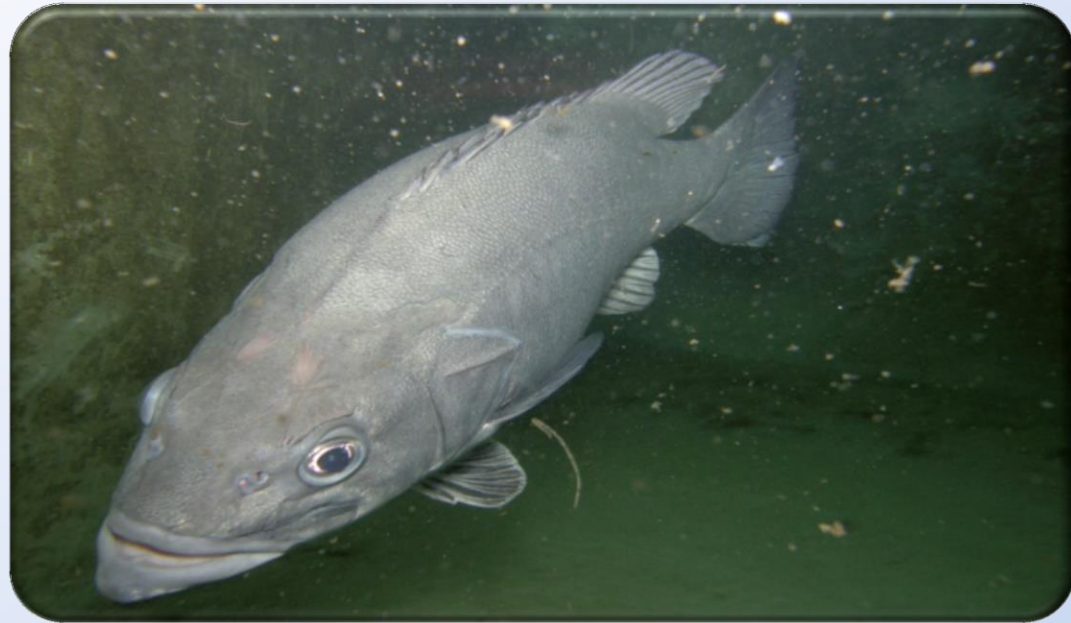


Wreckfish broodstock nutrition

Partners involved:

- ✓ CM: CIMA and IGAFA
- ✓ IEO (Oceanographic Center of Vigo)
- ✓ FCPCT (Las Palmas of Gran Canarias)



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Consellería do Mar. Xunta de Galicia.

July 2018



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

Objective and Tasks

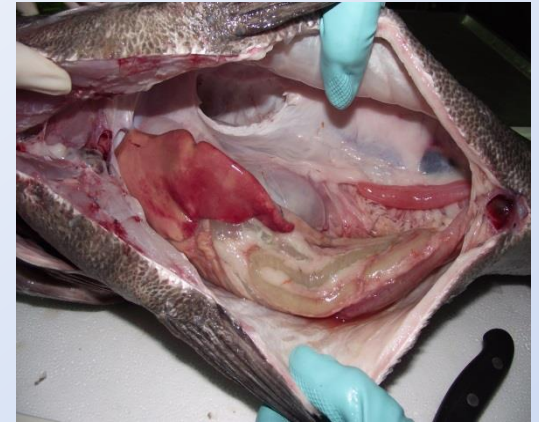
Determine the influence of broodstock feeds on fecundity and spawning quality

The study includes:

- ✓ A preliminary study about biochemical composition of some tissues of wild wreckfish and a comparison with the biochemical composition of tissues of intensive reared wreckfish
- ✓ The biochemical composition of different broodstock feeds with special attention to the fatty acid contents
- ✓ The effect of different feeding regimes based on fresh and commercial dry feeds on oocytes and eggs fatty acid composition
- ✓ The effect of feeding regime on fecundity and egg and sperm quality



Preliminary study: Composition of wild wreckfish tissues and a comparison with tissues of intensive reared wreckfish



Sampling: 91 fish were sampled from the Azores from February 2014 to April 2015

Objectives:

- To get some basic information about this species
- To advance in the knowledge of wreckfish nutritional requirements
- To identify potential nutritional deficiencies
- To formulate suitable diets for wreckfish broodstock



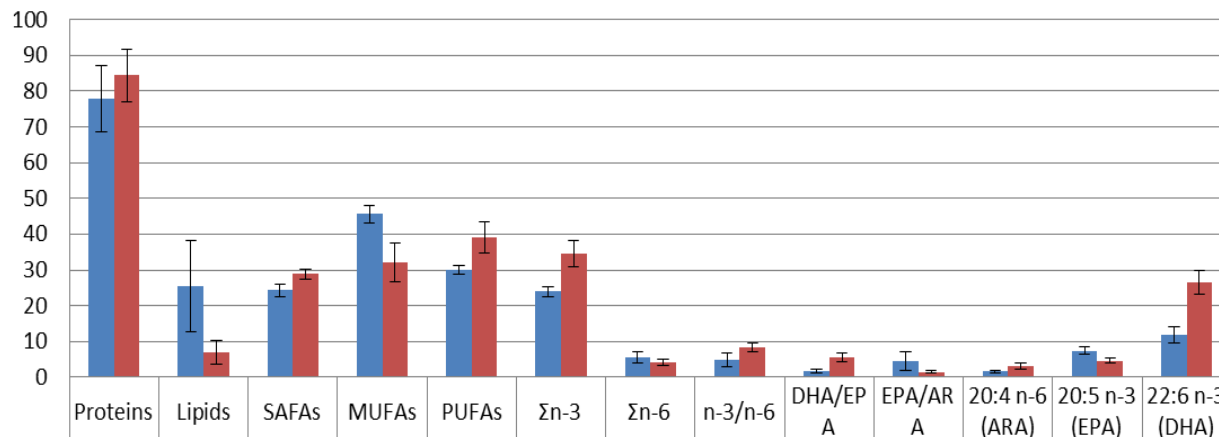
Biochemical composition of wild wreckfish tissues

	Muscle	Liver
Proximate analysis (% dry matter)		
Proteins	84.41±7.34	38.16±12.89
Lipids	6.92±3.39	39.34±15.03
Fatty acids content (% total fatty acids)		
SAFAs	28.83±1.28	26.48±5.21
MUFAs	32.09±5.43	55.64±10.72
PUFAs	39.08±4.41	16.66±8.88
Σn-3	34.51±3.75	14.01±7.69
Σn-6	4.08±0.81	2.44±1.29
n-3/n-6	8.50±1.18	5.48±1.75
20:4 n-6 (ARA)	3.11±0.79	1.48±0.92
20:5 n-3 (EPA)	4.55±0.70	2.90±1.54
22:6 n-3 (DHA)	26.38±3.33	8.69±5.37
DHA/EPA	5.69±1.23	2.92±0.96
EPA/ARA	1.54±0.37	2.02±0.75
PUFA.polyunsaturated fatty acids; ARA. arachidonic acid; DHA.docohexaenoic acid; EPA.eicosapentaenoic acid		



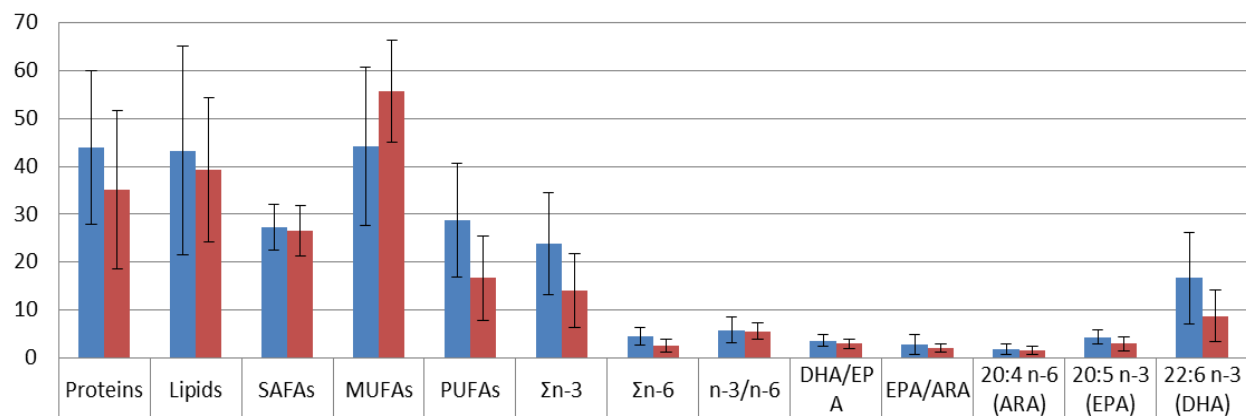
Biochemical composition: wild and reared wreckfish

Polyprion americanus



Muscle

Polyprion americanus



Liver

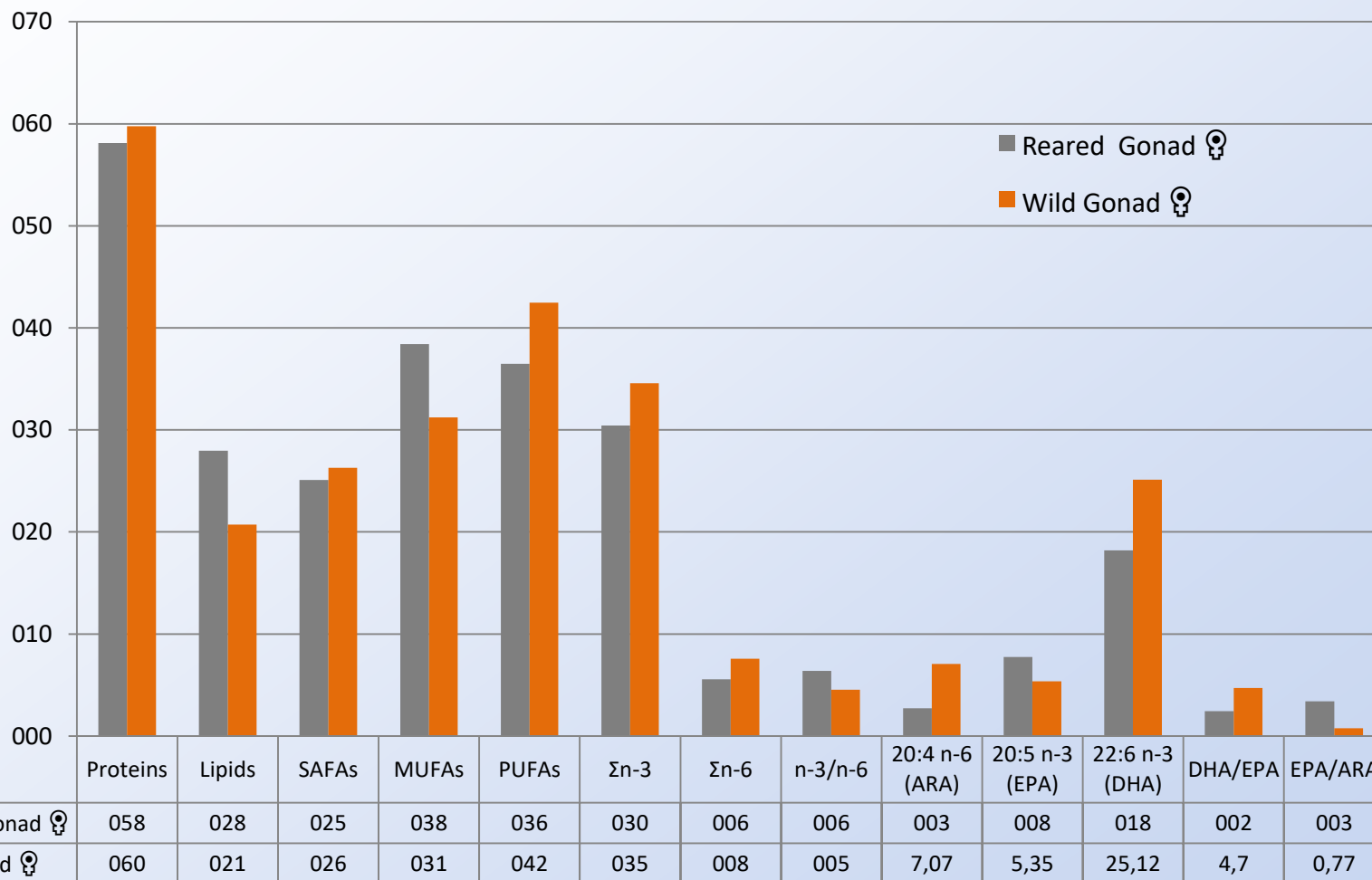


Biochemical composition of wild wreckfish mature gonads

	Wild ♀ Gonad	Wild ♂ Gonad
Proximate analysis (% dry matter)		
Proteins	59.77±10.50	44.02±3.77
Lipids	20.73±4.78	13.21±3.08
Fatty acids content (% total fatty acids)		
SAFAs	26.30±2.96	29.24±0.36
MUFAs	31.23±11.69	19.37±3.87
PUFAs	42.47±9.72	51.39±3.60
Σn-3	34.57±8.88	39.88±4.95
Σn-6	7.57±1.16	10.69±1.15
n-3/n-6	4.54±0.84	3.81±0.87
20:4 n-6 (ARA)	7.07±1.40	10.10±1.10
20:5 n-3 (EPA)	5.35±1.53	5.35±0.07
22:6 n-3 (DHA)	25.12±7.36	31.40±4.91
DHA/EPA	4.70±0.25	5.87±0.94
EPA/ARA	0.77±0.22	0.54±0.06



Females mature gonads Wild & Reared wreckfish



Feeding regimes for wreckfish broodstock

Ingredients

Dry food
%

<i>Fishmeal 70 LT FF Skagen</i>	25.000
<i>CPSP 90</i>	10.000
<i>Squid meal</i>	34.200
<i>Krill meal (Aker Biomarine)</i>	7.500
<i>Wheat Gluten</i>	7.000
<i>Wheat Meal</i>	7.250
<i>Tuna oil</i>	1.000
<i>Algatrium 70% DHA</i>	0.200
<i>Incromega DHA 500TG</i>	1.000
<i>VEVODAR</i>	1.300
<i>Vit & Min Premix PV01</i>	2.000
<i>Lutavit E50</i>	0.050
<i>Soy lecithin - Powder</i>	1.500
<i>Macroalgae mix</i>	1.000
<i>Antioxidant powder (Paramega)</i>	0.200
<i>Antioxidant liquid (Naturax)</i>	0.200
<i>SelPlex - Se yeast</i>	0.020
<i>Carophyll Pink 10% - astaxanthin</i>	0.050
<i>Nucleotides (Nucleoforce)</i>	0.030
<i>L – Taurine</i>	0.500
<i>Total</i>	100.000

Diets used for wreckfish broodstocks feeding

	2016	2017
<i>Stock IEO Tank S1 n = 5</i>	Semi-moist diet	Semi-moist diet
<i>Stock IEO Tank S2 n = 6</i>	Dry food	Dry food
<i>Stock IGAFA n = 10</i>	Squid	Hake/Squid



Semimoist diet:

- **14.8% blue fish**
- **14.8% white fish**
- **18% mussel**
- **17.6% squid**
- **34.8% fishmeal**



Protein and lipid composition (% dry weight) of broodstock wreckfish diets

<i>Diets</i>	<i>Proteins</i>	<i>Lipids</i>
<i>Semi-moist diet</i>	64.66	17.35 ± 2.45
<i>Dry food</i>	68.2	12.50 ± 0.53
<i>Hake</i>	82.5 ± 4.65	5.54 ± 0.46
<i>Squid</i>	57.54 ± 1.66	10.79 ± 0.38
<i>Hake/Squid</i>	62.93 ± 4.23	7.89 ± 4.23

High level of proteins in diets specially in Dry food (68%)
Hake/Squid has the lowest level of lipids (8%) and
Dry food has 12.5% of lipids and Semi-moist diet 17%



	<i>Semi-moist diet</i>	<i>Dry food</i>	<i>Hake/Squid</i>
SAFA's	29.96 ± 01.19	27.1 ± 0.21	29.92 ± 0.43
MUFA's	31.55 ± 0.97	26.01 ± 0.11	15.25 ± 0.78
PUFA'S	38.49 ± 1.65	46.89 ± 0.1	54.83 ± 0.97
Σn-3	29.40 ± 1.57	30.31 ± 0.14	49.44 ± 1.00
Σn-6	8.28 ± 0.56	16.25 ± 0.06	3.58 ± 0.01
n-3/n-6	3.56 ± 0.23	1.87 ± 0.01	13.81 ± 0.31
20:4n-6 (ARA)	1.25 ± 0.28	6.9 ± 0.12	2.73 ± 0.07
20:5n-3 (EPA)	8.84 ± 0.48	8.19 ± 0.11	14.27 ± 0.23
22:5n-3	1.29 ± 0.24	1.01 ± 0.14	0.97± 0.06
22:6n-3 (DHA)	15.91 ± 1.16	17.94 ± 0.05	32.55 ± 1.10
DHA/EPA	1.43 ± 0.66	2.19 ± 0.02	2.28 ± 0.09
EPA/ARA	1.86 ± 0.73	1.19 ± 0.04	5.22 ± 0.17

Hake/Squid has the highest level of PUFA(55%TFA), n-3 represents 49%TFA, EPA 14% & DHA 32%

Dry food has the highest level of n-6 (16%) and ARA (7%)

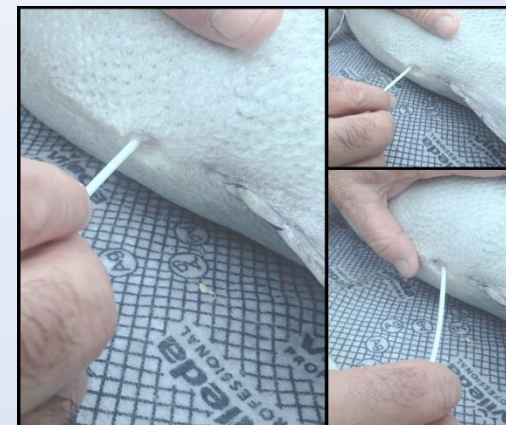
EPA/ARA is 1 in Dry food similar to tissues of wild wreckfish and 5 in Hake/Squid because of the high EPA content



Fatty acids of oocytes ($\emptyset >700 \mu$) from females fed with different diets (% total fatty acids)



Diets	Semi-moist diet	Dry food	Hake/Squid
Oocytes	OSM 0.710-2.033 μ	ODF 0.778-2.118 μ	OHS 0.831-1.388 μ
SAFA's	22.97 \pm 0.57	22.17 \pm 2.07	21.22 \pm 1.15
MUFA's	33.58 \pm 1.10	33.91 \pm 4.33	32.35 \pm 2.89
PUFA's	43.46 \pm 1.34	43.92 \pm 4.10	46.42 \pm 2.29
$\Sigma n-3$	36.87 \pm 1.59ab	31.35 \pm 3.31b	41.31 \pm 2.15a
$\Sigma n-6$	6.34 \pm 0.84b	12.32 \pm 1.21a	4.86 \pm 0.50b
n-3/n-6	5.90 \pm 0.93b	2.58 \pm 0.20c	8.63 \pm 0.90a
20:4n-6 (ARA)	1.88 \pm 0.36b	6.74 \pm 1.25a	1.61 \pm 0.99b
20:5n-3 (EPA)	7.57 \pm 0.19b	5.49 \pm 0.35c	10.72 \pm 1.13a
22:6n-3 (DHA)	24.60 \pm 1.53ab	21.32 \pm 1.90b	25.48 \pm 1.16a
DHA/EPA	3.25 \pm 0.15b	3.91 \pm 0.27a	2.40 \pm 0.18c
EPA/ARA	4.16 \pm 0.59b	0.86 \pm 0.10c	6.71 \pm 0.38a



There are not significant differences in SAFAS's, MUFA's and PUFA's of oocytes from females fed with the three diets

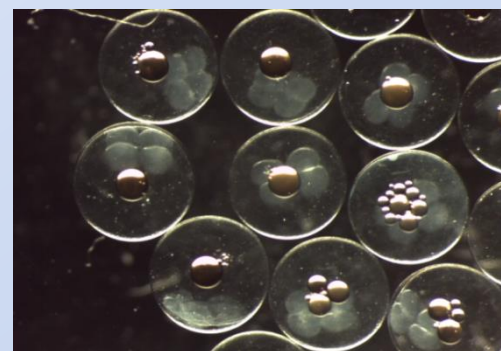
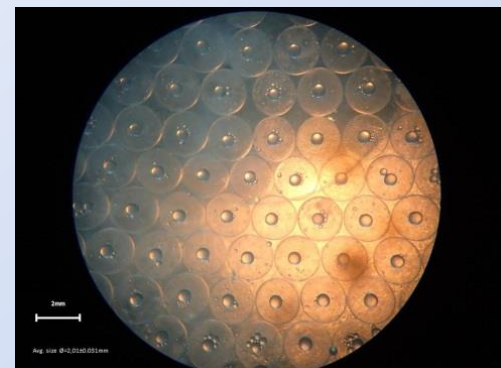
The highest content of n-3 was observed in OHS (41%), while OSM had 37% and ODF 31% of total fatty acids
n-6 PUFA values are much higher (12%) in ODF than in OHS and OSM (5-6%)

EPA/ARA is 0.86 in ODF and much higher in OSM(4) and OHS (7)

Lipid (% dry weight) and fatty acid composition (% total fatty acids) of eggs from females fed with different diets



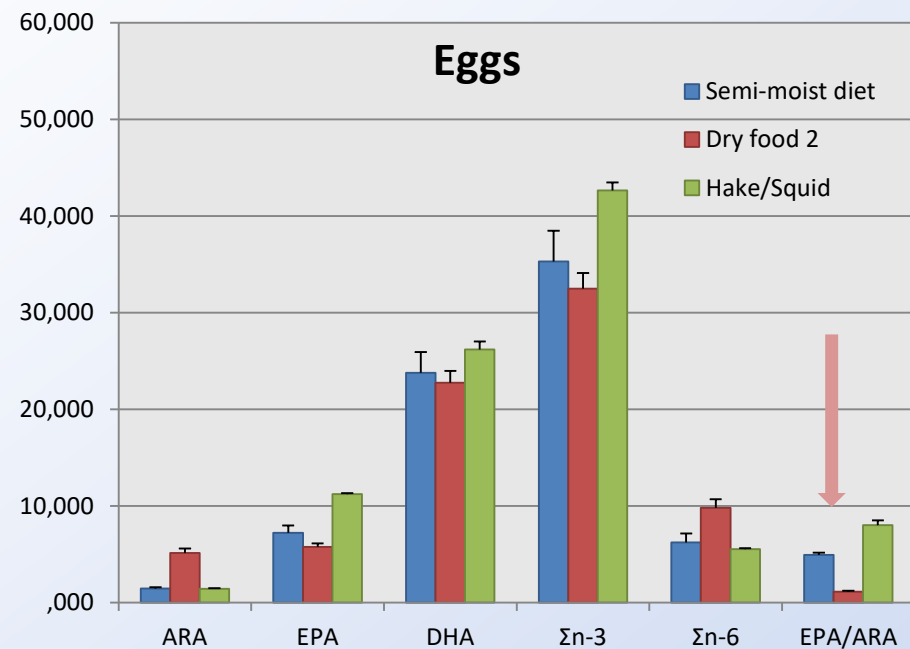
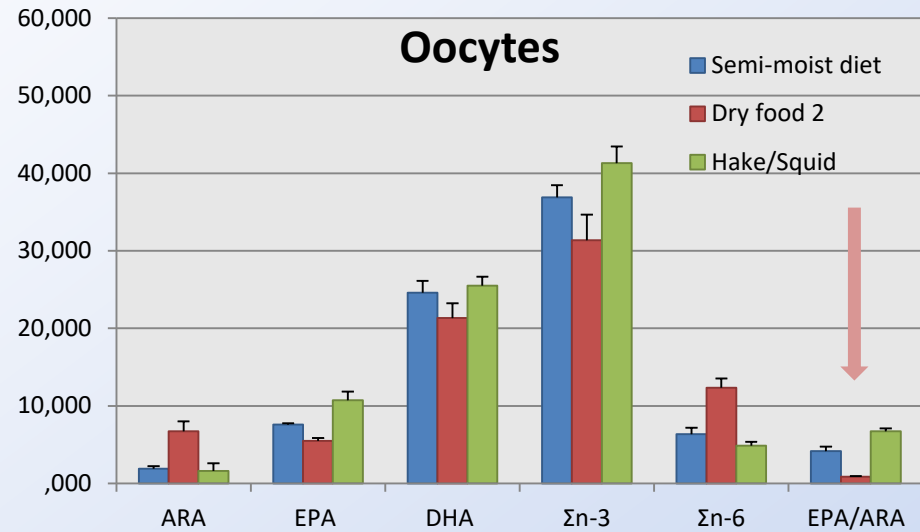
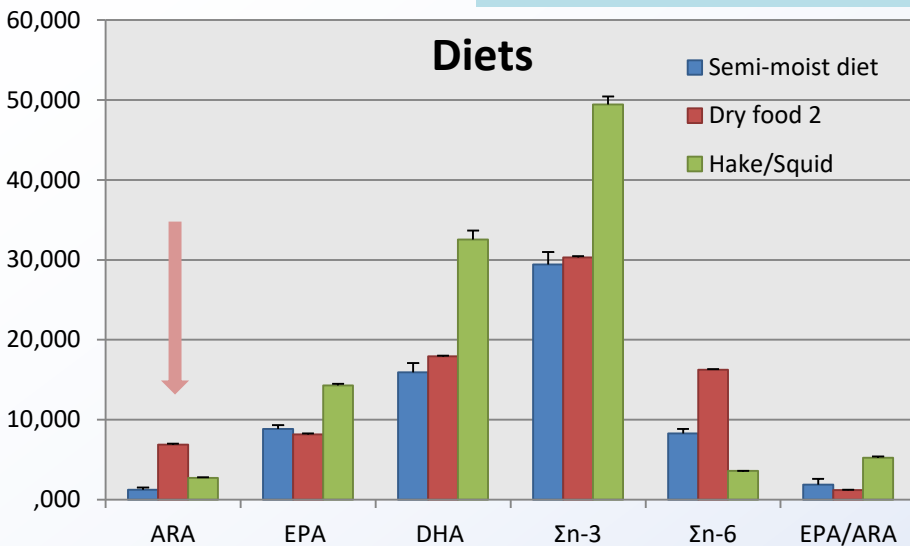
Diets eggs	Semi-moist diet OSM	Dry food ODF	Hake/Squid OHS
Lipids	24.65 ± 2.14a	18.94 ± 4.55a	9.40 ± 1.27b
SAFA'S	23.35 ± 0.96a	21.83 ± 1.29b	21.39 ± 0.66b
MUFA'S	34.93 ± 1.96a	35.10 ± 3.46a	30.11 ± 0.76b
PUFA'S	41.72 ± 2.38b	43.07 ± 2.64b	48.50 ± 0.72a
$\Sigma n-3$	35.29 ± 3.16b	32.48 ± 1.62c	42.64 ± 0.83a
$\Sigma n-6$	6.22 ± 0.93b	9.79 ± 0.91a	5.54 ± 0.10b
n-3/n-6	5.92 ± 1.76b	3.38 ± 0.39c	7.73 ± 0.31a
20:4n-6 (ARA)	1.46 ± 0.13b	5.12 ± 0.47a	1.41 ± 0.08b
20:5n-3 (EPA)	7.21 ± 0.76b	5.74 ± 0.39c	11.22 ± 0.10a
22:6n-3 (DHA)	23.76 ± 2.16b	22.73 ± 1.23b	26.17 ± 0.83a
DHA/EPA	3.30 ± 0.12b	3.6 ± 0.16a	2.33 ± 0.06c
EPA/ARA	4.92 ± 0.22b	1.13 ± 0.08c	7.99 ± 0.50a



The tendency of these results is similar to the one obtained in oocytes from females fed with the same diets

A correlation was found between the fatty acid profile of wreckfish feeding and eggs fatty acid profile

Fatty acid composition (n-3, n-6, DHA, EPA, ARA & EPA/ARA) of diets, oocytes and eggs



There is a clear relationship between the main fatty acid composition of diets, oocytes and eggs

The ratio of EPA/ARA in oocytes and eggs reflected the diet and was negatively correlated to dietary ARA content





Fatty acid composition (% total fatty acids) of sperm from males from different wreckfish broodstocks

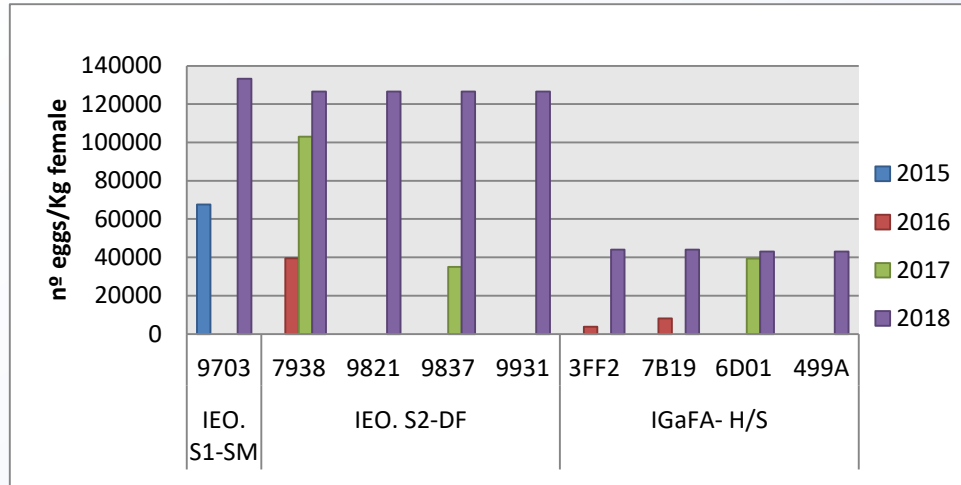
<i>Stocks sperm</i>	<i>Stock IGFA</i>	<i>Stock IEO</i>	<i>Stock MC2</i>
SAFA's	32.02 ± 1.92	32.56 ± 0.19	31.71 ± 1.96
MUFA's	10.82 ± 1.80	9.35 ± 0.79	9.81 ± 0.35
PUFA's	57.15 ± 0.60	58.10 ± 0.63	56.43 ± 1.93
Σn-3	51.50 ± 0.44	48.92 ± 4.54	49.86 ± 1.49
Σn-6	5.52 ± 0.62b	8.99 ± 3.97a	6.44 ± 0.45b
<i>n-3/n-6</i>	10.15 ± 1.96a	6.22 ± 2.62b	7.85 ± 0.28b
20:4(n-6) ARA	4.94 ± 0.75b	7.84 ± 3.85a	5.91 ± 0.40b
20:5(n-3)EPA	10.81 ± 1.12a	8.78 ± 1.76b	9.91 ± 0.36 ^a
22:6(n-3)DHA	36.67 ± 1.96	36.89 ± 2.96	36.47 ± 1.74
DHA/EPA	3.45 ± 0.53b	4.37 ± 0.78a	3.71 ± 0.29b
EPA/ARA	2.49 ± 0.70a	1.35 ± 0.69b	1.70 ± 0.16b



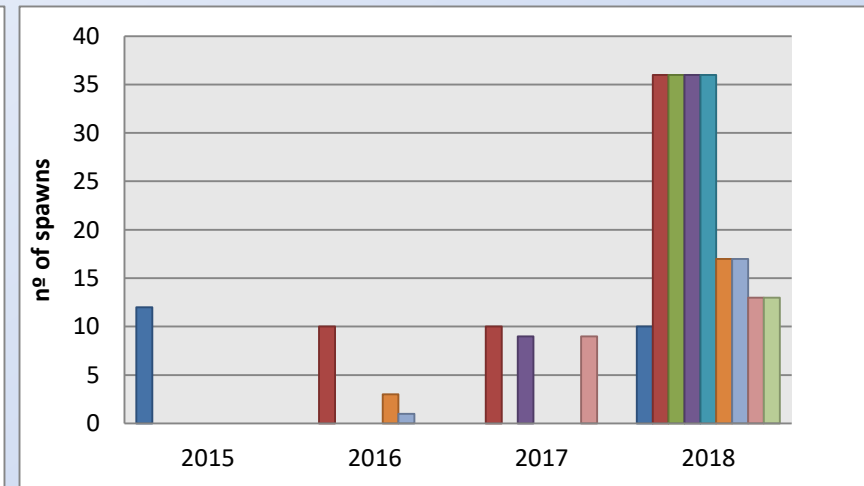
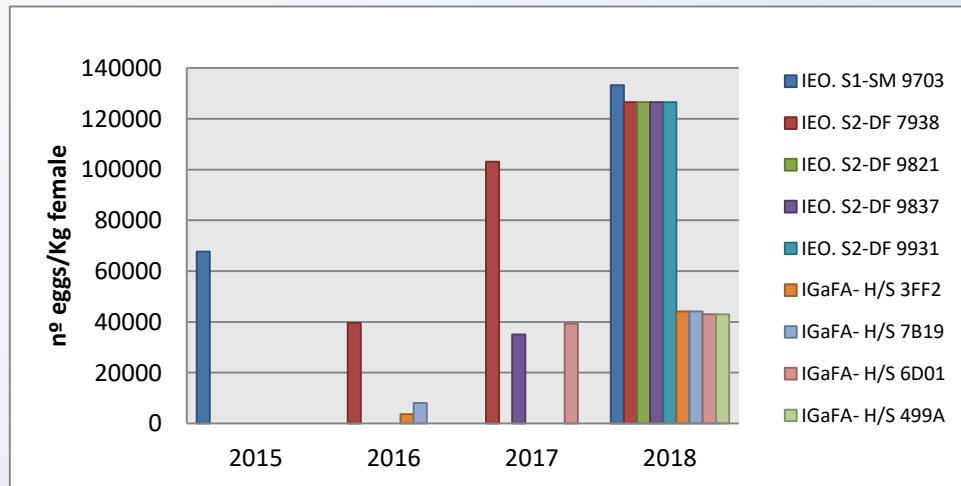
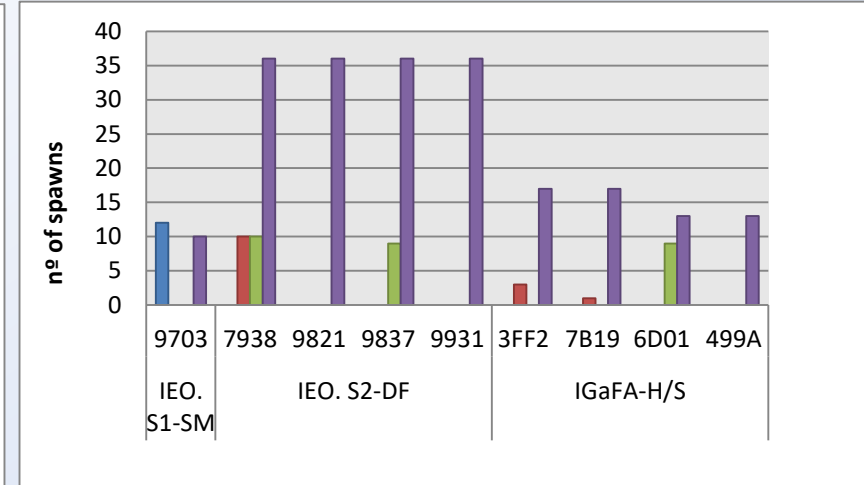
There are not significant differences in SAFA's, MUFA's and PUFA's in the sperm of males from the different stocks
 The n-3 PUFA content varies between 49-51% of the TFA and the n-6 PUFA content is higher in sperm from IEO stock (9%TFA) than in the rest of stocks

Fecundity of females and diet

**Relative fecundity
2015-2018**



**Spawns
2015-2018**



Relative fecundity and number of spawns per female have been increasing in females fed with dry feed over the years



SUMMARY

- ✓ Wreckfish has a big amount of proteins in muscle, 84% in wild fish and 78% in captive-reared fish
- ✓ Wild wreckfish have less lipids in muscle and liver than in muscle and liver from intensive culture and some differences were observed in the fatty acid profile between wild and reared wreckfish, PUFA and n-3 (%TFA) are higher in wild than in reared fish
- ✓ Gonads from females of wild wreckfish have a high level of ARA (7-10 %TFA) and EPA/ARA nearly 1
- ✓ A clear relationship between fatty acid profile of broodstock diet (semi-moisture, dry food and a mixture of hake and squid) and fatty acid profile of oocytes and eggs from females fed with these different diets was found
- ✓ Results obtained with dry food demonstrated that the wreckfish diet must contain a big amount of proteins, low level of lipids, a high amount of n-3 PUFA and the EPA/ARA ratio must be similar to the one obtained in wild females tissues
- ✓ First data of fatty acid profile of sperm from wreckfish males of different broodstock were obtained
- ✓ Relative fecundity (nº of eggs/Kg of female) and number of spawns per female have been increasing in females fed with dry feed over the years, from 2015 to 2018





Thanks for your attention!!!!

