

University of Lorraine, Nancy, France 2-4 February 2016

Minutes of the Annual Coordination Meeting 2016 (for Y3)



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Objectives

The objectives of the Annual Coordination Meeting (ACM) 2016 were to:

- (a) Present Scientific Discipline-specific summaries of the accomplished work during Y2 to the consortium members, as well as to a small number of invited guests,
- (b) Review and evaluate closely the work carried out in all Work Packages (WP) in the six Scientific Disciplines,
- (c) Plan the work to be implemented in all WPs in the following year,
- (d) Present the dissemination activities of the consortium (WP 31),
- (e) Inform the Partners of the 2nd Amendment that was submitted to the EU,
- (f) Organize the preparation of the Periodic Reporting (Scientific and Financial),
- (g) Have a meeting of the Steering Committee.

Description

The ACM 2016 was hosted by Dr. Pascal Fontaine of the University of Lorraine (P9. UL) and was held at the Museum-Aquarium of Nancy (Day 1) and the Brabois Campus of the University of Lorraine (Day 2 & 3) on 2-4 February 2016. The 3-day meeting was attended by 87 persons: 78 coming from the DIVERSIFY consortium and 9 invited guests from outside the consortium. No representative attended from three Beneficiaries (P26. GEI, P28. CANEXMAR and P37. EUFIC). Beneficiary P10. TU/e was unable to attend the first day of the meeting, but attended the second and third day.

As for the kickoff meeting, information regarding the meeting was uploaded continually on the project's web site (www.diversifyfish.eu/INTRA/Meetings & Activities/2016 Annual Coordination Meeting) to ensure that all participants had access to the most updated information. The Agenda (Tables 1 and 2) was developed with assistance from GWP leaders and consisted of:

- (a) DAY 1: a common session for all participants during DAY 1 (including invited guests) presenting summaries of the work implemented in all six Scientific Disciplines, specific presentations from various WPs or tasks, and presentations from invited guests,
- (b) DAY 2: Six (6) Scientific Discipline-specific workshops running in three parallel sessions during DAY 2, and
- (c) DAY 3: a common session dealing with Dissemination, Scientific and Financial Reporting, and Management. A meeting of the Steering Committee was also held at the end of the ACM. In addition, a special 2-hour meeting was held with all the Partners being involved in work with greater amberjack (*Seriola dumerili*) under any Scientific Discipline, in order to address some issues related to the grow-out experiments.

DAY 1 - Summary Presentations and invited guests

The morning session started with a welcoming presentation (Fig. 1) by the Project Coordinator (PC), Dr. C.C. Mylonas, presenting the Agenda for the meeting, welcoming the invited guests from outside the consortium and explaining the intentions of the consortium (as presented in the DOW, WP1 Project Management) for including other scientists and stakeholders in these ACMs. The invited guests included Dr. Pierrick Haffray and Anastasia Bestin from the Syndicat des Sélectionneurs Avicoles et Aquacoles Français (an animal breeding company), Dr Joao Correia and Mauricio Francisco from Flying Sharks (a fish capture and transport company), the secretary of the European Aquaculture Society Mr. Alistair Lane, Prof. Jonna Tomkiewicz from the Danish Technical University and members of the technical staff of four aquaculture production companies (Andromeda SA from Greece/Spain, Le Poisson du Soleill from France, Isidro de la Cal from Spain and Galaxidi SA from Greece).



Table 1. Agenda of DAY 1 of the Annual Coordination Meeting, which took place on the 2-4 February 2016, at the Museum-Aquarium of Nancy, France.

	y 2016,		Museum-Aquarium of		
DAY 1 Start	End	2 Feb	Tuesday Title	(Open Day - Summary presentation Presenter	os) Details
8.00	9.00		Registration	U of Loraine Staff	Register, receive badge, submit presentations
9.00	9.30		Welcoming	Constantinos Mylonas (HCMR), Pascal Fontaine UL)	Meeting logistics, agenda, welcoming from UL
9.30	10.00		GWP presentation - Repro & Genetics	Neil Duncan (IRTA)	
10.00	10.15		Dysfunctional reproductive maturation in captive greater amberjack	Aldo Corriero (UNIBA)	Reproduction & Genetics
10.15	10.30		Population genetic analysis of wild and domesticated pikeperch populations and their application to future breeding programs	Costas Tsigenopoulos (HCMR)	Reproduction & Genetics
10.30	11.00		GWP presentation - Nutrition	Daniel Montero (FCPCT)	
11.00	11.30	coffee			
11.30	12.00		GWP presentation - Larval husbandry	Bill Koven (IOLR)	
12.00	12.15		The nutrient profile of Artemia is greatly improved by ongrowing naulii for 3 days on Ori-Green	Kristin Hamre (NIFES)	Larval rearing
12.15	12.30		First larval rearing efforts with	Tito Peleteiro-Nikos	Larval rearing
12.30	13.00		wreckfish GWP presentation - Grow out husbandry	Papandroulakis-Antonio Vilar Nikos Papandroulakis (HCMR)	
13.00	14.00	Lunch	Lunch at a Restaurant "Cesar" a	nt Place Stanislas	
14.00	14.15		The effect of different stimuli on meagre feeding behaviour	Ioannis Papadakis (HCMR)	Grow out husbandry
14.15	14.30		Multifactorial approach to identify rearing conditions optimising growth, physiological status and immune defense in pikeperch	Patrick Kestemont (FUNDP)	Grow out husbandry
14.30	15.00		GWP presentation - Fish Health	Chris Secombes (UNIABDN)	
15.00	15.15		Pathologies of fish not included in the DIVERSIFY DOW	Pentelis Katharios	Fish health
15.15	15.45		GWP presentation - Socio economics	Gemma Tacken (LEI/DLO)	
15.45	16.00		Consumer value perceptions and attitudes towards farmed fish products in top-five EU markets	Marija Banovic and Thanassis Krystallis (AU)	Socieconomics
16.00	16.30	coffee			
16.30	16.45		Selection of new products and product development	Kriton Grigorakis (HCMR)	Socieconomics
16.45	17.00		Breeding selection in aquaculture fishes, with emphasis on the meagre	Pierric Hafray, SYSAFF	Invited guest
17.00	17.15		Capture of wild fish for aquaria and research	Joao Coreia (Flying Sharks)	Invited guest
17.15	17.30		European eel breeding, larval culture and first-feeding attempts	Jonna Tomkiewicz (DTU)	Invited guest
17.30	17.45		Greeting from the EU Officer of DIVERSIFY	Marta Iglesias (EU DG RTD)	EU Scientific Officer
17.45	18.00		Wrap up	Constantinos Mylonas (HCMR)	Agenda for next day
18.00	19.00		Visit the Aquarium	Pascal Fontaine	
20.00			Dinner at "Grande Salons Ho	tel De Ville, Stanislas Place"	

After the PCs introductory presentation (Fig. 1), the summary presentations started with Dr. Neil Duncan (P3. IRTA), the Group Work Package (GWP) leader for Reproduction & Genetics, presenting the overall objectives of the six (6) Work Packages (WP) in this Scientific Discipline, highlighting the important data obtained this second year of the project from each of the different WPs (Fig. 2). After the summary presentation (30 min) there were two 15 min presentations on the results obtained from WP 3 Reproduction & Genetics – greater amberjack (Dr. Aldo Corriero, P13. UNIBA) and WP 2 Reproduction & Genetics – pikeperch (Dr. C. Tsigenopoulos, P1. HCMR). Similarly, all the presentations from the GWP leaders explained the objectives of the WPs in each Scientific Discipline and provided an extensive summary of work implemented during the first year of the project, and the significant results that were obtained. The specific presentations from various other WP Lead Beneficiaries or Task leaders, allowed a more detailed presentation of the work that was carried out, as for example "The nutrient profile of Artemia is greatly improved by on-growing nauplii for 3 days on Ori-Green by Dr. K. Hamre (P17. NIFES), "Consumer value perceptions and attitudes towards farmed fish products in top-five EU markets" presented by M. Banovic (P11. UA), "Multifactorial approach to identify rearing conditions optimizing growth, physiological status and immune defense in pikeperch" from Mr. Baptiste Redivo, a graduate student from P16. FUNDP, and "Selection of new products and product development" by Dr. K. Grigorakis from P1. HCMR.

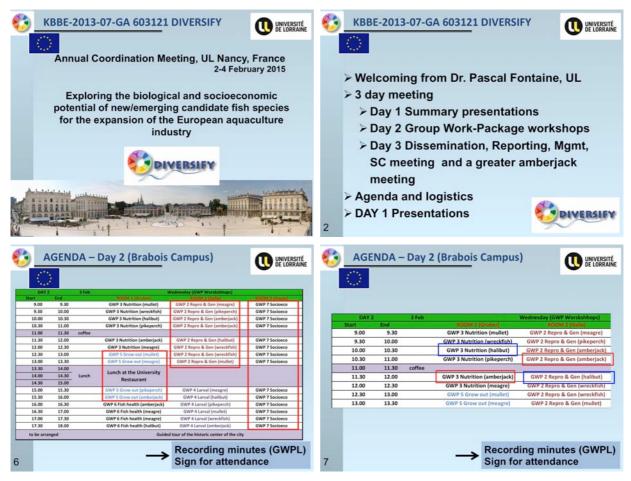


Figure 1. The opening slides for the Annual Coordination Meeting 2016, held by P9. UL, Nancy, France, explaining the Agenda of the meeting (upper right slide) and the slides explaining the organization of DAY 2 with the three parallel sessions, and the effort to organize the discussion in a way to allow most researchers to attend all sessions dealing with the species of their interest (lower slides).

The presentations from the invited guests, which followed the presentations from consortium GWP leaders and Partners, demonstrated both the interest of other organizations to participate in our ACMs and the interactions DIVERSIFY is trying to encourage with relevant researchers. Of great interest were the presentations of Dr. Pierrick Haffray (SYSAF, France) on the development of breeding programmes in aquaculture fish and of Prof. Jonna Tomkiewicz (DTU, Denmark) on the breeding and larval rearing research of Atlantic eel (*Anguilla anguilla*). Both presentations are extremely relevant to work undertaken in DIVERSIFY and we hope that we will establish further contacts with these researchers. The participation of four commercial aquaculture companies is also a clear indication of the relevance of DIVERSIFY to the EU industry, and the interest of their technical management to be updated with the current developments in the project. The connection with these companies also provides a means for DIVERSIFY to obtain relevant feedback from the sector, as well as having the potential to try some of the developed methodologies before the completion of the project and the release of the results.

Some of these companies, such as Andromeda SA and Galaxidi SA, continue to provide access to their facilities and fish stocks, and collaborate with DIVERSIFY as non-partners at no cost to the project. This ensures that expensive infrastructures and resources from outside the consortium are available to DIVERSIFY at no extra charge.

All presenting partners and invited guests agreed to have the presentations of the ACM 2016 available for the wider public, and they **have already been uploaded on the website** of the project, and are available to all interested stakeholders.



Figure 2. The opening slides from some of the presentations of some of the GWP leaders on DAY 1, including one presentation from an invited guest from outside the consortium (Prof. Jonna Tomkiewicz, DTU and Dr. Pierrick Haffray, SYSAF).



During the meeting, a professional company was hired to make a promotional video for the ACM 2016 of DIVERSIFY, which will be uploaded on our website and also examine the possibility of disseminating to various audiences (**Fig. 3**). Special 3-5 min interviews were given by the PC (Dr. C.C. Mylonas), the WP 31 Dissemination leader Dr. Rocio Robles, the host of the meeting from P9. UL Dr. Pascal Fontaine and the secretary of the European Aquaculture Society (EAS) Mr. Alistair Lane.



Figure 3. The crew video taping the proceedings of the meeting and Dr. Pascal Fontaine interviewing WP 31 leader Dr. Rocio Robles for the preparation of a promotional video.

After the completion of the presentations in DAY 1, all participants had dinner together at the beautiful hall of the Hotel de Ville (Municipality building), where they were treated to French cuisine and wine, with a special dishes of pikeperch (*Sander lucioperca*) and wreckfish (*Polyprion americanus*), two of the species of DIVERSIFY (**Fig. 4**).



Figure 4. The beautiful building and dinner hall of the Hotel de Ville (Municipality building) where dinner was offered by our host Dr. Pascal Fontaine and the University of Lorraine.



DAY 2 - Scientific Discipline-specific workshops

During the second day of the meeting, six Workshop Sessions were organized according to Scientific Disciplines with the objective of (a) reviewing and evaluating the work carried out and (b) planning the work to be implemented in the various scientific WPs during the third year (2016) of the project (**Table 2**).

Table 2. Agenda of DAYs 2 and 3 of the Annual Coordination Meeting, which took place on the 2-4 February 2016, at the University of Lorraine, Nancy, France.

DAY 2		3 Feb		Wednesday (GWP Worskshhops)	
Start	End		ROOM 1 (Gruber)	ROOM 2 (Galle)	ROOM 3 (Daum)
9,00	9,30		GWP 3 Nutrition (mullet)	GWP 2 Repro & Gen (meagre)	GWP 7 Socioeco
9,30	10,00		GWP 3 Nutrition (wreckfish)	GWP 2 Repro & Gen (pikeperch)	GWP 7 Socioeco
10,00	10,30		GWP 3 Nutrition (halibut)	GWP 2 Repro & Gen (amberjack)	GWP 7 Socioeco
10,30	11,00		GWP 3 Nutrition (pikeperch)	GWP 2 Repro & Gen (amberjack)	GWP 7 Socioeco
11,00	11,30	coffee			
11,30	12,00		GWP 3 Nutrition (amberjack)	GWP 2 Repro & Gen (halibut)	GWP 7 Socioeco
12,00	12,30		GWP 3 Nutrition (meagre)	GWP 2 Repro & Gen (wreckfish)	GWP 7 Socioeco
12,30	13,00		GWP 5 Grow out (mullet)	GWP 2 Repro & Gen (wreckfish)	GWP 7 Socioeco
13,00	13,30		GWP 5 Grow out (meagre)	GWP 2 Repro & Gen (mullet)	GWP 7 Socioeco
13,30	14,00		Lunch at the University		
14,00	14,30	Lunch	•		
14,30	15,00		Restaurant		
15,00	15,30		GWP 5 Grow out (pikeperch)	GWP 4 Larval (meagre)	GWP 7 Socioeco
15,30	16,00		GWP 5 Grow out (amberjack)	GWP 4 Larval (halibut)	GWP 7 Socioeco
16,00	16,30		GWP 6 Fish health (amberjack)	GWP 4 Larval (pikeperch)	GWP 7 Socioeco
16,30	17,00		GWP 6 Fish health (meagre)	GWP 4 Larval (mullet)	GWP 7 Socioeco
17,00	17,30		GWP 6 Fish health (meagre)	GWP 4 Larval (wreckfish)	GWP 7 Socioeco
17,30	18,00		GWP 6 Fish health (halibut)	GWP 4 Larval (amberjack)	GWP 7 Socioeco
to be arra	nged		Guide	d tour of the historic center of the city	

DAY 3		4 Feb	Thursday	(dissemination-reporting-administra	ntion)
Start	End		Title	Presenter	Details
9,00	9,30		WP31 Dissemination	Rocio Robles	
9,30	10,00		WF 31 Dissemination	Nocio Nobies	
10,00	10,30		Amendments (2nd), Reporting	Constantinos Mylonas	
10,30	11,00		Amenaments (zna), Reporting	constantinos irryionas	
11,00	11,30	coffee			
11,30	12,00		Deliverables, Participants Portal	Constantinos Mylonas	
12,00	12,30		Deliverables, Farticipants Fortai	constantinos irryionas	
12,30	13,00		Steering Committee meeting	Coordinator, GWP leaders, SME representatives (ARGO, ASIALOR) APROMAR	
13,00	14,00	Lunch	Lunch at the University		
13,00	14,00	Lunch	Restaurant		
14,00	14,30			Species Leader (Nikos	Address issues related to the
14,30	15,00		Greater amberjack meeting (Room	Papandroulakis) & greater	implementation of the large
15,00	15,30		Cuenot)	amberjack partners	scale grow-out experiments
15,30	16,00			amberjack partiters	state grott out experiments
18,00	22,00		Social event (to be arranged)	Pascal Fontaine	

The workshops of DAY 2 were running in parallel (3 Scientific Disciplines at a given time) in an attempt to minimize the potential time conflict for most Beneficiaries. The duration of each session was decided by the GWP leader based on the number of WP included in the Scientific Discipline, as well as the amount of work

that needed to be presented and discussed, and the workload expected for the upcoming year. Therefore, GWP Socioeconomics requested a full-day Workshop, so a room was dedicated to their work. In addition, the Workshops were organized in a way that the WPs dealing with the same species were planned at different times during the Workshops, to allow all scientists attending all the WPs of the same species (**Table 2**). This was also achieved, to a degree, by the participation to the ACM 2016 of more than one scientist from some of the beneficiaries that are involved in many GWPs. For example, P3. IRTA was represented by eight researchers and HCMR by nine.

Details of the different Workshops (Fig. 5) are provided in the following pages, in the minutes prepared by the GWP leader of each Scientific Discipline.



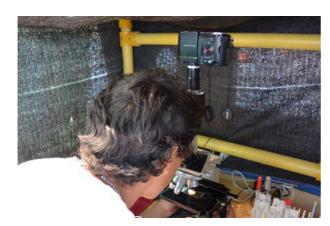




Figure 5. Photos from the DAY 2 Workshops of the GWPs Reproduction & Genetics (upper left), Socioeconomics (upper right) and Larval Husbandry (bottom).

Minutes of GWP Reproduction and Genetics workshop

Annual Coordination Meeting, Day 2 (3/2/2016, 9:00-13:30)



By Dr. Neil Duncan, IRTA (GWP Leader)

Participants

N°	Name	Lastname	Partner N°	Partner
1	Constantinos	Mylonas	1	HCMR
2	Costas	Tsigenopoulos	1	HCMR
3	Ioannis	Fakriadis	1	HCMR
4	Neil	Duncan	3	IRTA
5	Hanna	Rosenfeld	4	IOLR
6	Torstein	Harboe	7	IMR
7	Blanca	Alvarez-Blázquez	8	IEO
8	Jose Benito	Peleteiro	8	IEO
9	Salvador	Jerez	8	IEO
10	Virginia	Martín	8	IEO
11	Virginia	Martín	8	IEO
12	Alain	Pasquet	9	UL
13	Bérénice	Schaerlinger	9	UL
14	Pascal	Fontaine	9	UL
15	Aldo	Corriero	13	UNIBA
16	Christian	Fauvel	14	IFREMER
17	Covadonga	Rodríguez	15	ULL
18	José A.	Pérez	15	ULL
19	Kristin	Hamre	17	NIFES
20	Fátima	Linares	19	CMRM
21	Tasos	Raftopoulos	23	ARGO
22	Antonio	Vilar	32	MC2
23	Popi	Tsakoniti	Invited	Galaxidi

Summary of progress

Progress has been made on the reproduction and genetics tasks for all six species. Meagre is a species for which genetic improvement programmes need to be established. The genetic variation of breeders in the industry indicates that although the status of the existing stocks is healthy, care is needed in forming base

populations and managing crosses to produce families. Families can be produced using paired spawning, and a large number of genetic markers (microsatellites and SNPs) are now available. When these are associated with phenotypes they will facilitate breeding programmes. Similarly, pikeperch held for aquaculture have similar genetic variation compared to wild populations and careful management can use these stocks to form breeding programmes. Greater amberjack have been successfully spawned in facilities in both the Mediterranean and the Canary Islands. In the Mediterranean, gonadotropin releasing hormone agonist (GnRHa) has induced spawning in cages, but not in tanks, as females never reached an appropriate maturation stage. In the Canary Islands, natural and GnRHa induced spawning was obtained in tanks. The reproductive dysfunction of wild greater amberjack held in captivity for 4+ years in the Mediterranean has been described and compared to wild fish that were sampled at the moment of capture. Captive greater amberjack stocks had smaller gonads (GSI), higher incidence of atresia (females), higher incidence of germ cell apoptosis (males) and lower contents of polar lipids, docosahexaenoic and arachidonic acid.

Atlantic halibut hatched and reared in captivity had smaller and more frequent batches of eggs with poorer percentage fertilisation compared to captive wild halibut. The use of GnRHa implants synchronised egg batches and increased the size of the egg batches. Wreckfish exhibited advanced stages of maturation and some spontaneous spawning in captivity. Especially, sperm production was good from males and sperm management (sperm characteristics, cryopreservation and cool storage) protocols were developed. Application of GnRHa was successful in inducing ovulation. However, egg quality has been variable with many incidences of unviable eggs being collected and just a few fertilised spawns. The combination of induced ovulation and *in vitro* fertilisation is an approach that has given some success and that will be developed. Grey mullet sperm characteristics and management were described. The production of grey mullet recombinant gonadotropins (r-GtH) was developed and r-follicle stimulating hormone (r-FSH) induced a greater proportion of grey mullet to mature to the late stages of gametogenesis. The fish induced to more advanced stages of gametogenesis also gave higher responses and egg quality to induced spawning with GnRHa and dopamine antagonists. The DIVERSIFY project is on track to provide solutions to the identified bottlenecks in the area of Reproduction and Genetics for the six species.

WP2 Meagre

Outstanding deliverables

- **D2.4** Identification of genetic markers related to growth for use in marker assisted breeding programmes for meagre (M36, HCMR)
- **D2.5** Genetic characterisation of fast and slow growing meagre (M36, HCMR)
- **D2.6** Description of sperm characteristics and cryopreservation protocol of meagre sperm (M36, IFREMER)
- **D2.7** Protocol for the strip spawning of meagre females and *in vitro* fertilization (M36, IRTA)

D2.4 and D2.5 The deliverable 2.2 has been delivered, providing meagre's transcriptome for the liver and muscle tissues; moreover, thousands of microsatellites and SNPs related to genes that were expressed in meagre juveniles from six families and with different growth rates were identified. The plan is to genetically analyse large and small fish (fast and slow growers) at harvest from two cages containing either large grade of juveniles or small grade of juveniles from the same group of families. Fin clip samples and measures of weight and length have been taken from 400 fish in the cage of the 'large grade fish' by IRTA at the facilities of ANDROMEDA, Spain (a non-consortium collaborator) and sent to HCMR. The cage containing the 'small grade fish' will be sampled soon, hopefully during February 2016. The deliverable is expected to be on time.

ANDROMEDA asked about the results of the genetic analysis of their broodstock, which has not been given to them as agreed with P2. FCPCT. No representative of FCPCT was present in the room to clarify this issue. We were unsure of the situation with these samples, so the GWP coordinator and if necessary the PC will contact P2. FCPCT (Dr. Juan-Manuel Afonso) to ensure the results are delivered to ANDROMEDA.

A number of articles will be prepared from these Deliverables, including the one demonstrating the effectiveness of multiple GnRHa injections to induce reliable spawning (Dr. CC Mylonas, P1. HCMR) and the one with the paired spawning with male rotation (Dr. N. Duncan, P3. IRTA).

- **D2.6**: Work is on time and the data is sufficient for the deliverable. We will program visits between IFREMER and IRTA to train for handling sperm for *in vitro* fertilisation work.
- **D2.7** Initial IRTA trial gave preliminary *in vitro* results. We will induce 2-4 meagres each week during May-June 2016, in order to determine optimal stripping time for meagre females. Work with IFREMER to standardise sperm management. Suggested to look at relation of timing of hormone application and timing of stripping.

WP3 Amberjack

Outstanding deliverables

- **D3.2** Establishment of hormone specific ELISAs for measuring LH, FSH and leptin in greater amberjack (M30, IOLR)
- **D3.4** Establishment of a Computer Assisted Sperm Analysis (CASA) for the evaluation of greater amberjack sperm (M32, IFREMER)
- **D3.5** Description of the process of oogenesis in captive greater amberjack, including (a) aspects of growth and body indices, (b) histological evaluation of ovarian development, (c) pituitary levels of FSH (M46, HCMR)
- **D3.6** Description of the process of spermatogenesis in captive greater amberjack, including (a) aspects of growth and body, (b) histological evaluation of testicular development, (c) pituitary level.... (M46, HCMR)
- **D3.7** Comparative effectiveness of a GnRHa injection vs GnRHa implant treatment for the induction of spawning of greater amberjack in the eastern Atlantic (M48, FCPCT)
- **D3.8** Dose response of GnRHa implant therapy for the induction of spawning in F1 generation broodstock of greater amberjack in the eastern Atlantic (M54, IEO)
- **D3.9** Development of a spawning induction therapy for captive reared broodstock in the Mediterranean Sea based on the use of GnRHa in the correct mode of administration (hormone/implant), dose and timing of administration (M54, HCMR)
- **D3.10** Method for inducing spawning and collecting greater amberjack eggs in sea cages (M54, HCMR)
- **D3.2** has been delayed. The problem was leptin gene expression. The GtH assays have been established. The deliverable should be submitted as soon as possible, definitely before mid-term project review \sim September. The deliverable could be submitted without leptin results and resubmitted when results are available, but it is preferable to delay the Deliverable to early summer and submit it completed (June-July 2016).
- **D3.3** Delivered, but some work could be improved with information on age / size of maturity. Make some more samplings on smaller wild fish for comparison of gonads. Nutrition work will continue and be improved. There were eggs from successful spawning and the nutrition could be analysed to make comparison between different feeding regimens and the gonads from the dysfunctional group. This year we will collect more eggs from the two sea cage stocks (ARGO and GALAXIDI), one feeding on Skretting dry feed (Vitalis Cal) and the other one on live seabream/sea bass.

The comparison of the two farm cages gave different results. Repro cycle cage was dysfunctional while the other cage was in advanced stages and responded well to induced spawning. The fish were the same and were treated in the same way. Information and photos from spawning induction cage will be compared with the samples from the dysfunctional cage.

- **D3.4** Work in progress and will be delivered on time. IFREMER and HCMR will work together to coordinate more samplings and write the deliverable.
- **D3.5** and **D3.6** Already have almost all the data. Nutrition work will be continued. Deliverables will be on time.
- **D3.7** Eastern Atlantic stock produced good results, sufficient for the Deliverable and expected publication and PhD thesis. Work done.
- **D3.8** F1 spawning induction in May, June, July 2015, with very good results. Although in the DOW the proposal was to look at 3 doses, due to a mortality in 2014 just 9 F1 fish are left. Options are to repeat doses tested in 2015 or try different doses. Conflict as need to obtain eggs for larval rearing, should focus on different dose and possibly return to "successful" doses if eggs are not obtained. Last year we used a dose of $50 \mu g/kg$, so this year we will use the higher does of $75 \mu g/kg$ (as opposed to the lower) to ensure that we will get eggs (based on last year's results).
- **D3.9** In general, reproductive control is not working in tanks. Fish do not mature very well, with many fish having only primary oocytes and others poor vitellogenesis. Completely the opposite is observed in cages, which present late stages of gametogenesis and even late stages of oocyte maturation. Will work to stock fish from cages into tanks to see if this enables to achieve good spawning in tanks, for commercial production.

Suggested to stimulate earlier – mid gametogenesis with low dose of GnRHa implant aprox 10-15% of dose needed to stimulate spawning. We will try this, in one of the 2 captive stocks at HCMR. In general good sperm availability, but some difficulties to strip sperm. May be due to abdomen muscles but more likely due to low GSI.

D3.10 Cage spawning at ARGO, GALAXIDI and HCMR, with 1 cage in each site. Cage set up lower depth and curtain to aid egg collections. Very few or no eggs collected. Fish spawn low in cage, eggs are not strongly buoyant and eggs probably lost with currents. A cage was closed with a bag and this allowed collection of spawned eggs. This is an interesting approach, but it can result in problems of water exchange and oxygen levels and death of fish. Therefore, must develop semi-closed system that enables water exchange and collection of eggs. This will certainly be done at GALAXIDI, and perhaps also in HCMR.

WP5 Atlantic halibut

Outstanding deliverables

- **D5.1** Documentation of reproductive performance in wild-captured vs cultured female Atlantic halibut (M30, IMR)
- **D5.2** An optimised GnRHa therapy protocol to improve spawning performance of F1/F2 Atlantic halibut, and to increase availability of eggs of stable and predictable quality (M30, IMR)
- **D5.3** Identification of potential disturbances in reproductive development in F1/F2 Atlantic halibut females (M36, IMR) A 12 month delay has already been requested to allow the use of a new stock.
- **D5.1** Documentation of different stocks. Comparison of domesticated wild fish with farmed fish. Farmed fish smaller, more batches, lower interval between spawning. Total fecundity lower in spawning stocks, relative fecundity the same, smaller egg batches from smaller fish in farmed. Farmed fish must be stripped or eggs are retained and go to atresia. Domesticated wild fish will liberate eggs if not stripped. Fertilisation was lower and more variable in farmed fish. More egg quality characteristics will be obtained in 2016 to complete deliverable.
- **D5.2** GnRHa experiment in 2014. Synchronised ovulations into a shorter period and provided more eggs. Final trial is being made for deliverable M30. **Stirling White Halibut may close down or be sold**. Should not affect completing the deliverable, but we are awaiting for developments.

D5.3 Delayed and expected to be completed during 2016-2017. New wild fish are being captured for work to complete deliverable.

WP6 Wreckfish

Tito's last presentation, as he is retiring this month!

Outstanding deliverables

- **D6.3** Spawning induction methods with in vitro fertilization of wreckfish (M36, IEO)
- **D6.4** Establish reliable collection methods and protocols to form new wreckfish broodstocks (M36, CMRM)
- **D6.5** Description of the reproductive cycle of wreckfish (M48, IEO)
- **D6.6** An in vitro fertilization protocol to be employed by the industry to spawn wreckfish (M48, IEO)
- **D6.7** Spawning induction method for spontaneous spawning of wreckfish in large tanks (M54, HCMR)
- **D6.4** Capture of wild fish deliverable due this year. Two fish caught and acclimated in 2015. Contact with sport fisherman, who is catching a lot of small wreckfish and there is the possibility to collaborate and make a good capture to complete the deliverable.
- **D6.5** Reproductive cycle in progress and on time. Many small stocks. Problems with males fighting. Breeders swapped between centres to improve numbers of mature breeders in each centre. Worked with echography to examine ovaries. Different diets being used. Reduced fat content to reduce fat accumulation in fish.

Spawning from March to June. First spawning in IEO and then MC2 (Aquarium A Coruna). Three stripspawns made. Large oocytes $1000-1200~\mu m$ in maturing females during spawning season. No increase in oocyte size in non-maturing fish in IGAFA and part of the stock in IEO. Very prolonged period with sperm, only 3-4 months of the year without sperm. Concentration high $(x10^{10})$ during the spawning season of March-June.

Need to obtain pituitaries from mature and immature fish to send to IOLR. Blood samples taken in association with different stages of maturity. Continue sampling this year and send all samples to HCMR and IOLR.

D6.3, **D6.6** due this year. Spawning, hormone induced, *in vitro*.

IEO, CMRM and MC2 (Spain): Three females 27 kg +1100 μ m oocytes, 30 kg +1000 μ m oocytes, 16.5 +950 μ m oocytes implanted (500 μ g per fish), no ovulation. **Oocytes too small and dose to low**. Implant at 1300 μ m oocytes and at 50-100 μ g GnRHa per kg.

Need to identify time when fish have $1300+ \mu m$ oocytes and make induction trial with adequate dose (minimum $50~\mu g/kg$). Strategy should be to select females that have a history of large oocyte size and maintain in good conditions without sampling until the correct time for induced spawning with maximum oocyte size. In HCMR this should be done during April-May. Worry or problem is to obtain fish with 1300 μm oocytes at IEO and IGAFA.

HCMR (Greece): Vitellogenesis Feb-May. Spawns during April and May. Made strip spawning and obtained fertilisation. Another attempt will be done this year at HCMR, and hopefully will be able to start preparing the deliverable.

Will monitor stocks in aquarium, IEO and IGAFA.

Deliverables: Should be possible with results obtained by the end of 2016 from all partners. D6.6 is a protocol on induced spawning and *in vitro* fertilisation for the industry, and must be produced.

WP7 Grey mullet

Outstanding deliverables

D7.5 Description of the process of oogenesis in captive-reared vs hatchery-produced grey mullet, including aspects of growth, body indices, and histological evaluation of ovarian development (M48, UNIBA)

D7.6 Culture procedure that identifies the on-growing period for the production of grey mullet roe (bottarga) from wild and hatchery juveniles (M54, IOLR)

D7.7 Development of a breeding protocol for captive reared grey mullet broodstock based on optimized hormonal treatment, group structure and photo-thermal regime (M60, IOLR)

Good advances with development of bio-active r-FSH and r-LH. Hormone combinations of dopamine antagonist (females) or methyl testosterone (males) + r-FSH increases the number of fish maturing to advanced stages of gametogenesis. Combinations of GnRH and dopamine antagonist induce spawning. Deliverables D7.1-D7.4 delivered. Sperm protocols in place. More egg transport data needed to update deliverable D7.4.

Problems with spawning:

50-70% of females do not ovulate and spawn

Variable fertilisation

For the 2016-2017 season, the effort will be to improve spawning induction treatments, photoperiod and temperature manipulation. Obtain multiple spawning.

Look at group structures effect; fish selected from different groups based on good gonad development may not spawn well together. Plan to look at spawning inductions within established groups.

Minutes of GWP Nutrition workshop

Annual Coordination Meeting 2016, Day 2 (3/2/2016, 9:00-12:00)



By Dr. Daniel Montero, P2. FCPCT (GWP Leader)

During this workshop for each species we reviewed and discussed the progress achieved in 2015, pointing out to potential problems found, and deviations from the DOW. Besides, an update on the status of the deliverables was also commented, as well as interactions among partners and WPs.

Nutrition for grey mullet (WP13) Highlights:

The results on this WP were presented by Bill Koven (IOLR), and were based on previous results showed in the ACM 2014, with the aim to test if the taurine (Tau) growth advantage found early during larval rearing continued after 44 dph. An experiment was conducted with animals from rotifer treatments with different levels of taurine, showing that feeding starter diets with taurine at the fry stage allowed growth compensation in the smaller fish. A second experiment was conducted with grey mullet juveniles to study the effect of dietary taurine on growth. Although fish are omnivorous at this stage, 0.5% taurine DW diet improved juvenile growth.

Some analysis in progress:

- Task 13.1 (IOLR) Fatty acid and taurine analyses of muscle, eyes and liver, based on previous results obtained from white grouper, that Bill Koven discussed during the workshop.
- Sub-task 13.2.1 (IOLR) Determine expression of Tau rate limiting enzyme; cysteine sulfinate decarboxylase (CSD) at various stages (larval and grow out). The hypothesis is if this enzyme changes from carnivorous larva to herbivorous juvenile, as most of the carnivorous species do not have this enzyme, as Bill discussed.
- Sub-task 13.2.2 (IOLR) Determine expression of rate limiting enzyme of bile salt synthesis, cholesterol 7α-hydroxylase (CYP7A1) at various stages (larval and grow-out).

Problems and deviations from the plan:

As problems occurred during 2015, Bill Koven pointed out that during 2015 large spawns (liters) of unfertilized eggs were obtained. In 2015 only a few thousands of fish were produced, resulting in an important delay on the deliverables. As showed in the table, month due for deliverables D13.1 and D13.2 was 18. Due to the problems described above, both deliverables have been delayed and month 36 has been proposed as new month due for both of them.

Deliverable	Title	Month due

D13.1	Determine changes in the essential fatty acid requirement as a function of developmental stage and ambient salinity in grey mullet	18
D13.2	Determine a developmental stage ability to synthesize key enzymes in Tau and bile acid synthesis in grey mullet	18

Plans for 2016:

- Sub-task 13.1.1 (IOLR) Effect of DHA/EPA ratio on larval and juvenile performance during rotifer and *Artemia* feeding from carnivorous larva to herbivorous (omnivorous) juvenile. Will use the best performing Tau diet.
 - D13.1 (IOLR) Determine changes in the essential fatty acid requirement as a function of developmental stage in grey mullet.
- Sub-task 13.3.1 (IOLR) Effect of DHA/EPA ratio in non-fish meal grow-out diets on fish performance.
- In addition, an analysis of hatchery control diet needs to be conducted to determine effective missing nutrients, as discussed based on the better results obtained within animals from this experimental group in contrast to those from Tau treatments.

Nutrition for wreckfish (WP12) Highlights:

The second species treated was the wreckfish, presented by Fátima Linares (CMRM). The first task (task 12.1) deals with live prey enrichments. Some wreckfish larvae from different spawnings of IEO and MC2 broodstocks were obtained, but a low survival rate at hatching was obtained and mortality was 100% at 20 days after hatching, and whole experiments could not be conducted with those larvae. However, some samples of larvae were taken out on days 0, 5 and 10 of life, to obtain the fatty acid profile of wreckfish larvae. Fatty acid profile has little variation in the first 10 days of life of wreckfish. Some comments were done (Kristin Hamre) on the special care to be taken with Selenium content in enrichment for this species.

In the second task (task 12.2), that deals with broodstock feeding regimes, a comparative study on the composition of wild fish *vs* captive wreckfish broodstocks was conducted. Analysis of tissues of cultured wreckfish and comparison with those obtained from wild fish was done. Cultured fish have higher lipid content in muscle and liver, whereas values of PUFA were higher in wild wreckfish than in cultured fish, pointing out the higher values obtained for DHA and ARA in wild fish.

In addition, a comparison of feeding of broodstock with semi-moist diet and a "new dry food", as defined by Fatima, was conducted. This new dry food was formulated to obtain results on composition more similar to those obtained for wild fish. The "new dry food" is based on high protein content and with Vevodar supplementation to increase ARA in the diet. A general comment on the problem to base this new diet on Vevodar product was done, since this commercial product is not available anymore. The main result is an increase of n-6 PUFA, especially ARA, in the oocytes of cultured fish.

Problems and deviations from the plan:

No deliverables were planned in this WP until M54 and M57 (as seen in Table) but Fatima pointed out the necessity to formulate the different enrichments (to be done by FCPCT) during 2016 to start to work with them as soon as larvae will be available.

Deliverable	Title	Month due

D.12.1.	Effect of live prey enrichment products on wreckfish larval performance.	Month 54
D.12.2	Recommendations for wreckfish broodstock feeds.	Month 57

Plans for 2016:

Task 12.1. The first study will focus on EFA content in live preys. The influence of the different live food enrichments on the nutritional composition of the larvae will be studied. Biochemical analysis of samples of live food, as well as larvae will be done (CMRM). Emulsions will be formulated (FCPCT).

Task 12.2. A new experiment will be done in 2016 with

- Semi-moisture diet supplied to the IEO broodstock 1
- A second formulation of new dry food specifically formulated for wreckfish (FCPCT and SPAROS) that will be supplied to the IEO broodstock 2.

Nutrition for Atlantic halibut (WP11) Highlights:

Kristin Hamre (NIFES) presented the 2015 results for Atlantic halibut nutrition. In Task 11.1 (early weaning for Atlantic halibut larvae), Kristin presented the experimental design and some preliminary results, pointing out that those larvae fed "Otohime" feed (Japan) had high feed intake the first day. The feed was superior with respect to feed intake throughout the experiment and the tanks were cleaner.

In task 11.2 (nutrient profile of on-grown *Artemia*), Kristin summarized some results, pointing out that on-growing for three days gave increased concentrations of Protein, free amino acids, taurine, phospholipids, and decreased concentrations of glycogen and lipid. Different results for fatty acids at IMR and SWH were obtained, with different values of increases or decreases in DHA, EPA and ARA obtained. On-grown *Artemia* has been shown to improve halibut juvenile quality.

In task 11.3 (Nutrient retention and digestive physiology in response to on-grown *Artemia*), an experiment was conducted; Atlantic halibut larvae fed *Artemia* nauplii until 14 dpff (days post first-feeding). One group was fed nauplii, and the other on-grown *Artemia* until 28 dpff (2+ out of 3 meals). No difference in larval performance. 100% pigmentation and good eye migration were obtained in both groups. Samples have been taken for nutrient analyses (NIFES) and analyses of digestive capacity (ULL) after the end of feeding on-grown *Artemia*.

No actions in Task 11.4 and 11.5 have been presented (to be finished by months 36 and 48).

Problems and deviations from the plan:

No problems or deviations have been reported. Deliverables D11.2. and D11.3 are planned by month 36 (see Table), whereas Deliverable D11.1 was due by month 24, and was presented in time and no incidences (in webpage).

Deliverable	Title	Month due
D.11.1.	Report on nutrient profile of <i>Artemia</i> nauplii and ongrown <i>Artemia</i> from IMR and SWH.	Month 24. COMPLETED
D.11.2	Report on optimal characteristics of feed particles and feeding environment for early weaning of Atlantic halibut larvae.	Month 36
D.11.3	Report on the nutrient retention and digestive physiology in	Month 36

	Atlantic halibut larvae fed <i>Artemia</i> nauplii and on-grown <i>Artemia</i>	
D.11.4	Report on the nutrient retention and digestive physiology in Atlantic halibut larvae reared in RAS vs FTS	Month 36
D.11.5	Report on the effect of dietary phospholipids on Atlantic halibut juveniles	Month 48

Plans for 2016:

- Task 11.1. Weaning at early stages will be conducted within 2016.
- Task 11.2. Deliverable completed.
- Task 11.3. Analysis of digestive capacity (by ULL).
- Task 11.4. The experiment will be performed this spring/summer (March-July)
- Task 11.5. The experiment will be performed this Autumn (August-October)

Nutrition for Pikeperch (WP10) Highlights:

The 2015 results for pikeperch nutrition were presented by Ivar Lund (DTU). Regarding Task 10.1 (Effect of selected dietary nutrients on pikeperch larval development and performance), a trial was conducted on the effect of phospholipid levels and HUFA levels on ontogenetic development and performance of pikeperch larvae. Ivar presented the experimental design based on different levels of phospholipids (soy lecithin) plus the addition of EPA and DHA. Ivar pointed out the differences found in PE of larvae fed the different experimental diets, as well as on growth after 30 dph, better for those fed on the diet with high soy lecithin plus high DHA and EPA. The study on larval enzymatic response was also presented (done by FUNDP). Larval FA composition, skeletal deformities, staining & gene expression, tissue proteomics are in progress.

Regarding task 10.2 (Influence of salinity and dietary n-3, n-6 on pike perch larvae performance, metabolism and FA elongation/esterification capability), Ivar explained the design of the experiment conducted during 2015, with two different levels of fatty acids (w-3 vs w-6) in *Artemia* and three different salinities. Ivar presented some results including larval growth, SGR and mortality by confinement among the experimental groups and some preliminary results on C14 fatty acid and phospholipid metabolism performed by ULL after one week *in vivo* assays carried out at DTU. Larval FA composition, enzymatic response, eicosanoid production uptake and metabolism of lipid classes/esterification, deformities, staining and gene expression analysis are in progress by the different partners involved in this task.

Problems and deviations from the plan:

No problems were pointed out by Ivar. Deliverables of this WP are planned by Months 36 and 48 (Table).

Deliverable	Title	Month due
D.10.1.	Recommended Ca/P, vitamins and phospholipids to improve larval development and reduce skeleton alterations in pikeperch.	Month 36
D.10.2	Protocol for optimal early fatty acid enrichment to reduce stress sensitivity in pikeperch.	Month 36
D.10.3	Formulation for a diet better adapted to pikeperch requirements.	Month 48

Plans for 2016:

- Task 10.1. A Multifactorial approach (frSub-taskal and factorial experimental design, 16 diets) will be conducted with the following Variables; Ca/P, EPA+DHA, ARA, vit E, D, C, A, Se, two modalities per conditions (high and low levels). Expected trial period: February-March 2016.
- Task 10.2. Marine vs Vegetable Phospoholipids and DHA/ARA on welfare indicators. Planned by September-October 2016.

Ivar has pointed out that the deadline for deliverable 10.2 is very close to the finishing of expected trial.

Nutrition for Greater Amberjack (WP9) Highlights:

Firstly, José Pérez (ULL) presented the results obtained by IEO and ULL in their lab and facilities regarding greater amberjack nutrition within Task 9.1 (Improvement of larval enrichment products to enhance production of larvae and juveniles), subtask 9.1.2 (Combined effect of PUFA-rich lipids and carotenoids). In a first step, an assay was conducted to determine the optimum enrichment protocol and a trial was conducted with larvae after definition of the best protocols. José highlighted the results that Rotifers enriched (3h) with a polar rich emulsion containing a marine natural lecithin (LC60) and ARA, combined with 10 ppm of Naturose (E1,10), resulted in a significant advantage for amberjack larval growth, survival and welfare.

Within this task, Daniel Montero (FCPCT) presented the results obtained in subtask 9.1.1. (Determination of optimum essential fatty acid in enriched products for live preys) implemented by FCTCT. In a first trial, Optimum DHA in enrichment products for live preys for greater amberjack was determined. In a second trial Optimum EPA in enrichment products for live preys for greater amberjack was also determined, but these results were not presented due to the short time to explain all the results. Daniel highlighted that $1.5 \text{ g} 100 \text{ g}^{-1}$ DHA DW was determined to be sufficient to promote fast growth in greater amberjack larvae, whereas increased levels were associated to skull anomalies.

All these results belong to D.9.1, and Daniel presented a list of optimum levels and ratios of EFA and carotenoids in enrichment products, results coming from the deliverable.

DHA in enrichment products for *Artemia* 10-17% TFA EPA in enrichment products for *Artemia* 14-20% TFA DHA/EPA in enrichment products for *Artemia* 1-5 DHA in enrichment products for rotifers 14% TFA EPA in enrichment products for *Artemia* 6% TFA DHA/EPA in enrichment products for rotifers 2.3 Carotenoids levels in enrichment products 10 ppm

Regarding Task 9.2. (diets for grow out of greater amberjack), Yannis Kotzamanis (HMRC) did not present any results. Six diets with different levels of Lysine were formulated and experiment of on-growing is being conducted at this moment.

Regarding task 9.3. (Broodstock nutrition) Subtask 9.3.1 (Optimum ARA, EPA and DHA for reproductive success of greater amberjack), implemented by FCPCT and SARC, Daniel Montero presented some results of a trial conducted with some diets formulated by SARC and containing protein, taurine and histidine, and presented the highlight of better number of eggs per spawn in the diet supplemented with histidine.

Problems and deviations from the plan:

No problems were pointed out by José or Daniel. D 9.1 was delivered as planned in month 24.

Deliverable	Title	Month due
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D.9.1.	Optimum levels and ratios of essential fatty acids in relation to Tau and combined PUFA-carotenoids in greater amberjack enrichment products	Month 24 COMPLETED
D.9.2.	Lys requirements of greater amberjack juveniles	Month 36
D.9.3.	Performance of grow-out diets for greater amberjack developed in order to maximize growth potential	Month 58
D.9.4.	Recommended protein, carotenoids, Tau and EFA levels in greater amberjack broodstocks	Month 58

Plans for 2016.

For Task 9.2, HMRC presented a plan to conduct the chemical analyses (*body proximate composition, blood analysis, etc.*) and the evaluation of the amberjack growth performance, corresponding to subtask 9.2.1. No inputs have been done for subtask 9.2.2. by CANEXMAR, but the deliverable is not due until month 58.

Nutrition for Meagre (WP8) Highlights:

Daniel Montero (FCPCT) has presented the results from the studies of FCPCT. In the first Task (task 8. Improvement of larval weaning diets), two experiments were conducted. A first experiment to obtain the optimum essential fatty acids and related micronutrient levels in weaning diets for meagre was conducted, and the highlighted results were that 0.4% dietary HUFA is not enough to cover the essential fatty acid requirements of larval meagre and, since their elevation up to 3% markedly improved lipid absorption, essential fatty acids levels and growth, a high HUFA requirement in weaning diets is foreseen for this species. Besides, it is necessary to point out the importance of dietary vitamin E and vitamin C to protect these essential fatty acids from oxidation, increase their contents in larval tissues and promote growth, suggesting as well high vitamin E and vitamin C requirements in meagre larvae (higher than 1500 and 1800 mg kg⁻¹ for vitamin E and vitamin C, respectively).

In a second experiment, Daniel presented the results obtained for dietary vitamins A, K and D in weaning diets for meagre, and the results suggest a supplementation of meagre weaning diets with 2.4 mg kg⁻¹ vit K, since the absence of this vitamin markedly reduced larval survival. This species seems to be very sensitive to hypervitaminosis D and, only mildly to hypervitaminosis A, since supplementation with these vitamins leaded to a growth reduction. Taurine supplementation did not have any effect in meagre larvae performance under these experimental conditions.

For Task 8.2 (diets for grow out of meagre) some experimental diets have been formulated by SARC, based on different type of oils.

Problems and deviations from the plan:

No problems were pointed out by Daniel. D 8.1 was delivered as planned in month 24.

Deliverable	Title	Month due
D.8.1.	Improvement of larval weaning diets	Month 24 COMPLETED



D.8.2. Recommended essential fatty acid content in diets to promote meagre growth, welfare and health Month 48

Plans for 2016:

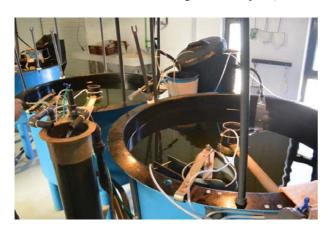
For Task 8.2, Daniel presented a plan to be implemented by FCPCT to conduct the on-growing experiment of meagre, fed diets based on different oils to determine the recommended essential fatty acid content in diets to promote growth, welfare and health. Experiment is planned to start by late summer 2016.

GENERAL DISCUSSION

It is interesting to comment that in the general discussion at the end of the workshop, some participants (i.e. Patrick Kestemont or Kristine Hamre) have recommended for the next meeting to present an overview on the results in a general way and not species by species, pointing out the different process (i.e. enrichers, microdiets, weaning diets) or maybe focusing on different bottlenecks to try to discuss and to have an overview of recommendations for "diversification" processes in aquaculture). The WP leader commented that this is an item to be conducted in following meetings, as probably this is difficult to be done nowadays with the results we have, but we will try for the next meeting.

Minutes of GWP Larval Husbandry workshop

Annual Coordination Meeting 2016, Day 2 (3/2/2016, 15:00-18:00)



By Dr. William (Bill) Koven, P4. IOLR (GWP Leader)

Present: Representatives of all 6 larvae species

- 1. Meagre-Alicia Estevez (P3. IRTA), Enric Gisbert (P3. IRTA)
- 2. Pikeperch-Alain Pasquet (P9.UL), Ivar Lund (P21.DTU)
- 3. Atlantic halibut- Torstein Harboe (IMR), Kristin Hamre (P17.NIFES)
- 4. Grey mullet-Bill Koven (P4. IOLR)
- 5. Wreckfish-Tito Peleteiro (P8.IEO)
- 6. Greater amberjack- N. Papandroulakis (P1.HCMR), José Pérez (P15. ULL), Virginia Martín (IEO. P8), Salvador Jerez (IEO. P8)

Summary of achievements/progress so far

The weaning of meagre larvae at 20 dph compared to the current protocol of 30 dph resulted in significantly poorer growth and survival suggesting that the presence of undefined nutritional factors in Artemia continue to give live food an advantage (IRTA). Studies on pikeperch found that in order to obtain homogenously sized pikeperch larvae with the best weight gain, light intensity should be 50 lux with a water renewal rate of 100%/h entering at the surface. In Atlantic halibut, flow through (FT) rearing systems gave better survival in yolk-sac and first feeding larvae than recirculating aquaculture systems (IMR). In addition, no differences were found between feeding Artemia nauplii or on-grown Artemia to metamorphosing larvae, in terms of eye migration, pigmentation and growth. These results are at odds with the striking nutritional differences between these different aged Artemia (NIFES). Research on 2-15 dph grey mullet demonstrated that an optimal level of algal turbidity significantly increased rotifer consumption independently of the algal species used (IOLR). Moreover, rotifer consumption in early development markedly influences juvenile survival much later on. During larval rearing of wreckfish, larval growth was documented from 2-24 dph as well as tentatively identifying blue sac disease (BSD) and swollen sac syndrome (SYSS) (IEO and HCMR). High larval mortality, resulting in complete loss of the population by 30 dph remains a stumbling block to the successful larval culture of this species. In greater amberjack, intensive rearing systems gave significantly better larval growth than in the semi-intensive mesocosm approach (HCMR). In addition, 24 h of continuous light produced the fastest growing 2-29 dph larvae, while green tanks gave better larval performance than black or white tanks. In addition the ontogeny of greater amberjack visual and digestive systems reared in mesocosm and intensive rearing systems was determined (HCMR/ULL). HCMR also successfully designed primers for determining somatotrophic axis protein and hormone gene expressions, which will be a major tool to investigate the endocrine and autocrine regulators for skeletal muscle growth.

- **1. Meagre:** Dr. Estevez (P3. IRTA) summarized Task 14.1 "Determining the earliest and most cost effective weaning period" that was carried out in two studies in 2014 and 2015. This is the only meagre task in WP 14 for Larval Husbandry. There was discussion why lipase activity was significantly greater in earlier weaned larvae and there was speculation that the higher lipid in the weaning diet may have caused this. However, IRTA is still in the process of preparing the deliverable (D14.1) and samples are being analyzed and will be completed in 2016. It is not clear at this stage if early weaning can be advanced from 20 to 10 dph. Cannibalism was a major problem in carrying out these studies and severely influenced results. A discussion on how to limit cannibalism included reducing light intensity and increasing the frequency feeding with *Artemia* and dry feed.
- 2. Pikeperch: Dr. Alain Pasquet (P9.UL) summarized their results using a multifactorial approach to look at environmental factors affecting pikeperch. They concluded that in order to obtain homogenously sized and best larval weight gain, light intensity should be 50 lux, water renewal rate must be 100%/h, siphoning tanks should be carried out in the morning and water inflow should occur at the water surface. This will be deliverable D16.1 "Determine effect of environmental factors on pike perch larval rearing". UL plans during February-March of 2016 to test nutritional factors that will include: food distribution (continuous or discontinuous), co-feeding, early or late weaning and weaning duration (3 or 9 days). The effect of population factors will be tested from April to June 2016 and will include fish density, single or mixed siblings, geographical origin and whether to do size grading. Cannibalism was also discussed in the pikeperch presentation. UL is involved in separating and identifying traits of individuals that are aggressive and demonstrate cannibalistic behavior. A discussion ensued about the merits of using bubbles (used with cobia larvae) or upwelling to limit the focus of larger individuals on smaller cohorts, thereby reducing aggression or whether bubbles will damage the larvae.
- 3. Atlantic halibut: Dr. Torstein Harboe (P7.IMR) summarized their work on Task 17.1 "Recirculation (RAS) vs flow through (FT) systems during yolk sac and first feeding stages and the effects on larval survival, quality and growth", Task 17.3 "Production of on-grown Artemia" and Task 17.4 "Comparison of feeding on-grown Artemia versus Artemia nauplii on larval performance". The results demonstrated that FT systems appear to give better performance than RAS during the yolk sac stage, that on-grown Artemia can be produced, but there was no difference in larval performance, in terms of eye migration, pigmentation and growth if fish are fed Artemia nauplii or on-grown Artemia. These results are at odds with Dr. Kristin Hamre's work, which found striking nutritional differences between Artemia nauplii and on-grown Artemia. A discussion followed that debated whether or not clearer results would have occurred if the experiment was run longer or other analyses were carried out, and that pointed out that if on-grown Artemia gave superior larval performance, the considerable costs in labor and growing these older Artemia would have to be taken into account. Deliverable D17.1 "Production protocol of on-grown Artemia" has been submitted. The deliverables D17.2 "Determine if RAS is a more effective protocol than FT for Atlantic halibut", D17.3 "The effect of probiotics on Atlantic halibut larval microbiota and survival" and D17.4 "Comparison of feeding on-grown Artemia versus Artemia nauplii on Atlantic halibut larval performance" will be completed by the end of 2016.
- **4. Grey mullet:** Dr. Bill Koven (P4.IOLR) summarized the work carried out in task 19.1 which (1) determined if microalgal concentration and its subsequent turbidity facilitate prey hunting and (2) compared the turbidity effect of *Nannochloropsis oculata* and *Isochrysis galbana*. The primary results showed that (1) algal turbidity significantly affected rotifer consumption independent of algal type and (2) rotifer consumption in early development markedly influences juvenile survival. This task was submitted as a deliverable but it was agreed with the PC to postpone submission until analyses of digestive tract enzymes and larval fatty acid composition will be completed in the next few months (IOLR, IRTA). In 2016 IRTA will compare the best performing diet from task 19.1 with a lyophilized substitute while IOLR will also compare the selected microalgae treatment against clay giving the same turbidity. There was some

discussion on the merits of clay, which has been used successfully in the culture of Atlantic halibut. Task 19.3 "Determining the effect of co-feeding ciliates and rotifers on digestive tract maturation and enzyme production" was also discussed. Since the writing of the DOW, the company that was to be subcontracted to produce the ciliates has closed, so the merits of co-feeding rotifers and copepods was discussed. The advantages and disadvantages of buying commercially produced copepod eggs were debated. The culture of locally occurring copepod species is preferable, but the continuous culture of copepods is very problematic although IOLR made notable progress during their Atlantic bluefin tuna experiments. The advantages of mastax counting was discussed, as well as possible reasons underlying the poor fertilization rate of the liters of grey mullet eggs that were spawned in the past 2015 season that limited the completion of a number of tasks.

- **5. Wreckfish:** Dr. Tito Peleteiro (P8.IEO) summarized the work carried out in the larval husbandry of this species. Although transport of eggs was successful, and studies on tank depth were attempted it has not been possible to grow the larvae more than about 30 dph. It seems that the fish tend to adhere to the side of the tank and although they are actively feeding they do not survive. Information on malformed individuals with blue sac disease (BSD) and swollen sac syndrome (SYSS) was documented, as well as total length from hatching to 24 dph. It was generally agreed on the imperative to improve larval survival in order to carry out planned tasks. Possible approaches to reduce mortality were discussed, which included light, photoperiod and tank color.
- 6. Greater amberjack: Dr. Nikos Papandroulakis (P1. HCMR) summarized the results of Task 15.2 comparing semi-intensive (mesocosm) with intensive systems in the culture of amberjack larvae. In 2014 there was significantly better growth in the mesocosm compared to intensive culture, while in 2015 this was reversed as intensive culture produced significantly larger larvae than the semi-intensive mesocosm. Possible explanations discussed were that the batch of larvae was of higher quality in 2015 and the improvements in the intensive rearing protocol. Sub-task 15.2.3 (ULL) that focused on the ontogeny (12 and 30 dph) of the amylase, alkaline protease, lipase and pepsin of the digestive system of greater amberjack larvae in the mesocosm and intensive systems was completed and discussed. In sub-task 15.3.2, 24 hours of light (24L: 0D) gave better growth compared to a 18L:06D photoperiod in 2-29 dph larvae while green tanks gave the best growth compared to black and white tanks. The possible problems of exposing larvae to continuous light were discussed. In sub-task 15.2.1 (HCMR) the primers for investigating larval somatotrophic axis proteins and hormones have been defined and were presented. This will be a major tool to investigate in 2016 the endocrine and autocrine regulators for skeletal muscle growth, which are known to play key roles in the regulation of metabolism and physiological processes. This Sub-task will contribute to D15.3 "Optimum hydrodynamics and light conditions during greater amberjack larval rearing". Sub-task 15.2.1 (HCMR) of this WP "Ontogeny of visual system through histological procedures" has been completed and will be submitted as part of D15.4 "Ontogeny of greater amberjack larval visual and digestive system reared in mesocosm and intensive rearing system". Deliverable D15.1, from sub-task 15.2.2, on larval stocking densities has been submitted by FCPCT recommending 50 eggs/l as the preferred stocking density. However, due to poor survival in the experiments and the ineffective use of primers for gilthead sea bream, skeletal deformities and gene expression of stress and skeleton related genes were not completed at this time. In 2016, Task 15.1 "The effect of feeding regime and probiotics" will be investigated (IEO) as well as Task 15.2.1 "larval oxidative stress through the activity of specific enzymes" (HCMR) and Task 15.3.1 (FCPCT) "The effect of tank hydrodynamics" will be studied.

Minutes of GWP Grow out husbandry workshop

Annual Coordination Meeting 2016, Day 2 (3/2/2016, 12:30-16:00)



By Dr. N. Papandroulakis, P1. HCMR (GWP Leader)

Participants

Nikos Papandroulakis (npap@hcmr.gr), Neil Duncan (neil.duncan@irta.cat), Estevez (alicia.estevez@irta.cat), Gisbert, <Enric.Gisbert@irta.cat>, Enric **Tassos** Raftopoulos <argofishsa@yahoo.gr>, Manolis Daniil, Daniel Montero, Salvador Jerez (salvador.jerez@ca.ieo.es), Virginia Martín (virginia.martin@ca.ieo.es), Jose Perez (janperez@ull.es), Robert Mandiki (robert.mandiki@unamur.be), Patrick Kestemont <patrick.kestemont@fundp.ac.be>, Ivar Lund <ili>agua.dtu.dk>, Batist XXXXX (FUNDP), Jan Batiste XXXXXX (Asialor) Bill Koven (bmkoven@gmail.com), Babis Syrigos (FORKYS).

Not present: GEI, DORAQUA, CTAQUA

Summary of achievements/progress so far

Meagre: Juveniles present high size variability and after selection no compensatory growth is observed. The SGR remains higher for groups with large individuals compared to groups with small ones. For on-growing, evidence exists that (a) cage depth is an important parameter for rearing of individuals between 200 g and 1 kg and that deeper nets result in better feed utilization and better survival; and (b) meagre exhibit feeding behavior during night. Meagre is able to learn and be trained to feeding stimuli (mechanical and optical).

Greater amberjack: Feeding frequency is important and 7 meals per day promote better feed utilization and growth. Stocking density at juvenile stages (5 g) affects significantly the growth performance. The species perform better at 26°C compared to 22 or 17°C presenting also morphological differences (elongated body shape at 26°C).

Pikeperch: From a multifactorial experiment 3 combinations of husbandry parameters (related to light, density, temperature, feed type and handling) were selected for further evaluation towards industrial application.

Grey mullet: A weaning diet has been developed with high level of FM substitution with plant protein sources that has been successfully tested, as it did not affect any of the performance and conditioning parameters tested.

WP20-Meagre

T20.1 IRTA presented the work done and the conclusion of the respective deliverable

T20.2 HCMR presented the work implemented and planned

- T20.2.1 The experiment is completed. Analyses are pending. No deviations from the DOW
- T20.2.2 Experiments are implemented. A second trial is designed to start during summer 2016. No deviations from the DOW.

T20.3 HCMR presented the work done

- T20.3.1 experiments implemented. Analysis is pending. No deviation from the DOW.
- T20.3.2 This task has not started yet. It is planned to start in Summer 2016.
- T20.3.3 This task will be implemented in 2016. It has been decided to re-organize the trial and to perform it only with one size class. An additional trial will be performed to test the night feeding of meagre in cages.

T20.3.4 to be implemented at IRTA.

The objective is to determine natural feeding rhythms and establish if these feeding rhythms improve growth performance (increase growth rate and decrease size dispersion).

Perform the trial in experimental scale: Comparison in each season of the year of (a) demand feeding and (b) feeding with automatic feeders programmed to follow the feeding routines. Use three replicate control tanks (automated feeding) compared to three experimental demand-feeding tanks. The parameters to be evaluated would be: feeding time, feed delivered, growth, size variation in the population, FCR, pattern of fish activity, level of aggressive behaviours and fin condition. Experimental conditions will be natural photoperiod and simulated natural temperature controlled to be similar to sea cage growing areas for the specific season.

- Obtain 10-g juveniles in June-July.
- Initiate experiment after 2-4 week acclimation and training with demand feeders.
- Continue experiment for 1 year with 6-8 week periods of monitoring in each season of the year.

No deviation following the 2nd amendment, which removed the work planned for P30. CULMAREX that exited the consortium.

WP21 Greater amberjack

In general the work planned is as in DOW. A general concern from the partners exists in terms of fry availability.

T 21.1

- T21.1.1 The trial will be performed as in DOW depending on fry availability. FORKYS and HCMR will decide on type of cages (diameter) to be used. Planned to start in 2016 Plan B: in case of low fry availability use cages of smaller volume.
- T21.1.2 CANEXMAR has some administrative issues for the installation of the submerged cage that are resolved in collaboration with ULPGC. The cage is expected to be on site in early 2016 The trial will be performed as in DOW depending on fry availability.

T 21.2

- 21.2.1 Implementation in 2016 as in DOW is expected.
- 21.2.2 IEO has implemented part of the trial and is expected to be concluded during 2016 as described in the DOW.

- 21.3.1 The task is to be implemented in part by HCMR and in part by ULPGC. The ULPGC part (individuals of 5 g) has been already implemented. The trials with 200 and 500 g individuals will be implemented according to DOW in 2016.
- 21.3.2 IEO has implemented the trial with individual size of 5 g. Rearing trial with individual size of 150 g will be implemented as in DOW during 2016.

During the special "greater-amberjack-meeting" on day 3 decisions were taken to secure proper implementation of all tasks.

WP22 Pikeperch

Dr. Patrick Kestemont (FUNDP) presented the work implemented and planned.

- T22.1 The trial has been implemented and some analyses are pending. The pending analyses need 2 months because of unexpected long steps for selecting immune gene primers for this species. So we will have more relevant results on key-immune gene expressions and brain serotonin by 8 April 2016. The deliverable, was due for mid February, but it will be delivered by the end of April 2016.
- T22.2 The task will start in the second part of 2016 as in the DOW, but confirmation is needed from ASIALOR for availability of juveniles and facilities. **Deliverable 22.2** is expected to be late by 6 months.
- T22.3 The starting date depends on the availability of different broodstocks. After the work performed in Reproduction & genetics WP, the available data will be used for identification of required stocks. Until June 2016 companies will be contacted and the stocks to be used will be defined. Larval rearing at UL will follow.

WP 23 Grey mullet

IOLR presented the work planned.

T23.1 IRTA concluded the work and the deliverable was submitted.

T 23.2, 3 and 4 The trials are under implementation in Greece and Spain and a new trial is designed in Israel to start during Spring 2016.

Deliverable 23.2, due for June 2016 will be late by approximately 12 months.

Minutes of GWP Fish Health workshop

Annual Coordination Meeting 2016, Day 2 (3/2/2016, 15:00-18:00)



By Dr. C. Secombes, P6. UNIABDN (GWP Leader)

Present

Chris Secombes (CJS), Douglas Milne (DoM), Daniel Montero (DM), Pantelis Katharios (PK), Karl Andree (KA), Cindy Campoverde (CC), Ramon Fontanillas (RF), Virginia Martin (VM), Salvador Jerez (SJ), Covadonga Rodriguez (CR).

Apologies: Sonal Patel.

CJS welcomed the group and gave an overview of the deliverables and milestones of the project, with a focus on the deliverables due in the next 12 months, so that everyone was reminded of the dates by which tasks should be completed.

WP25 greater amberjack

To date there were no deliverables due for this WP.

Task 25.1 There have been no incidences of Epitheliocystis so far. In gilthead sea bream the infectious agents appear to be two new species of bacteria (one being a new genus), rather than Chlamydia. A paper is in press on this work.

Task 25.2/ 25.3 DM reported work studying *Neobenedenia girellae* infection in greater amberjack, which appear more sensitive to the parasite than other *Seriola* spp. The fish appear to be immunosuppressed in that they get secondary infections. Mucus samples were collected from the infected fish for determination of the lysozyme and bactericidal activity. Lymphocytic infiltration is seen in skin histology. To date a freshwater bath is the only effective treatment. Since monogeneans are highly host specific, and only fish of >100-200 g are infected, discussion of what factors may attract them to the larger fish was considered.

Planned future diet trials with prebiotics/functional feeds that may impact on mucus defenses were described, with disease challenge possible in summer of 2016. The immune genes now characterized by CJS/DoM in Task 25.3 would be available to help analyze the responses.

Photoperiod was also considered as a way to block the lifecycle in tanks.

DoM had visited FCPCT in November 2015 to collect samples for analysis of induced responses *in vivo* and *in vitro* to PAMPs. The analysis is on-going.

Task 25.4 ULL/IEO reported on the success with using 0.25M mannose to detach *Zeuxtapta seriolae* from gill arches. Further work is needed but looks a very promising approach.

Task 25.5/25.6 On-going work is describing and diagnosing infections seen during grow out trials. These will be collated by all groups at the end of the project to prepare the practical manual (Task 25.7).

WP24 meagre

CJS pointed out that deliverables D24.1 and D24.3 had been submitted and that D24.2 was moved to Month 30 (from Month 24) and is on target to be submitted on time.

Task 24.1 Discussion of the difficulty in comparing scoring for Systemic Granulomatosis (SG) was had, with HCMR using larger fish and visual scoring, vs smaller fish used by FCPCT with histological analysis. The issue of when SG would be irreversible, in relation to the planned diet trials was discussed. HCMR used fish of 4g for the vitamin D trial (D24.1).

The planned trial with high plant protein was considered in terms of lead time for diets from RF/SARS (3 months), and facilities at FCPCT.

Task 24.2 The trial using seawater vs borehole water had been undertaken and the samples were being analysed to study the development of chronic ulcerative dermatopathy.

Task 24.3 Preliminary trials looking at anti-parasite treatments at IRTA had been encouraging with cinnamon essential oil having the best effect on antimicrobial function, with no effects on growth.

Task 24.4 *Nocardia* vaccination was proving problematic, in that to date no bacteria had been isolated from diseased fish in the DIVERSIFY program. DM mentioned he had isolated *Nocardia* from healthy fish, that was pathogenic for meagre, and would look into the possibility of using this strain to achieve the project's deliverables.

Action: DM to confirm to partners the conditions from his institution to do this.

Since this precludes preparation of the autogenous vaccine (DL24.6) or downstream evaluation of immune responses (DL24.13) and efficacy (DL24.12), it would seem sensible to defer these deliverables by 6 months, to allow more time to look for *Nocardia* strains and to investigate the possibility of FCPCT involvement.

Action: CJS to discuss with CCM.

Task 24.5 Immune gene sequencing and design of primers for qPCR have been finalized by IRTA/ABDN. The ontogeny study was completed and samples were ready to be analyzed.

Action: CJS/KA to confirm which genes/samples to be used by DoM/CC.

Task 24.6 The need for a *Vibrio anguillarum* vaccination trial was discussed and when to fit into the IRTA challenge facility timetable. Since the task relates to study of the immune response, it was agreed that pathogen exposure was not needed for mortality testing, but rather for examination of memory responses. Hence a few days only would be needed in the challenge facility.

Action: KA to look at best timing for this experiment, and CJS to coordinate input from DoM as needed. CJS/DoM to optimize ELISA assays for Ab analysis.



Task 24.7/24.8 On-going work is describing and diagnosing infections as seen during grow out trials. There was discussion of the group effort needed to put together the diagnostic manual.

WP26

CJS pointed out that deliverable 26.1 was submitted on time. The next one, D26.2, is due for the end of November 2016.

Minutes of GWP Socioeconomics workshop

Annual Coordination Meeting 2016, Day 2 (3/2/2016, 9:00-18:00)



By Dr. G. Tacken, P5. DLO (GWP Leader)

Attendees:

Michel van der Borgh (TU/e)

Ed Nijssen (TU/e)

Javier Ojeda (APROMAR: Spanish fish producer's association)

Matthias Keller (BVFi: German fish industry association)

Niki Alexi (HCMR)

Martiña Ferreira (ANFACO: Spanish seafood manufacturers)

Kriton Grigorakis (HCMR)

Ioannis Pelekanakis (FGM: Federation of Greek Mariculture)

Hellas Saltavarea (HRH)

Konstantinos Larentzakis (HRH)

Covadonga Rodríguez (ULL Tenerife)

Ricard Bou (IRTA)

Oxana Lazo (IRTA)

Luis Guerrero (IRTA)

Machiel Reinders (LEI Wageningen UR, minutes)

Marija Banovic (AU MAPP)

Thanasis Krystallis (AU MAPP)

Rocio Robles (CTAQUA)

Gemma Tacken (LEI Wageningen UR, chairman)

Summary progress so far

In 2015, 9 Deliverables have been finished and 2 scientific papers have been written and submitted to peer reviewed magazines. This is a very good accomplishment from everybody in the team.

In 2016, tasks 29.2 and 29.3 are planned. Since all tasks are sequential in this GWP, close cooperation between all work packages and tasks are necessary. In this meeting the following decisions have been made that are relevant for all other tasks that follow:

- 6 products will be tested in the sensory test
- The sensory test will be done in 5 countries
- The sample per country will be 100 consumers in the segments: *involved traditionals* and *involved innovators*
- In the experiments of 29.3 only the most interesting products of 29.2 will be tested. The maximum will be 6 products

The following readjusted planning of deliverables in this GWP will be proposed:

28.4 March 2016

29.3	March 2016
29.4	June 2016
29.5	June 2016
29.6	November 2016

Additional agenda point:

The following meeting is the next ACM planned in Barcelona in December 2016 or January 2017. The data and location will be determined at the end of this month or beginning of March.

> Since all deliverables are sequential, it might be necessary to have an interim meeting end of June, beginning July, when task 29.2 is finished and 29.3 is starting. When necessary, this meeting will be organised with the involved researchers. Please let the WP-leader know when such a meeting is wished.

In the agenda, a discussion about 28.3 is planned, however this deliverable is due at May 2018, so there is no direct necessity to discuss it now.

Matthias: Are the presentations of the plenary meeting available? Yes, all presentations become available. However, parts of the presentation of Marija and Thanasis will be published. As long as the publication is not ready, parts are not available. The presentation of Gemma and Kriton are available.

> Kriton and Marija will send their presentations to Gemma so that she can send it to all socioeconomic partners and Rocio and Dinos for uploading on the website.

Evaluation of year-2 of the project

What went very well:

- The focus groups were organized in a short time frame.
- Organization overall went well, taking into account the different countries that are involved in the project.

Attention points/ problems that we should take into account:

- In this GWP, we now have a delay of 2 months. Since all projects are sequential, a delay has a direct effect on the work of others. We are now in Month 27.
 - Gemma: Please be aware that we have to be ready in December 2018 and that all your delays have an effect on other deliverables.
- Logistics to get all raw products in place at the right time (Task 28.2.1).
- For some of the species it is a problem that there is not enough raw material to create new products to test with consumers (Task 29.2). Especially wreckfish is not available, so no new products of wreckfish have been developed.
- Arrangement of the expert interviews in some countries for Task 28.1.1 was difficult. But in the end we had an acceptable deliverable.
- Methodology of Deliverable 28.2 had to be revised: in the beginning, there was some misunderstanding of what to do with all ideas that came out of focus groups. But in the end the Deliverable had a good quality.
- Dissemination: there is a lack of information that is suited for communication in press or professional journals.
 - o Javier/ Matthias: Can you help us to develop some non-academic summaries from deliverables (executive summaries, main points) and send them to Javier. As long as information is not confidential, because papers are in preparation.

- For deliverables of WP27 and the already published deliverables of WP28 and WP29 these accessible summaries (not scientific) should be made, so that they can be used for dissemination to the EU companies in aquaculture. All summaries should be sent to Gemma before end March. Gemma will do WP 27, Marija will write summary for D28.1 and Machiel for D29.2.
- > All task leaders of 28, 29 and 30 include such an accessible summary (not scientific) for each new deliverable. Gemma will check whether these summaries are made.
- o Gemma: We made an easy readable article (in Dutch and English) for a Dutch aquaculture magazine. This article can also be used in the other countries.
- > Gemma will send the article to all participants in GWP7

Presentation Ricard Bou - Deliverable 28.4

Physical prototypes developed from ideas that came out of D28.2 and D28.3. These results will be presented in D28.4 and they are the input for the sensory test in 29.2 and mockups in 29.3.

IRTA: meagre and pikeperch

- > Frozen fish fillet with recipes (meagre)
 - Decide recipes, long shelf life
- Ready to eat meal: salad with fish (meagre)
 - Shelf life depends on the added vegetables
- Fish burgers shaped as fish (meagre)
 - Minimum difficulties, long shelf life
- Fish spreads/ pate (pikeperch)
 - Difficulties with the packaging (tubes)
- Fresh fillet with different 'healthy' seasonings and marinades (pikeperch)
 - Decide seasonings, relatively limited shelf life
- Ready-made fish tartar with additional soy sauce (pikeperch)
 - Shelf life is 6 days, however, with HPP can be extended and eventually combined with avocado

Javier: Due to the fact that one of the most important assets of European aquaculture is freshness, perhaps the prototypes should be based on it (and provide it). Nevertheless, more processed prototypes could be prepared from left-overs and trimmings from filleting. Highly processed product concepts can easily substitute EU aquaculture production with cheaper imports.

Ed: how can the prototypes be related to the main market segments (innovators, traditional, indifferent) or characterized so that retailers recognize the qualifications (e.g. high or low convenience)?

CTAQUA: greater amberjack and grey mullet

Greater amberjack:

- Frozen fish fillets that are seasoned or marinated (soya or honey)
- Tartar with additional soy sauce
- Fresh fish steak for grilling in the pan

Grey mullet (NB. Farmed fish not available, so wild fish has been used):

- Thin smoked fillet (salting and smoking with wet oak chips)
- Fresh fish fillet with different 'healthy' seasonings or marinades
- Ready-made fish fillet in olive oil



Rocio: What do we do with the requirement that products should be ASC certified (as stated in the product ideas)? For European aquaculture this requirement will be a problem (third countries are much more ahead with ASC labeling). So, maybe we should be less strict, so that any type of environmental certification will do.

Thanasis: We can incorporate certification as one of the parameters in the consumer experiments. The type of certification can vary, so we also check which one consumers recognize and place importance in their decision.

Matthias: Certification is often a license to enter a retail-chain, not because consumers are willing to pay a premium for it.

Javier: Align this discussion of certification with what was concluded in D27.2 about certification.

Javier: Food safety positive assets of EU aquaculture could be used to make the prototype preparations more attractive to consumers. For example, in Spain guaranteeing the absence of *Anisakis* could appeal to them. Thanasis: Better don't put certifications like parasite-free on the package, you create a negative association/ anchor.

Gemma concludes that these results are good work and she is looking forward to the Deliverable. It is a pity that this Deliverable was not far enough to present this in the plenary meeting.

Task 28.3

See above.

Planning of upcoming Deliverables 2016

Deliverable 29.3 (Due M28, 1st April 2016): development of the actual product samples from the selected species for the sensory testing with consumers in the five countries investigated. This is the design of the sensory consumer study > no delays are expected (ready by 15th March)

Deliverable 29.4 (M29, 1st May 2016): report on the actual products' sensory profiling in the five countries investigated. > two-months delay is expected (ready by 15th June)

Deliverable 29.5 (Due M30, 1st of June 2016), is dependent on the outcomes of D29.4, so this one will be delayed too. > one-month delay is expected (ready by 15th of June)

Deliverable 29.6 (Due M36, 1st of December 2016), will be ready in time again.

So at the end of 2016, this GWP is expected to be back on track according to the planning.

Task 29.2 - Consumer sensory tests (Luis)

Discussion points:

- > Sample. In the DOW, it says that we have to have at least 80 respondents per country. However, the consumer research identified 3 segments. So, Luis proposes to have at least 100 participants. Ideally 50 respondents per segment is the minimum.
 - Hellas indicates that there is no budget in recruiting, hiring locations and respondent fees to nearly double the sample.
 - Marija/ Thanasis: we can also choose the 2 most interesting segments (*Involved innovators* and *involved traditional*), 50 participants per segment.
 - Budget is anyway a problem, since sensory testing is relatively expensive. We can lower cost if we can make use of partner's facilities.
 - Conclusion: the sample will be 100 respondents per country in two consumer segments.

- Luis will contact local partners (university facilities including cooking and hosting) in the five focal countries (Germany, France, Spain, Italy and UK), which can be used as a location for the sensory testing. Hellas then will recruit the consumers for each location and pay the participant incentive and use of the facilities.
- Another point is whether other demographic aspects should be included in the sample requirements? Like age and income? What about fish consumption frequency?
- Luis will send the results with the outcomes of the consumer test to Marija.
- Organisation of the fieldwork. Do we think that the fieldwork has to be performed at the same moment in all countries? No, this is not necessary. However, given the preservation of products, it would be best when all the fieldwork will be done in one month. Luis will make a protocol on the basis of availability when the test will be done in each country. Spain will be first in order to be able to adjust things that won't work.
- Sample handling/ preparation/ shipment: The amount needed will be 50 kg/product (based on 100 g/individual/ participants). Gemma has spoken to Dinos who offered to help us. When Luis determines when he will need what amount of product, he can help organizing it.
- Luis makes a planning soon when he will need what amount of fish
- o **Cooking protocol:** Some products need cooking. A cooking protocol including the heating source will be included in the testing protocol. Preferably, there will be worked with cooks.
- o **Control of actions.** During the fieldwork in all countries, Oxana will attend the fieldwork activities and she will be responsible for controlling everything at the different locations.

New products to test:

- At this moment, 18 products, of three processing degrees, have been developed, by using raw materials from four species (meagre, grey mullet, greater amberjack and pikeperch). However, the number of products/ species to test (PREFMAP requires at least 6 products) is limited. Ten products is the maximum.
 - ➤ Decision: 6 products, also because the duration of sensory tests will be maximum 1 hour, given the budget for field research.
- There are 4 species for which we choose 6 products in total:
 - > Luis, Ricard and Rocio choose the 6 most interesting products. Luis should involve Matthias, Javier, Marina and Ioannis in choosing the products to be tested, because of their market experience.
- > It is not clear whether enough fish is available from all species. Gemma informed the GWP leaders and species leaders whether they could help. Bill Koven says that for grey mullet enough product is available in Israel.
 - > Rocio should timely contact him with the quantities needed at which place
- > In the GWP meeting, Gemma has received contacts for meagre and pikeperch and given the contact information to Luis. Meagre is quite available.
 - Lluis and Rocio should inform the species leader early on how much he needs and where, greater amberjack is probably available in Malta.
- > Dinos will send a contact in Malta to Rocio
 - > Criteria for selection should be:
 - Shelf life and transportation is an important criterion.
 - Raw material availability
 - Realistic handling and preparation procedure
 - Uniqueness in the market

Scalability of processing

> Protocol/design test:

- 1. 1st impression and overall expectations regarding the product (on base of pictures of the 6 products already cooked and prepared, ready to eat) without information.
- 2. Blind test: overall acceptability first, then acceptance of taste, odour and mouth feeling. Finally CATA (descriptors obtained from QDA, FCP, free terms elicitation, etc).
- 3. Overall expectation in full information condition (on base of a picture and description of 6 commercial products as consumers could come across it in the supermarket) after having tasted the different products.
- 4. Willingness to buy (regardless of price).

Presentation order:

Balanced order for both expectation measurements and same order for blind tasting in each tasting session.

Additional analyses to be performed:

- Sensory profile of the new products: Nikki (HCMR) will be responsible for that
- Preference Mapping: IRTA
- Other ideas/ suggestions

Task 29.3 - Discussion issues for experiments (Thanasis):

Planning of 29.3

The experiments will be based on mock-ups of products that present how the products will appear in the supermarket: packaged, labeled, etc. The choice of the mock-ups will be based on the outcomes of the sensory test.

Therefore, the Deliverable 29.5 will be delayed until the Deliverable 29.4 is ready.

Attributes. What are the attributes we want to include in the experiment (and how many attribute levels):

- Price (plus pricing > base price on retail prices as reported in Mintel database)
- Existence/ type of certification
- Origin
- Processing type/level

Thanasis and Marija will make a concept protocol that will be discussed in the task team.

Sample

Which segments? In the sensory test, we have chosen for the two most promising segments (involved innovators and involved traditional). It is best that we are consistent in all tests.

Number of test products

- Ideally, the 6 products that were selected for sensory testing should also be used in this experiment, but maybe these are too many products (to cover in the experimental design). If this is the case, then only the best-rated products of the sensory test will be included.

Rating of the products

For the rating of the products, a 7 points scale can be used, but a probability scale would be a good alternative. Ed suggested we use a ranking scale so he can also use this result for WP30.

Timing

D29.5 (M30: May 2016): the mock-ups, protocol and design should be ready by June 15, 2016 (1 month delay).

D29.6 (M36: November 2016): still on scheme > fieldwork is planned in September (after summer), so analyses can be ran and report can be written by December.

Task 30.1 Business models (Ed/ Michel)

Based on the input of customer demands (sensory testing and mock-up experiments), value propositions can be developed in cooperation with SME fish producers and processors of fish products. Ed and Michel ask Javier, Matthias, Martina and Ioannis, which companies could be relevant? To be able to make a selection, a definition of products and companies is necessary.

Javier: In order to receive feedback on this, it will be more interesting to approach processing and retailing companies than fish farmers.

- As soon as the selection of products is made for the sensory test and preferred products are selected, Ed and Michel make a protocol for the SME chain parties that should be approached for making business models.
- To get insights in SME companies in the aquaculture sector it is interesting to visit: Seafood Expo Global (26-28 April 2016 in Brussels), Fish International (14-16 February 2016 in Bremen)

Why do we focus on four species instead of six? Atlantic halibut had no commercial difficulties and wreckfish is not available as farmed fish in the short term. So, it is decided that no business models will be made for these products, since content of other tasks is needed for good business models.

Test market

In the test marketing task, products are needed that are presented in stores. Would it be as difficult to find product as now in the sensory test? In 2017/2018, the project and the producers are further, but contrary to the sensory test you will need SMEs that can produce the focal fish products and retailers that want to sell the product. It is legally not allowed to sell products made at a university or knowledge institute in stores.

> Just after the pre-test contacts with processors and producers should be made for test products.

This is priority in the next ACM

Gemma informs Ed and Michel that in case a real test is not possible a test with the virtual supermarket of Wageningen UR is a possibility. This is a back-up that can be used as plan B.

SMEs and retailers should be contacted right after the summer of this year already with the outcomes of the sensory tests and the experiments and discuss/convince them. Javier and Matthias can help to arrange the right companies.

Material exchange and operational planning

Operational things will be discussed bilaterally between the involved partners.

Other remarks:

Matthias:

In November, EuroTier will be held in Hannover Germany. This is a nice trade fair to disseminate the results from the socio-economic work package. The aquaculture sector from Europe is visiting this fair. Matthias is asked whether he is interested to organize a meeting there.

> Decided that Matthias will contact Thanasis and Luis for presentations. Gemma would like to be informed about date and time, so that she can attend too.



Matthias:

Fresh fish is gaining popularity, but at the expense of frozen fish. Even stronger: consumers cannibalizes the frozen for fresh fish but in smaller amounts. It could be that color plays a role here: fresh fish looks more attractive.

Matthias would prefer to look at fresh fish in the project, because that's most interesting.

Evaluation of the day

Rocio: Main challenge is to have the right amount of fish and that is not so easy to get from the producers at the moment

Thanasis: I'm optimistic. If we manage to overcome all the contingencies, then we did a good job. We are entering in a very interesting phase where all socio-WPs converge towards very concrete outcomes.

Marija: Nice to meet everyone.

Machiel: Nice to see a concrete product development project, but difficult to manage all the dependencies.

Lluis: I feel a lot of pressure now, because the rest of the project depends on the consumer tests.

Oxana: Optimistic as well: it is a challenge, but that's life.

Ricardo: Impressive all the work that has been done.

Konstantinos: Good collaboration is necessary.

Ioannis: I'm more an outsider. Nice to see the discussions.

Kriton: Creating the products is a huge step and people from IRTA and CTAQUA should get all the merit for that. But: getting the right amount of fish available will be the main challenge.

Martina: First contact with the project. Amazing what is done here.

Niki: Like to see the compromises that had to be made in a multi-disciplinary team.

Matthias: Nice to see all the developed products, but a little bit upset that wreckfish is not available. I feel that now it's my turn to get the producers, processors and retailers moving.

Javier: It should not come as a surprise that we do not have the kilos to provide for the market. This is the reason why the call has been produced in the first place by the European Commission. That is why the other researchers in the project are doing the technical work.

Gemma: Big compliment that we made the steps and that we could have the discussion that we had during this meeting. Thank you all.



DAY 3 - Dissemination, Scientific and Financial Reporting

During this day the agenda included a presentation by Dr. Rocio Robles on WP 31 Dissemination, presentations by the PC on Scientific Reporting and Financial Reporting, and a meeting of the Steering Committee (**Table 2**).

Dissemination

The presentation of WP 31 Dissemination begun with a brief reiteration of the WP's many objectives, emphasizing the need for all Partners to participate actively in the preparation of dissemination materials and activities (**Fig. 6**). Then there was a presentation of the various dissemination activities carried out in the last 2 years (2014-2015), which included the publication of four semester Newsletters that are uploaded at the



Figure 6. Photos from the presentation of WP31 leader Rocio Robles on Day 3.

website of the project and three species-focused articles published at the quarterly magazine of the European Aquaculture Society (for greater amberjack, meagre and pikeperch). A special session was held at the annual conference of the European Aquaculture Society (Deliverable 31.10), where presentations were given by all Species leaders, as well as many researchers from the consortium. The Species Leaders' presentations have been uploaded on the DIVERSIFY website.

As mentioned in the first ACM 2014, dissemination activities started as early as October 2013 (two months before the official starting date of the project) and so far the project has produced 120 Dissemination actions (from 48 as of the previous ACM 2014), which include (**Fig. 7**):

- 1. Magazine articles for the Aquaculture industry, and magazines addressing Politics, Policy and People (The Parliament Magazine, Paneuropean Network, CommNet, etc.),
- 2. Newspaper and magazine articles, press releases and media briefings,
- 3. Interviews in newspapers, radio or TV,
- 4. Web articles and movies of DIVERISFY research (www.ytube.com)
- 5. Oral presentations (18) and posters (17) in scientific conferences/meetings, including the dedicated Special Session "New/Emerging Finfish Species (EU DIVERSIFY Project) at the European Aquaculture Society's (EAS) "Aquaculture Europe 2015" conference,
- 6. Distribution of the project's flyer and bookmark to aquaculture professionals, regulators and administrators.

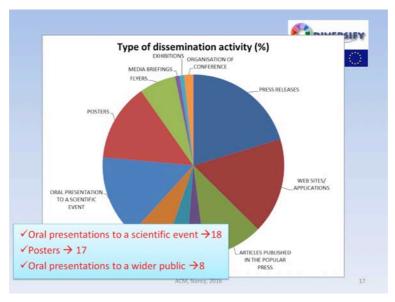


Figure 7. A pie chart showing the various dissemination activities of the project.

The vast majority of these dissemination activities have been already registered in the Participants Portal, though it has become apparent that not all Partners have been active in uploading their activities in the Participants Portal, especially presentations and posters in scientific conferences. AS pointed out, most of the dissemination activities were undertaken by P1. HCMR (the PC) and P18. CTAQUA. This is understandable to a large extend as these two partners have a major involvement and budget for WP 31 Dissemination, but the other Partners need to also dedicate some effort in this area. Dr. Robles encouraged the members of the consortium to pay more attention into uploading their activities on time, and a presentation of the Participants Portal was made to familiarize the partners again with the use of the site for uploading dissemination material. As before, dissemination material was mainly produced in English, but some material has been produced also in Greek, Spanish, German and Italian. It has also become known that some researchers have already started submitting their work for publication in scientific magazines. For

these publications there is a special page in the Participants Portal, and Dr. Robles introduced this page to the attendants of the meeting. She emphasized that it is time to start producing some scientific articles, in order to comply with our contractual obligations. Also, in order to make the work that will be coming out of DIVERSIFY more prominent and to increase the visits to our website, it was decided to modify the organization of the website. The modification will include moving the "scientific articles" page to the home page of the website. This will make this page more prominent and will make it easier for the visitors to see and access the work published in scientific journals.

As regards the DIVERSIFY website, the partners were informed that the website of the project (www.diversifyfish.eu) is averaging 250 visitors per day, a number that has not increased from the last ACM 2014, and needs to be increased. In order to facilitate the production of short reports on implemented work and acquired results to be uploaded in our site, the Dissemination leader prepared in 2014 a format file to be used by all scientists to prepare dissemination materials, in a way that would be easy for the partners to fill. The format file is available in the INTRA page of the DIVERSIFY website. Unfortunately, not many such reports have been produced so far, and more effort must be dedicated to encourage DIVERSIFY scientists to start preparing these short dissemination material from their activities. Invitations must be sent personally to the participating scientist from now on, as opposed to using the project's distribution list.

In terms of upcoming events, it was mentioned that oral presentations will be made in the EuroTier Trade Fair (http://www.eurotier.com/home-en.html), which is "the world's leading trade fair for animal production" to be held in November 2016 in Hanover, Germany. The invitation to present DIVERSIFY at the Aquaculture section of this Trade Fair (Fig. 8) was achieved through the actions of Dr Matthias Keller, from the German Association of Seafood Processors (P34. BVFi). It was agreed that DIVERSIFY will be represented with oral presentations by the WP 31 Dissemination leader and by Task leaders for Socioeconomics from IRTA (Spain) and University of Aarhus (Denmark).

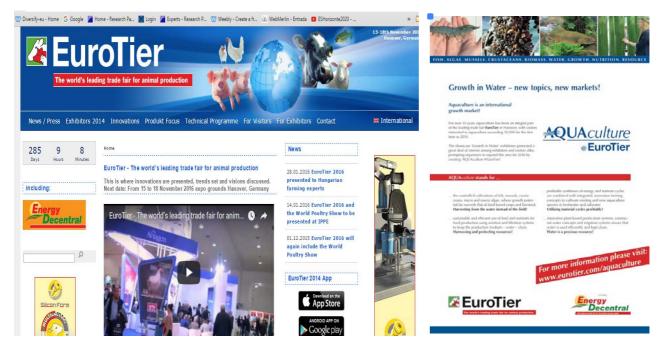


Figure 8. The home page of **EuroTier** (the world's leading trade fair for animal production) and the advertisement of the Aquaculture section.

An oral presentation is also going to be given by the PC at the upcoming Offshore Mariculture Conference 2016 (http://www.offshoremariculture.com/europe), which will be held in Barcelona, Spain (6-7 April 2016).

Dr. C.C. Mylonas was invited to give a presentation on DIVERSIFY, as well as prepared a small article about the project that will be distributed to the attendants in a Book of Abstracts. The chair of the conference is Dr. Alessandro Lovatelli, Aquaculture officer, FAO-UN and it has been supported by the leading aquaculture organizations – e.g. EAS, EATIP, GAA, FEAP, APROMAR, SEA. Regarding our participation to other conferences, no special session associated with the DIVERSIFY project is planned for the EAS 2016 conference, though any partner interested in presenting their work at the conference was encouraged to do so. A special DIVERSIFY associated conference will be prepared for the EAS 2017 conference to be held in Dubrovnik, Croatia (October 2017), for which the PC Dr. C.C. Mylonas is the president of the Scientific Program committee. As in 2014, the Species leaders will be asked to make oral presentations summarizing the work achieved.

The project was also considering the participation to the upcoming Seafood Expo organized in Brussels in April, and arrangements will be made with the commercial partners. The promotional workshops (Task 31.6) planned for the four strategic countries will start in Year 4 as planned in the DOW, although erroneously Deliverable 31.16 (1st Workshop) was listed to be delivered on Mo 37.

In agreement with the intentions of the consortium to be as open as possible and to disseminate the results as promptly as possible, all the presentations of the ACM 2016 will be uploaded on the website of the project, to be available to all interested stakeholders. In addition, it was agreed that all GWP leaders will submit a paragraph with the major highlights of the work implemented so far in their Scientific Disciplines, in order to prepare a 1-2 page flyer, which will then be translated to various languages by our Professional Association partners and disseminated to their members (e.g. in Greece, Spain, Hungary and Germany).

Management (Amendment, Scientific and Financial Reporting)

The Partners were informed of the submitted 2nd Amendment to the Annex I (**Fig. 9**). The reasons for the amendment were explained (loss of greater amberjack broodstock in P.24 ITTICAL and transfer of activities to P23. ARGO; exit of P30. CULMAREX and modification of work carried out in WP20, etc.).

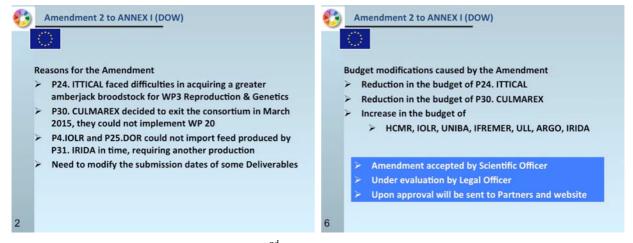


Figure 9. Explanation of the reasons for the 2nd amendment and the changes it will bring to the budget.

As in the previous ACM 2014, a presentation was given by the PC to explain how the Scientific reporting needs to be done for the project. This included both Deliverables and Periodic Reports. The presentation had begun with a reiteration of the roles of the PC, the Species Leaders (SL) the GWP leaders and the WP leaders (Lead Beneficiaries), in an effort to clarify their responsibilities and to remind the information that must be provided by the rest of partners to the WP leaders and GWP leaders.

Regarding the Deliverables, it was emphasized again to the Partners that they must be uploaded **on time** electronically in the Participants Portal. To ensure that all Deliverables are uniform throughout the consortium –in terms of appearance, format, and content quality and extent--, the PC has prepared a specific format file (**Fig. 10**), as well as explicit instructions on the preparation of the Deliverables, which are included in the website (http://www.diversifyfish.eu/2016-annual-coordination-meeting-feb.html). The Participants were also reminded of the Participants Portal and its functions that are relevant to the uploading of the Deliverables.



Figure 10. Instructions on the format file created by the PC for the preparation of the Deliverables of the project (available at www.diversifyfish.eu/INTRA/Forms & Protocols) and presentation of the Participants Portal section where Deliverables are listed and can be uploaded and downloaded by consortium members.

Then the PC discussed the status of the Deliverable submission, making a summary of the number of Deliverables that have been submitted so far and the ones that have requested a delay (**Fig. 11**). So far only 80% of the expected Deliverables have been submitted, but it is expected that as time goes on less delays will be faced and by the time of the Mid Term evaluation we will have almost all due Deliverables submitted. To ensure that the all Deliverables are not only uniform throughout the consortium in terms of appearance, but also that they are of high scientific quality, the PC has explained and emphasized the procedure that has been employed so far for the preparation and submission of the Deliverables (**Fig. 12**).



Figure 11. A summary of the Deliverables due and submitted so far and a presentation of the DIVERSIFY web page where all the submitted Deliverables are available to the consortium members, in the INTRA section of the website (http://www.diversifyfish.eu/deliverables.html).

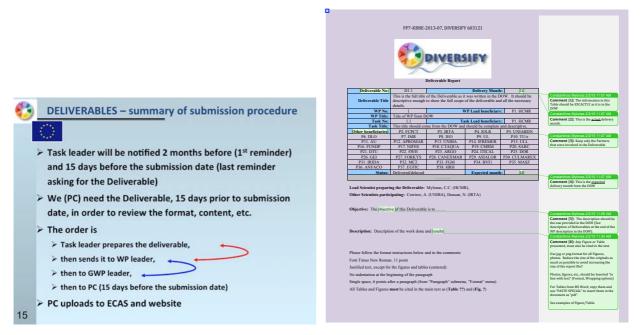


Figure 12. Explanation of the procedure that has been employed for the preparation and submission of the Deliverables, in order to ensure high quality and uniform presentation of all Deliverables, and the format document to be used by all Partners.

The session continued with a brief discussion dealing with the upcoming Periodic Report (Period 13-30 months, due July 2016). As for the Deliverables, special format files have been produced by the PC for each Work Package and will be sent to the Lead Beneficiaries of each Work Package (in April 2016) to help them compile the results and data from each Task (**Fig. 13**). It was stressed that the Periodic Report must include the work carried out during the reporting period with enough detail, but without excessive and unnecessary information, as it has been done for the 1st Periodic Report. This will allow the Consortium members to follow the major achievements as well as problems encountered during the 2nd period, and will enable both the EU Scientific Officer and the Mid-Term Evaluation committee to evaluate the work in relation to the DOW, and be able to make any necessary recommendations. A question must be made to the EU Scientific Officer regarding the inclusion in the report of the data obtained in the 1st Reporting Period (1-12 months).

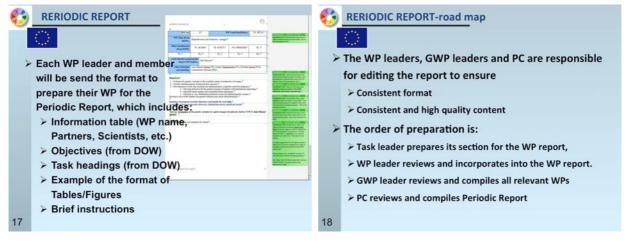


Figure 13. Representative slides from the instructions provided in the presentation for the procedure for the preparation of the Periodic Report for 13-30 Months, due in July 2016 (the whole presentation is available at http://www.diversifyfish.eu/2016-annual-coordination-meeting-feb.html).



As before, to speed up the process of preparing the report, while at the same time ensuring that a uniform and high quality document is presented (both in terms of format and content), the work has been delegated to all Consortium members as follows (**Fig. 13**):

- a. The Lead Beneficiaries for each Work Package (WP leaders) will request the text/figures/tables for each Task from the Task leaders, who are responsible to coordinate their writing with all scientists participating in their task. This process has already begun to some extent, in preparation for the ACM 2016, but must be updated with work that will take place in the next 3 months and completed by 20 May 2016,
- b. The Lead Beneficiaries then will compile all the information into a single document for each Work Package, review it for content/format/editorial errors and submit it to the GWP leader (30 May 2016),
- c. The GWP leaders will then compile all the Work Packages into a single document for each GWP, review it for content/format/editorial errors and submit it to the PC (10 June 2016),
- d. The PC will then compile all the GWPs into a single document to prepare the 2nd Periodic Report and review it for content/format/editorial errors (30 June 2016),
- e. The GWP leaders will also have to prepare following information (20 June 2016):
 - i. 3.1 Publishable Summary (0.5 page per GWP),
 - ii. 3.2.1 Project objectives for the period (0.5 page per GWP),
 - iii. 3.2.2 Project progress and achievements for the period (1 page per GWP)

and submit them to the PC for incorporation in the Periodic Report. The PC will prepare the remaining sections required (e.g., 3.2.3 Project management for the Period, Deliverables and Milestones, etc.) and will complete the 2nd Periodic Report by the end of June 2016 and upload it in the Participants Portal. All Partners agreed to the procedure and time schedule, and will do their utmost to complete the documents as requested and within the proposed deadlines.

Then the PC made a special mention to the work and effort of the GWP and Species Leaders, who are responsible for coordinating the work in their respective discipline or species, and thanked the six GWP leaders and six Species leaders. A change in one of the GWP leaders (Nutrition) had to be done in the few months prior to the meeting, due to the retirement of Dr. Hipolito Fernández Palacios from P2. FCPCT. The new GWP leader for Nutrition is Dr. Daniel Montero from the same Partner.

The PC then discussed the issue of preparing the work done in DIVERSIFY for submission to scientific magazines. Already 4 manuscripts have been submitted for publication and many more researchers expressed their intention to start submitting their work. The PC encouraged the Partners to publish their work as soon as possible (**Fig. 14**), not only to abide by the contractual requirements of the DIVERSIFY (2 articles per GWP per year, for a total of 60 articles), but in order to disseminate the work done and have as rapid an impact to the stake holders as possible. A change was agreed on the project's website, by moving



Figure 14. Representative slides from the discussion on Scientific Publications.

the "Scientific Publications" page to the main menu bar, so that visitors will have a more rapid and direct access to the scientific work of the Consortium.

Financial Reporting

Then, the PC discussed briefly some Financial Issues, regarding the payments received and the need for all Partners to abide by the budget allocation as described and agreed in the DOW, as much as possible (Fig. 15). Partners were reminded that each Partner has requested a specific budget, and allocated the EU support to clearly defined and described types of expenses, such as Personnel, Subcontracting, Travel, Consumables, Durable Equipment and Other types of expenses. The objective is to avoid unnecessary modifications that would require transfer of budget between types of expenses, or spending of the money in items not indicated in the DOW. Of course, it is recognized that there is always the possibility of unforeseen costs, but the Partners were encouraged to keep this to the absolute minimum. The PC mentioned that so far a number of minor modifications have been made, always after the agreement of the EU Scientific Officer, who has so far always accepted such requests, provided a reasonable explanation and justification has been provided.

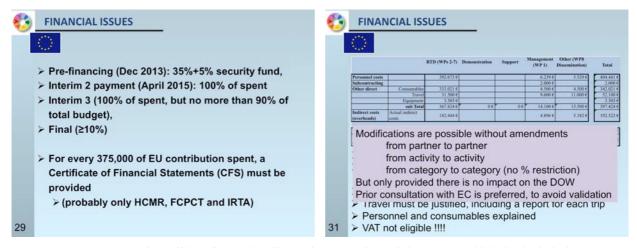


Figure 15. Representative slides from the discussion on Financial Issues, which included the payments received so far and an urge to the Partners to respect as much as possible their budgets and resource allocation (the whole presentation is available at http://www.diversifyfish.eu/2016-annual-coordination-meeting-feb.html).

It was agreed that Form C would be submitted to the PC by all Partners before 15 June 2016, so that the PC would have time to review and ask for corrections (if necessary) to complete the process by the end of the year. It was emphasized that in the previous reporting, all Form Cs were submitted to the PC except from one Partner, thus delaying the submission of the Financial Report by almost 1 month (still it was submitted before the official deadline!).

At the end of the presentation, the date and location of the **next ACM meeting** was discussed. The next ACM has been proposed for **Dec 2016 – Jan 2017** and **will be hosted by IRTA (P3. UL) either in San Carles de la Rapita or in Barcelona, Spain**. The local organizers will examine the best alternative both in terms of time and place and will inform the PC of their decision in early March 2016, at which time we will begin the process of preparing for the meeting. As this time no Periodic Report is due, the PC suggested that the format of the meeting is modified slightly. It was suggested that we hold 2 Open Days, where instead of summary presentations of the GWP leaders, we allow as many Partners as possible to present their work in 20-30 min presentations. This will allow the consortium members to have a more detailed view of the work carried out and will encourage the Partners to prepare their work for presentation and then publication.

Finally, the Partners were presented with a slight modification of the project's logo that we used recently for the new t-shirts. The new logo provides some information as to the objective of the project and the PC thinks it is more informative (**Fig. 16**). Still, the decision was not to modify all the DIVERSIFY documents with this "new" logo, but to use it only in our new t-shirt and some of our new activities and presentations.



Figure 15. The modified logo for the new t-shirts.

Steering Committee meeting

At the end of the third day, a meeting of the Steering Committee (SC) was held, as planned in the DOW. The SC members are the PC, the six GWP leaders, two representatives of SMEs (since P30. CULMAREX that was the third industry representative has exited the consortium) and the representative from a professional organization. The people attending this meeting were Mylonas, C.C. (PC, P1. HCMR), Duncan, N. (GWP leader, P3. IRTA), Montero, D. (GWP leader, P2. FCPCT), Koven, W. (GWP leader, P4. IOLR), Papandroulakis, N. (GWP Leader, P1. HCMR), Secombes, C. (GWP leader, P5. UNIABDN), Tacken, G. (GWP leader, P6. DLO), Daniil, M. (P23. ARGO), Deves, K. (P29. ASIALOR) and Ojeda, J. (P12. APROMAR).

No official agenda was prepared for the meeting, but the PC addressed the following issues:

- 1. 2nd Amendment The PC explained in more detail the major aspects of the amendment, especially the budget reallocation amounts to the various Partners.
- 2. Some discussion was made on possible changes of Partners and a further Amendment to the Annex 1. Specifically, it was mentioned that the PI from one Partner may be moving to another organization and would like to continue his involvement in DIVERSIFY. The PC will contact the EU Scientific Officer and examine the procedure for this. Secondly, one organization may be changing its structure, which would involve a change in name but also of legal documents (e.g. VAT registration number). As above, they would still like to be in the consortium, so we need to address the procedure for their validation and then joining of the consortium.



- 3. Also, two of the commercial partners may also be changing legal status or stopping their activities, so the consortium must already start examining the potential of other partners joining the consortium to undertake the work planned for these Partners.
- 4. Management Substitution of the leader for GWP Nutrition Dr. Hipolito Fernandez by Dr. Daniel Montero, due to the retirement of Dr. H. Fernandez has been requested by P2. FCPCT and has been reported and approved by the SC.
- 5. The PC mentioned that he is encouraging the participation/collaboration of more commercial operations outside the consortium, and introduced Isidro de la Cal as one company interested in having more collaborations with the consortium in the area of wreckfish reproduction and larval rearing. The company has a large stock of wreckfish breeders (more than 30!) and would be a very valuable collaborator.
- 6. Next ACM 2016b or 2017a will be held in the end of December or beginning of January in Spain, organized by P3. IRTA and hosted by Alicia Estevez The location of the meeting and the time will be determined by the host organization in the next month and will be announced to the consortium. The GWP leaders were encouraged once again to invite relevant scientists from the international community to attend this meeting.

Special Session on Greater Amberjack

Present

P1.HCMR: N. Papandroulakis, CC. Mylonas, P. Katharios, I. Papadakis, I. Fakriadis

P2.FCPCT: D. Montero, F. Acosta

P4.IOLR: H. Rosenfeld

P8.IEO: S. Jerez, V. Martin

P13.UNIBA: A. Corriero

P14.IFREMER: C. Fauvel

P15.ULL: C. Rodriguez, J. Perez

After a brief overview of the work carried out and some of the problems encountered, the following was decided:

1) Reproduction

- i. Spawning experiments will continue as planned in the DOW and implement corrective actions where required to improve egg quality and availability. Spawning for the acquisition of eggs will concentrate in the sea cage broodstocks, which showed the best performance so far.
- ii. Egg transportation from FCPCT to IEO and HCMR should be done during the next spawning period in order to allow comparison of methods/quality of eggs (particularly in case of natural spawnings). The problems regarding import of eggs have been resolved.
- iii. Genetic analyses should be done to see the differences, if any, between Mediterranean and Atlantic broodstock at IEO and FCPCT, given the significant differences in reproductive biology and performance in captivity between broodstocks from the two regions. Fin clips must be taken in the following samplings and sent to HCMR for analysis.

2) Larval rearing

- i. The main objective of the next year will be to produce juveniles to implement ongrowing trials, as most of the larval experiments have been concluded.
- ii. Trials already implemented, but with inadequate results related to husbandry (e.g low survival) may be repeated without performing the associated sampling or reducing it to two points (start end), in order to be able to match the results with previous trials.
- iii. However trials not completed or without covering the deliverable requirements will be repeated (e.g. samples have to be sent to ULL from FCPCT to complete the analyses from the density trial).
- iv. IEO-ULL will provide the info available for rotifer enrichment in order to be implemented in next year's larval rearing trials (as additive to already available commercial diets)
- v. FCPCT will provide the available info on *Artemia* enrichment as previously.

3) Growout

- i. CANEXMAR has to solve the issue regarding license very soon, otherwise measures regarding experiments have to be taken (these can include change of a partner or abandoning the task).
- ii. FORKYS should be ready to receive the juveniles, if available, otherwise the possibility to purchase juveniles from the market should be considered (the administrative and budgeting issues are to be discussed). The PC expressed the opinion that there should be no problem to transfer budget from consumables to the purchase of juveniles.
- iii. A "handling protocol" of the species has to be established, with compiled knowledge of different sources (HCMR, FCPCT, IEO) containing also up to date information regarding juveniles and growout.
- iv. A diet with increased content of protein is used in FCPCT, so the same could be used by other partners.
- 4) Establish a more close and "solid" collaboration with Dr. Robert Vassalo-Agius from Malta, as an expert in greater amberjack larval rearing.
- 5) Implementing the tasks as described in the DoW will require a better sharing of information among partners, as it is needed to implement the best practices and knowledge, even if they are still at experimental level.

Actions to be taken:

- 1) D. Montero (FCPCT) will send email at the end of February regarding points 3.i and 1.ii (and also the opinion of his colleagues on the decisions),
- 2) S. Jerez (IEO) will collect fin clips from their stocks (May 2016),
- 3) N. Papandroulakis (HCMR) will handle point 3.iii together with P. Katharios (HCMR),
- 4) N. Papandroulakis will handle point 4.

This concludes the minutes of the ACM 2016.



A group photo of some of the participants of DIVERSIFY ACM 2016 at the building "Presidence" of the University of Lorraine, Campus Brabois, Nancy, France.

Deviations from the DOW

The ACMs were planned in the DOW to consist of 2-days of open presentations and 1 for consortium activities. Instead, as for the previous ACM 2014 (Bari, Italy), the ACM 2016 contained only 1 open day and 2 days reserved for consortium activities. This was considered again necessary because of the large number of Work Packages in the project, and the need for as much time as possible to be allocated to the discussion of obtained results and future planning of the work.

For the next ACM (2016b or 2017a), which will be held between December 2016 and January 2017, we are considering having a 2-day Open Session, where all Partners will be allowed to present their work, instead of presenting only summary presentations. This will provide to all Partners a detailed view of the progress of the project after 3 years and will disseminate the information to a larger invited guest audience. Then we will have a full day of Scientific Discipline-specific Workshop (as of DAY 2 of the ACM 2014 and 2016) and a SC meeting in Day 3 and we will not have any other organizational/management session, since there will not be any Reporting Period associated with this meeting. The next Reporting Period ends in month 48 (November 2017), and just prior to this time we will have another ACM to coordinate the preparation of the Scientific and Financial Reports.

There were no other major deviations from the DOW at this time. Some delays in the uploading of the Deliverables have been discussed (and mentioned in the minutes of the GWP Workshops), but they are not considered major in kind. Also, there are a number of expected delays in some of the upcoming deliverables, but so far there is no expectation of any Deliverables not been completed within the lifespan of the project. These expected delays have been mentioned within the minutes of the specific GWP workshops reported in the previous pages.