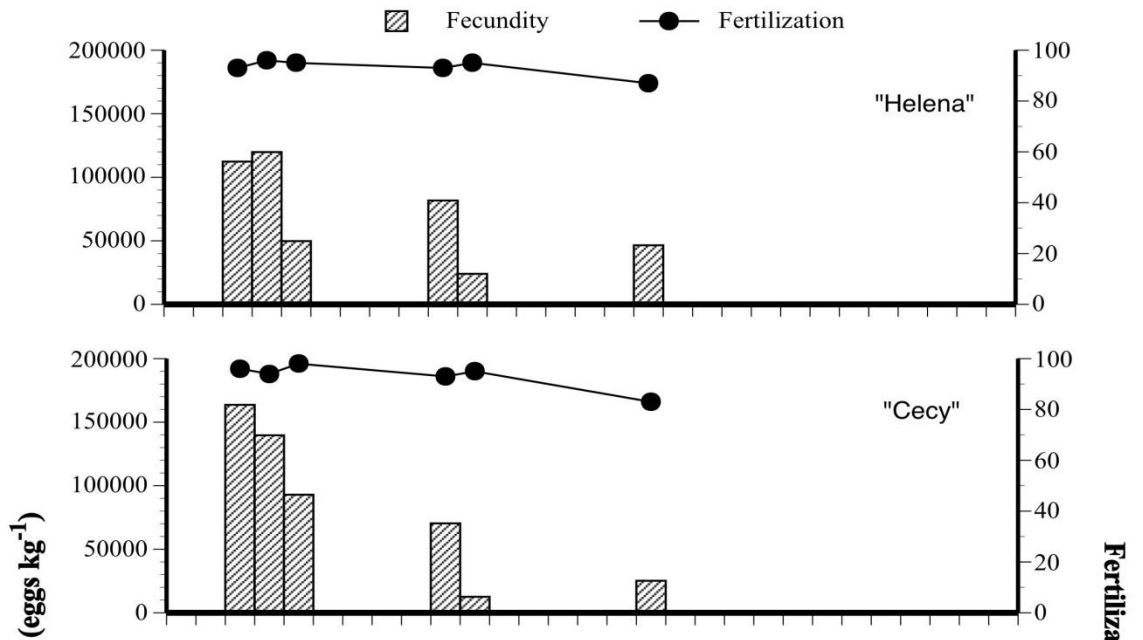
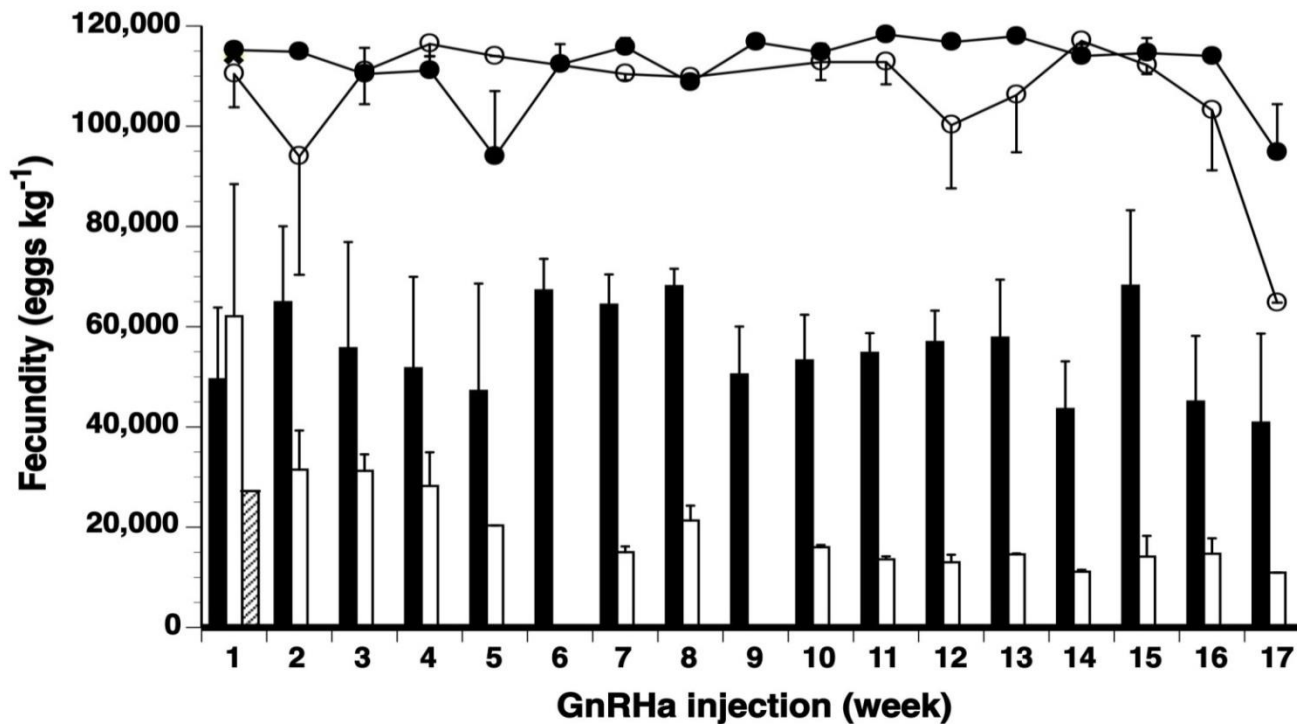


DIVERSIFY



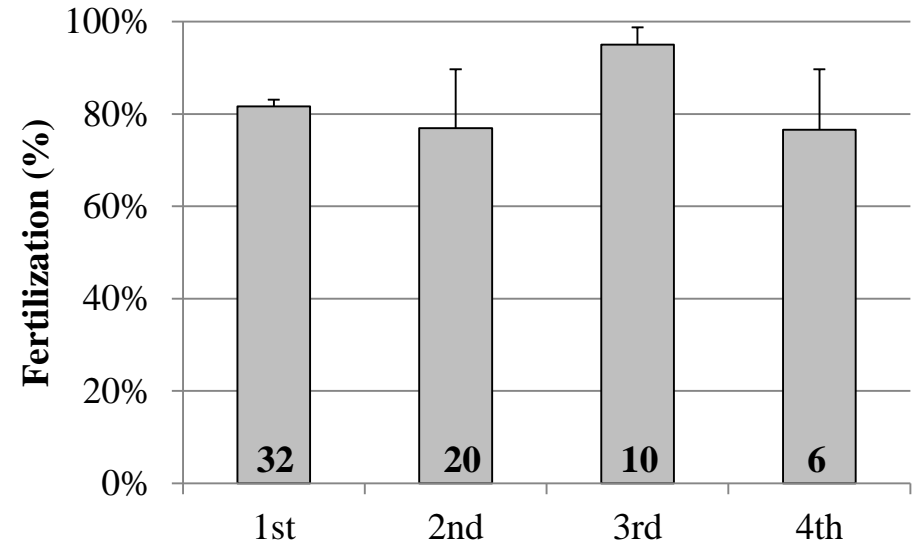
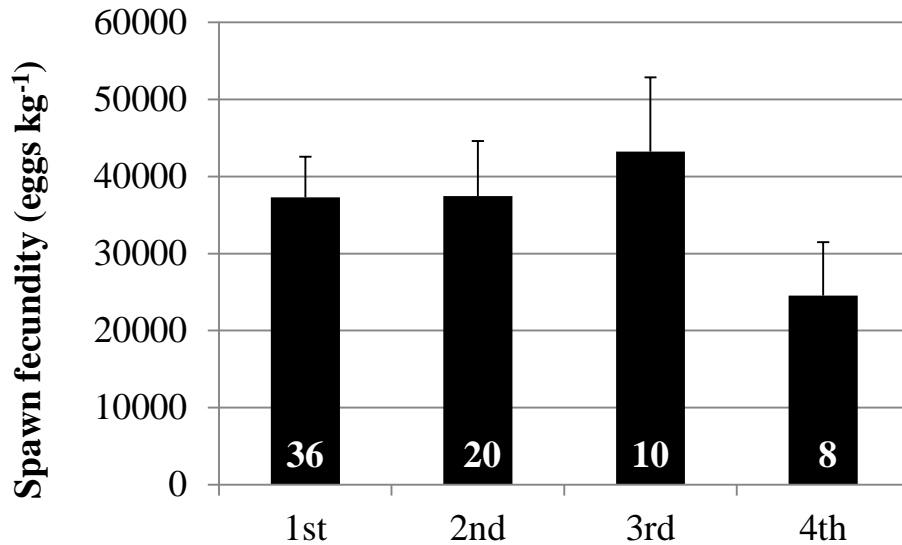
Results: Fecundity

Effect of male rotation?
Stress?



Results: Fecundity

Significant decline in fecundity with successive injections of GnRH α

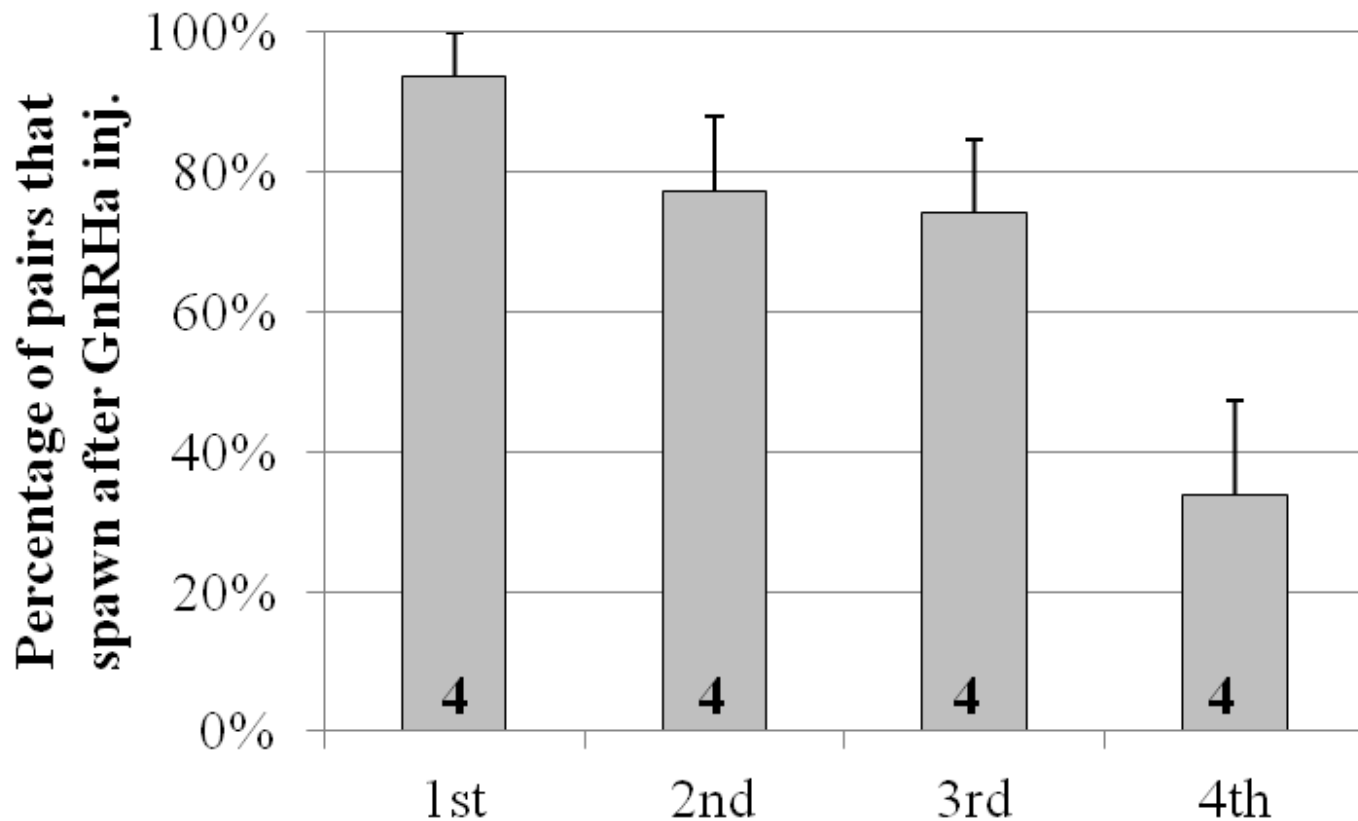


GnRH α injection for female



Results: Spawning success

Decline in percentage of pairs that spawn with successive injections of GnRHa





Meagre

Conclusion

**The success of spawning pairs with male rotation was 76%
to produce 61 families (full and half-sib)
that had >200,000 good quality eggs.**





Meagre

Conclusion

Paired spawning of meagre was possible for the production of known families from parents with known phenotypes





Meagre

Conclusion

A successful “proof of concept” for the paired spawning approach, highlighting the both the positive potential of the approach and possible drawbacks





Co-funded by the Seventh Framework Programme of the European Union



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This project has received funding from the European Union's Seventh Framework Programme for research, technological development and demonstration (KBBE-2013-07 single stage, GA 603121, DIVERSIFY).



**Thank you,
for all technical help
In HCMR and IRTA**



Gracias por su atención

El fin.



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