



**European Cooperation  
in Science and Technology  
- COST -**

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**Secretariat**

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**COST 4152/12**

**MEMORANDUM OF UNDERSTANDING**

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Subject : Memorandum of Understanding for the implementation of a European Concerted Research Action designated as COST Action FA1205: Assessing and improving the quality of aquatic animal gametes to enhance aquatic resources - The need to harmonize and standardize evolving methodologies, and improve transfer from academia to industry (AQUAGAMETE)

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Delegations will find attached the Memorandum of Understanding for COST Action as approved by the COST Committee of Senior Officials (CSO) at its 185th meeting on 6 June 2012.

**MEMORANDUM OF UNDERSTANDING**  
**For the implementation of a European Concerted Research Action designated as**

**COST Action FA1205**  
**ASSESSING AND IMPROVING THE QUALITY OF AQUATIC ANIMAL GAMETES TO**  
**ENHANCE AQUATIC RESOURCES. THE NEED TO HARMONIZE AND**  
**STANDARDIZE EVOLVING METHODOLOGIES, AND IMPROVE TRANSFER FROM**  
**ACADEMIA TO INDUSTRY (AQUAGAMETE)**

The Parties to this Memorandum of Understanding, declaring their common intention to participate in the concerted Action referred to above and described in the technical Annex to the Memorandum, have reached the following understanding:

1. The Action will be carried out in accordance with the provisions of document COST 4154/11 “Rules and Procedures for Implementing COST Actions”, or in any new document amending or replacing it, the contents of which the Parties are fully aware of.
2. The main objective of the Action is to harmonize and standardize evolving analytical methodologies used in assessing the quality of aquatic gametes, improving their different uses, enhancing aquatic resources and transfer from academia to the industry.
3. The economic dimension of the activities carried out under the Action has been estimated, on the basis of information available during the planning of the Action, at EUR 60 million in 2012 prices.
4. The Memorandum of Understanding will remain in force for a period of 4 years, calculated from the date of the first meeting of the Management Committee, unless the duration of the Action is modified according to the provisions of Chapter V of the document referred to in Point 1 above.

## **A. ABSTRACT AND KEYWORDS**

During the past six years, three international workshops on fish gametes demonstrated a rapid development of methodologies that encompass extensive opportunities for promising use in basic reproductive biology, genetic research, biotechnology and aquaculture practice. All of these can have far-reaching consequences on conservation of endangered species, assessment of anthropogenic and climatic impacts on aquatic species and application in aquaculture, as well as in fisheries management. In particular, it has been recognized that there are many highly diverting details in the practical application of these new methods used by most scientists and laboratories, which can cause highly variable if not contradicting results, even using the same species.

There is an urgent need towards a universal scale to assess both the precise state of sexual maturation (for secure broodstock use) and related life history traits (gamete quality assessment, incubation of eggs) in teleost fish and other commercially important invertebrates used in either bioassays or aquaculture.

The aim of the Action is to reach a consensus on protocols and guidelines (using internationally defined terminology, units of measurement and format of reporting) that permit the use of results in relational databanks for sound and common application in aquaculture research and commerce.

**Keywords:** Aquatic species gametes, Aquaculture, Endangered species, Biotechnology, Cryopreservation

## **B. BACKGROUND**

### **B.1 General background**

Europe needs aquatic animal species. The markets' interest in fish consumption and the reduction of captures has partially been compensated by aquaculture, centered in controlled-reproduction species. These species can assist the assessment of anthropogenic and climatic impacts and fisheries management. Also, the need for conservation of genetic resources from endangered species has been addressed by gamete cryobanking or new techniques such as primordial germ cells (PGCs) transplantation. Moreover, the increasing use of aquatic biotechnology models such as zebrafish requires the use of transgenic lines that need adequate storage.

Scientists (and producers) find problems, such as extreme diversity of fish species and aquatic environments; necessity of conservation of genetic resources (biodiversity, aquaculture, biotechnology); diversity and need of standardization of the techniques used for assessing gamete quality and cryopreservation; existence of emerging techniques (PGCs handling, spermatogonia selection, proteomics, etc); high cost of experiments; lack of scientific networks; need of establishment of a forum on aquatic gametes research showing outcomes, defining goals and avoiding labs overlapping; need to harmonize methodologies linking academia and industry; and lack of commercial-scale know-how for scaling-up.

In particular, many innovative techniques for gamete production, evaluation, handling and storage are appearing. However, different labs are using them with very different modifications, reducing the comparability of results, making urgent the need of meetings for intercalibration and standardization, coordination and identification of gaps in research and technology, and exploring ways of transfer to industry.

These necessities of networking for international standardization, coordination and scientific exchange seem a perfect context for a COST Action, better than other research frameworks. On the other hand, most of the involved groups participate in EU or national projects funding their research, considering the AQUAGAMETE COST Action as complementary due to the nature of its objectives. Moreover, the research coordination within this COST Action will allow participants to successfully apply for future EU funding in the future.

## **B.2 Current state of knowledge**

The diversity of species, lab techniques and protocols results in a quite complex and diverse area of research, difficult to review. Briefly, research groups involved in the AQUAGAMETE Action work with a high number of aquatic species (mainly fish, but some invertebrates as well) and they use similar basic techniques (gametes quality evaluation, sperm activation, cryopreservation) but with small differences that make results difficult to compare. This problem obviously needs a standardization effort.

Probably, the most evident example is the use of techniques for sperm quality assessment such as CASA (computer assisted sperm analysis) and ASMA (assisted sperm morphology analysis) softwares, used to evaluate sperm motility and morphology parameters, respectively. Laboratories use software and hardware systems provided by different market suppliers or different versions of the same system. Some of the project partners are even actively involved in the development of these systems primarily on the software level. These systems might work on the same principle but each one is different in many details which render their results incomparable. Currently there is very little or no intercalibration of methods and protocols among labs and even less among market suppliers. On the other hand their use is becoming a standard requirement for sperm motility or morphology evaluation which forces scientists to use them. In addition, methods of preparing samples for analysis (pre-dilution of sperm, activating media) also differ according to research groups and laboratories which contributes to the low comparability of results.

On the other hand, new areas of research and techniques are being incorporated into studies on gametes, and in a network context pioneer groups can be the way to spread these new tools. To list a few of the new areas of research: proteomics, genomics, epigenetics, molecular biology, sperm motility mechanisms, water & ion control in gametes, oxidative stress and antioxidant protection or molecular mechanisms controlling oocyte competence. In addition, new techniques include short-term storage of gametes, manipulation of primordial germ cells (PGCs), spermatogonia selection, flow cytometry or FTI spectroscopy applications.

In a period when availability of funding is getting reduced in many EU countries, the need of research coordination to avoid overlapping studies and efforts becomes clear. One example is the cryobanking of aquatic genetic resources that is currently a need with different objectives (endangered species protection, aquaculture selected lines, biotechnology-generated fish strains). This area requires the analysis of requirements, standardization of selected freezing protocols used in different aquatic species and links to know-how acquired with mammals.

Most of the applicant groups have a common background consisting of the series of biennial International Workshops on Biology of Fish Gametes (IWBFGE). The First IWBFGE was held in Vodnany (Czech Republic) in 2007 and was entirely devoted to sperm ([www.vurh.jcu.cz/Website/Abstracts.pdf](http://www.vurh.jcu.cz/Website/Abstracts.pdf)). The Second one was held in Valencia (Spain) in 2009 and was opened to female gametes ([www.workshopbfg.upv.es](http://www.workshopbfg.upv.es)). The Third IWBFGE organized in Budapest (Hungary) in 2011 ([www.fish-gametes2011.org](http://www.fish-gametes2011.org)) was extended to shellfish gametes, and had 82 attendees from 17 countries, presenting 79 works. Next Workshops are planned in Faro (Portugal, 2013) and Ancona (Italy, 2015).

Proceedings of past IWBFGEs were published in the Journal of Applied Ichthyology. Approximately 30 papers are expected from the last meeting. Summaries and recommendations included in these Proceedings of Workshops (i.e.: Rosenthal et al., 2010: On the biology of fish gametes: summary and recommendations of the Second International Workshop, Valencia, Spain, 2009. J. Appl. Ichthyol. 26: 621-622.) already have expressed the main goals of AQUAGAMETE Action, mainly the need of research coordination and standardization on the application of lab techniques.

Organizers of the First Workshop are involved in this proposal. The main applicant and proposed AQUAGAMETE Action's Chair was the organizer of the Second one, while the organizer of the last meeting acts as Co-chair. Organizers of the upcoming two meetings are involved, too. This team, together with the rest of applicant groups, guarantees a good knowledge of the state of the art, as well as the role of the Workshop as main forum of this area of research.

These are global necessities, and the work with different local aquatic species, different techniques, etc. increases the diversity of work systems. In fact, several research groups from USA, Canada, Brazil, Japan, Singapore and South Africa joined the previous Workshops and have already expressed their intention of participating in this Action (most of them during the last Workshop in September 2011).

Apparently there are no other networks in Europe working specifically on the biology of the gametes of aquatic species, as the COST action LARVANET concentrates entirely on the culture of fish larvae for aquaculture without paying attention to gamete quality which is a similarly important issue in aquaculture management.

Possible links with AQUAEXCEL project, offering the chance of using aquaculture research facilities, will be explored.

### **B.3 Reasons for the Action**

Although there are many publications that present definitions and methodological details in this area of research, they are prepared for very specific and sometimes unilateral purposes and present all very different modifications, failing to address the many marginal conditions that affect the results while causing substantial experimental background noise. These add considerable uncertainty to the application of these methods. In this context, the existence of an expert network will serve as a reference in terms of techniques application and format of disseminating results. AQUAGAMETE will add value to published scientific results in this area of research because it will enhance the strength of the results by increasing their comparability.

Moreover, a series of Workshops and hands-on intercalibration exercises to assess the level of standardization needed, will greatly enhance the practical application of these methods and identify gaps in science and technology, thereby providing incentives to industries to improve performance of the equipment to be used in this field of application.

Research groups involved in the preparation of the Action work with most of the commercially produced fish species in Europe, as well as other species from the rest of the world. The AQUAGAMETE COST Action will have both immediate and future benefits as well as multiple applications. Firstly, the increase of scientific collaboration will directly potentiate the work of PhD students by giving them training in new techniques, that they will bring with them to their respective countries. This will have immediate application in running and future research projects.

These projects can have a wide range of topics, from aquaculture (with a direct impact on the European economy) to ecology or biomedicine (with evident societal benefits). On the other hand, the improvement of coordination of research can foster standardization and intercalibration of existing technologies and development of new ones.

A research network on the biology of aquatic gametes seems necessary in order to facilitate international cooperation, exchange of scientists and students, and efficient use of resources at all levels. AQUAGAMETE will pay a special attention to the training of young scientists (by organizing STSMs and specific training courses) and will continue the organization of biennial IWBF as a forum for researchers directly involved in the Action and many others.

The coordination of research will be increased with specific meetings and by the Working Groups (WG), avoiding the overlapping research funded by EU, and trying to maximize the dissemination of results. The intention is to arrive from the street level to the elaboration of recommendations for policy-makers (national, EU, bilateral cooperations, etc.). Lists of expected deliverables can be found at the WGs structure description.

AQUAGAMETE will find and describe common features to be considered to assess gamete quality and will pinpoint specific discrepancies considering the extreme diversity of fish species and the different characteristics of freshwater and seawater environments.

The main objective of the Action will be the assessment of relevancy and standardization of selected analytical methods, facing in this way the diversity of evaluation techniques and the need of standardization of the techniques used on assessing quality of aquatic gametes.

Another expected outcome is the development of cryobanking of aquatic genetic resources, solving the necessity of conservation of genetic resources and cells of interest (endangered species, aquaculture, biotechnology-generated fish strains). This will require the analysis of requirements and standardization of selected freezing protocols used in different aquatic species.

The Action will develop an active survey to support most promising axes, as new and relatively unknown techniques (PGCs handling, spermatogonia selection, proteomics, etc.).



AQUAGAMETE will solve the lack of previous scientific networks on the biology of aquatic gametes, establishing a forum on the scientific outcomes of this area of research. The creation of this targeted scientific network will improve the collaboration between specialized labs and will allow the share of specific and expensive tools. The Action will help in the definition of strategic goals of research on aquatic gametes, avoiding overlapping of research.

The Action will evaluate the need to harmonize and standardize methodologies linking academia and industry. The direct involvement of industry will help with the lack of commercial-scale know-how and the need of scaling-up for the application of techniques developed at the lab scale.

AQUAGAMETE will try to potentiate the communication with industries using internet resources.

#### **B.4 Complementarity with other research programmes**

All project partners have the adequate level of project funding for their gamete research (see section E.3.). These projects cover a very wide research area regarding both fundamental science and applied aquaculture and conservation research. The main areas of research of these partners can be classified as follows: research on gamete biology (activation, gametogenesis, fertilization), research on gamete quality (viability, DNA damage), gamete preservation (sperm cryopreservation and short-term preservation, cryopreservation of PGCs, spermatogonia, embryos), applied aquaculture research (improvement of gamete and embryo management at hatcheries, transfer of scientific results into aquaculture practice), applied species conservation research (cryobanking, use of cryopreserved gametes in conservation work) and various others such as toxicology studies using gametes. Thus, there is a level of complementarity with various nationally and EU funded projects, however, there is a clear lack of cohesion among these groups that sometimes work on very similar areas using different methods and sometimes finding different or even contradicting outcomes. Thus, the COST network would greatly enhance the cohesion of these groups and foster collaboration in related areas of research.

## **C. OBJECTIVES AND BENEFITS**

### **C.1 Aim**

The main objective of the Action is to harmonize and standardize evolving analytical methodologies used in assessing the quality of aquatic gametes, improving their different uses, enhancing aquatic resources and transfer from academia to the industry.

### **C.2 Objectives**

AQUAGAMETE COST Action will enhance scientific networking to improve the exchange of basic and fundamental knowledge, experimental capacities and academic resources as a way to reach secondary objectives:

- To review the current state of knowledge related to different aspects of research on aquatic species gametes (WGs 1-3).
- To coordinate activities related to the main research goals in this area by meetings of senior scientists dedicated especially to intercalibration and standardization of techniques of analysis, and to identify gaps in research and technology, to avoid overlapping research funded by EU and/or national sources, and to explore ways of transfer to industry (WG 4).
- To reach a consensus on protocols and guidelines (using internationally defined terminology, units of measurement and format of reporting) that permit the use of results in relational studies (WGs 1-4).
- To train students and young scientists (by STSMs and training courses), organizing courses for specific techniques and activities (WG 4).
- To continue the series of biennial International Workshops on Biology of Fish Gametes as a forum for the exchange of information between researchers directly involved in the Action, as well as other groups interested in this scientific area (WG 4).
- Maximize the dissemination of results at different levels and using different channels. Our intention is arriving from the street level to the elaboration of recommendations for national or international agency calls (EU, bilateral cooperations) (WGs 1-4).

### **C.3 How networking within the Action will yield the objectives?**

As has been mentioned, AQUAGAMETE COST Action will face several aspects related to the study of aquatic species gametes. This will be accomplished through scientific multidisciplinary networking, that will allow to share facilities, equipments, experience and knowledge. Methodical development will be carried out in close collaboration with industry partners in order to foster the application of developed technologies to aquaculture practice.

### **C.4 Potential impact of the Action**

This COST Action will promote cooperation between European research groups and others outside of the EU space in order to improve sharing of knowledge and attract and increase the attention for the reproduction problems of aquatic animals.

The Action will increase the level of coordination of research in this area, avoid overlapping, identify knowledge gaps and prepare joint proposals addressing them.

This Action will contribute to the standardization and development of new tools to control gamete quality. This would mean advances in production technologies, increased reproduction efficiency and ultimately reduced production costs which can be transferred to the industry.

Improvements in gamete evaluation, handling and storage will provide standard protocols for biotechnology and biomedicine research, genetic improvement in aquaculture and protection of endangered species.

These societal benefits will derive from advances in the understanding of the biological mechanisms determining gamete performance, identification of quality markers and will contribute to technical advances in production technologies.

## **C.5 Target groups/end users**

AQUAGAMETE will serve as a platform for several types of stakeholders: Aquaculture companies (especially hatcheries) working with seawater or freshwater species, research groups, Universities (being an added value for Universities having Aquaculture programmes, especially specific Aquaculture Masters, some of them involved in the Action), Biotechnology and Reproduction companies (including human applications), NGO-s involved in the conservation of aquatic species. This Action will use the COST tools to pioneer the integration of a broad range of multidisciplinary research in gametes of aquatic animals, connecting research groups and industry. For the moment, six companies directly interested will participate in the Action.

## **D. SCIENTIFIC PROGRAMME**

### **D.1 Scientific focus**

The AQUAGAMETE Action wants to solve main scientific problems in this area of research by 1) standardization of existing techniques, 2) creation of new techniques and tools, 3) sharing of knowledge on innovative techniques, 4) coordination of research. Different Working Groups (WGs) will face diverse aspects.

### **D.2 Scientific work plan methods and means**

The activities of the AQUAGAMETE COST Action will be organized under four Working groups:

WG1. Techniques for evaluation of gametes quality

The WG1 main goals will be to standardize current techniques for gamete quality evaluation and to develop new protocols for field, lab and industry applications.

## Objectives of WG1:

1. To organize and implement a survey of updated gamete quality assessment methods, both through cross information within the partners and through contacts with other groups of animal realm such as human and mammals.
2. To be incentive (or stimulating) for the test and use of updated methods in the research programs of the partners
3. To try and collect original data in order to standardize and propose adapted or dedicated analytical as well as field techniques of gamete assessment in aquaculture and reproduction biology academic studies.

## Expected deliverables from WG1

- Results and conclusions to be shown in the IWBFs organized by WG5.
- Suggestion of subjects for training courses organized by WG4.
- Description of WG1 activities and conclusions will be included in progress (annual + final) reports to COST.
- Specific reports published as user-friendly instructions manuals for the industry
- On-line delivered information (COST webs, AQUAGAMETE web and blog, Partners webs)
- Collaboration in the periodic edition of COST Action newsletters and press releases
- Collaboration in academic activities (on-line resources, links with Aquaculture Masters)

Milestones for WG1 includes at least 1 coordination meetings/year (month 9; coinciding with IWBFs or European Aquaculture Society Annual Meeting), at least one training course per year (month 3) and 1 reporting period/year (month 12). Other milestones include:

M1 Survey organization set up (month 6)

M2 Survey results acquired and proposition for new tools in running programs (month 12)

M3 Original results collected (month 24)

M4 Proposition of standardized methods (end of the project)

WG2. Gametes storage and preservation

The WG2 goals will be the standardization of protocols for sperm cryopreservation, and manipulation, short-term sperm storage and cryopreservation of spermatogonia; optimization of egg storage at sub-zero temperatures; coordination of tests of new cryoprotectants, and standardization of protocols for cryobanking of endangered species, aquaculture-selected or transgenic lines.

#### Objectives of WG2

1. To standardize cryopreservation protocols, proceedings during freezing/thawing, equipment and identify key bottlenecks in cryopreservation of different types of reproductive cells/tissues (oocytes, embryos, gametes, germ cells, reproductive tissues)
2. To provide techniques for cell and tissue preparation for cryopreservation
3. To simplify procedures and adapt protocols for specific situations: a) germplasm cryobanking of endangered and threatened species, b) selection of gametes from specific breeders for aquaculture production, c) broodstock reproductive management d) model species or specific transgenic lines (e.g zebrafish)
4. To articulate knowledge within workgroups (especially WG1 and WG3) to identify possible causes for cryodamage (oxidative stress, DNA fragmentation, etc.).
5. To integrate industry interests (fishfarms, cryobanks and cryogenic companies) with research knowledge and identify gaps in collaboration and needs.
6. Networking.

#### Expected deliverables from WG2

- Results and conclusions to be shown in the IWBFs organized by WG4.
- Suggestion of subjects for training courses organized by WG4.
- Description of WG2 activities and conclusions will be included in progress (annual + final) reports to COST.
- Specific reports (i.e.: standardization of cell collection, isolation and freezing techniques) published as friendly user's manuals for the industry
- On-line delivered information (COST webs, AQUAGAMETE web and blog, Partners webs)
- Collaboration in the periodic edition of COST Action newsletters and press releases
- Collaboration in academic activities (on-line resources, links with Aquaculture Masters)

Milestones for WG2 includes at least 1 coordination meetings/year (month 9; coinciding with IWBFs or EAS Annual Meeting), at least one training course per year (month 6) and 1 reporting period/year (month 12). Other milestones include:

- M1 Standardization of cryopreservation protocols for sperm and germ cells (month 10)
- M2 Identification of bottlenecks in cryopreservation of different types of reproductive cells/tissues (oocytes, embryos, gametes, germ cells, reproductive tissues) (month 18)
- M3 Establishment of techniques to collect, isolate and visualize cells for cryopreservation
- M4 Evaluation of cryodamage in different reproductive cell/tissue types as new tools in reproductive biotechnology (month 28).

WG3. Basic and applied research on gametes biochemistry and physiology, including omics.

The WG3 effort will be centered on providing a critical overview of the knowledge on the mechanisms responsible for the function of gametes (sperm motility mechanisms, gamete proteins, water & ion control in gametes, gametes oxidative stress and antioxidative protection, molecular mechanisms controlling oocyte competence) at physiological and pathological conditions. Special attention will be paid to integration of existing knowledge and development of emerging methods such as transcriptomics, proteomics and analysis of epigenetic modification of gametes. The acquired knowledge will be used for identification of critical factors of gamete quality.

#### Objectives of WG3

1. To provide critical overview of current knowledge related to following key areas: 1.1. Primordial germ cells (PGCs). 1.2 Epigenetic modification of gametes. 1.3. Genome quality in gametes. 1.4. Mechanisms of sperm motility. 1.5. Identification and characterization of gamete proteins and maternal mRNAs. 1.6. Water and ion control in gametes. 1.7. Oxidative stress to gametes and antioxidative protection. 1.8. Role of maternally-inherited factors in embryonic developmental success/potential.
2. To integrate research related to above key areas through:
  - 2.1. Networking such as STSMs, organizing specialized sessions in conferences, technical workshops and training schools (through WG4).
  - 2.2. Addressing identified gaps in the scientific knowledge.

3. To prepare joint proposals addressing identified gaps and to use basic knowledge to develop objectives related to applied research, such as gamete quality and biotechniques related to reproduction in aquatic animals.

#### Expected deliverables from WG3

- Results and conclusions to be shown in the IWBFs organized by WG4.
- Suggestion of subjects for training courses organized by WG4.
- Description of WG3 activities and conclusions will be included in progress (annual + final) reports to COST.
- Specific reports (Scientific reviews on fundamental knowledge and its potential applied significance related to areas 1.1-1.8)
- Important gaps in the scientific knowledge will be identified and a report on this issue will be prepared.
- On-line delivered information (COST webs, AQUAGAMETE web and blog, Partners webs)
- Collaboration in the periodic edition of COST Action newsletters and press releases
- Collaboration in academic activities (on-line resources, links with Aquaculture Masters)
- Direction of future studies will be defined and joint proposals will be prepared.

Milestones for WG3 includes 1 coordination meeting/year (month 9), at least one training course per year (month 10) and 1 reporting period/year (month 12).

#### *-Overview of current knowledge*

First, specific areas will be defined and leaders for particular topics will be named. Then, thematic sessions, covering key areas 1.1-1.8 will be planned for IWBF as described in WG4. Each session will include a review lecture and several reports concerning specific issues. At the second meeting of IWBF specific gaps will be named and addressed.

#### *-Proposed training courses*

Name of the Course: Micromanipulation techniques for Zebrafish Primordial Germ Cells (PGCs).

Program: Techniques for zebrafish genital ridge extraction, PGC, visualization and selection.

Techniques for sterilization of recipient animals and transplantation of PGCs from donor embryos.



Name of the Course: Identification and characterization of gamete proteins

Program: Extraction of proteins, electrophoresis and chromatography, identification and characterization of proteins, introduction to proteomics.

Name of the Course: Gene expression during gamete formation and early embryonic development in relation with developmental potential.

Program: Gene expression using quantitative PCR and in situ / in toto maternal mRNA localization during gametogenesis and embryonic development. Microarray analysis including design and data analysis. Use of next-generation sequencing (NGS).

During preparation and execution of training courses, opportunities for STSMs will be identified and offered through WG4 activities.

WG4. Organization of training courses, coordination meetings, and two next International Workshops on Biology of Fish Gametes

The WG4 will be dedicated to the organization of training courses, coordination meetings, IWBFs and will help with the organization of STSMs. The Executive Group will receive the groups' proposals for the use of funding and select year activities.

Objectives of WG4

- Student training courses. Working Groups 1-3 will define the necessities and topics of the most relevant training courses, and WG4 will coordinate with the hosting groups the organization of each of the courses.

An initial list of possible training courses including several subjects has been proposed by participant institutions: use of CASA/ASMA (sperm quality testing), sperm cryopreservation, egg and larvae microinjection, PGCs and spermatogonia handling, in vitro fertilization procedures, assessment of egg quality using embryonic survival and larval malformations, unfertilized egg holding at low temperature, analysis of egg transcriptome using microarray, gene expression in the oocyte using qPCR and in situ hybridization, protein methods (identification, proteomics), flow cytometry, FTI spectroscopy.

However, this is an open list. In fact, some of these courses could be repeated or offered in upgraded levels, and new topics can be incorporated depending on final participants (bringing new disciplinary perspectives), funding, network interest, etc.

- Coordination meetings. Focused on the standardization of techniques, creation of new tools, and coordination of research. Initially, proposed subjects are: CASA software development, gamete cryopreservation and quality evaluation (standardization), short-term storage of gametes (standardization), cryobanking (standardization, up-scaling know-how), and links with know-how in mammals and industry needs.
- STSMs. They will constitute an integral part of training within this action. STSM-s will be awarded to postgraduate or PhD students or other early stage research scientists based on proposals submitted to calls that will be announced preferably three times as year (depending on the budget). Their development will be under decision of the Executive Group.

International Workshops on Biology of Fish Gametes (IWBFGE). As it has already been mentioned, most of the applicant groups have a common background consisting in the series of IWBFGEs.

The WG4 will organize the 4th and 5th IWBFGE, as a forum for exchange of state-of-the-art scientific knowledge. But, more specifically its detailed objectives will include hosting AQUAGAMETE coordination meetings, exploratory sessions for EU projects application and plenary sessions for the presentation of conclusions made by AQUAGAMETE WGs, and including practical sessions presenting new techniques and standardized protocols using existing techniques.

The Action funding will allow WG4 leaders to increase the impact of coming IWBFGEs, trying to increase the number of attendees (they were 82 from 17 countries during the last meeting) and presented works (79 works at the last meeting).

Expected deliverables from WG4.

- Results and conclusions to be shown in the IWBFGEs organized by WG5.
- AQUAGAMETE has the preliminary intention of organizing every year 1 coordination meeting/WG1-4, 1 training courses/WG1-3 (see timetable), and a budget-dependent number of STSMs (making 3 calls annually).

- Description of WG4 activities and conclusions will be included in progress (annual + final) reports to COST.
- On-line delivered information (COST webs, AQUAGAMETE web and blog, Partners webs).
- Participation in periodic edition of COST Action newsletters and press releases.
- Academic activities (on-line resources, links with Aquaculture Masters).
- Grants for STSMs for exchange of students and researchers (seminars, tutorials, practical exercises).

Proceedings of coming IWBFs that will be published as special numbers in the Journal of Applied Ichthyology. In this case summaries and recommendations will be especially important because they will include the main goals of AQUAGAMETE Action, both in terms of research coordination and standardization on the application of lab techniques.

Milestones for WG4.

At least initially, the time table includes every year one training course suggested per each one of the WGs 1-3 (months 3, 6 and 10 respectively).

The 4th IWBF will take place in the Universidade do Algarve in Faro (Portugal) in September-October 2013, while the 5th IWBF will be host by the Università Politecnica delle Marche in Ancona (Italy) in September-October of 2015. During the last edition (Budapest, September 2011) researchers from both institutions committed themselves as coming Workshops organizers. Funding from this Action will help guarantying the successful organization of these meetings.

## **E. ORGANISATION**

### **E.1 Coordination and organisation**

AQUAGAMETE COST Action will be implemented through a concerted action, with the research carried out and financed by the participating countries, while COST provides the necessary co-ordination.

Main milestones include 1 coordination meeting/year (month 9) and 1 reporting period/year (month 12). Coming 4th and 5th IWBFs (in 2013 and 2015) will serve as a full network (including even external researchers) forum with the chance of having specific sessions (if required), coordination meetings, delivery of conclusions and reorientation of some research if necessary.

The Action will serve both at international and national levels, in especial in countries with several participating institutions/groups of research. Researchers representing each country in the Management Committee will have the task of serving as a connection point for all the country groups and coordinate their research. AQUAGAMETE Workshops, STSMs, training courses and websites will serve as linking for these groups, as well as a to increase the involvement of Non COST countries.

AQUAGAMETE organization will be based on a Management Committee (MC), an Executive Group (EG) and 4 Working Groups (WGs).

#### Management Committee

The MC will elect the Chair of the Action, the Vice-Chair, the WGs leaders (Executive Group). The MC will have one coordination meeting per year. They will be in charge of planning MC meetings, and will decide the subjects of training courses. It will be responsible of financing issues, WGs progress reports and preparation of annual reports. The MC will promote contacts with other ongoing COST Actions and relevant European and international initiatives.

#### Executive Group

The EG will be formed by the Chair of the Action, the Vice-chair and the WG leaders. They will have a regular contact using video-conferences if necessary. They will be responsible for ensuring the coordination between WGs (although they will be developed in parallel and relatively independent) and the proper dissemination of results. They will face the obligation to set up an Action specific website, while the WGs leaders will keep it updated.

The EG will decide the allocation of resources, in especial those dedicated to organization and assistance to STSMs and training courses. The Executive Group will receive the groups' proposals for the use of funding and select year activities.

## Working Groups leaders

They will detail plans of work, and coordinate research activities and dissemination of results through different ways. In especial, they will set up and keep updated the AQUAGAMETE Action website. This website will try to serve the needs of the participants giving detailed information of coming activities (as training courses, STSMs or links to webs of the next IWBF) and a blog open to partners-participation for fast-daily information.

Links to dropbox will be set up for the share of information and to give the possibility of working on line in common files (reports, papers, etc).

The website will help in the aim of ensuring the dissemination or exploitation of the results of the Action, showing results, documents (publications, conclusions, reports) and including links to other participating institutions to increase interactions).

## **E.2 Working Groups**

Four WGs has been established:

WG1. Techniques for evaluation of gametes quality

WG2. Gametes storage and preservation

WG3. Basic and applied research on gametes biochemistry and physiology, including omics

WG4. Organization of training courses, coordination meetings and two next International Workshops on Biology of Fish Gametes

The main objectives, deliverables and milestones of each of the WGs are shown in detail in section D.2.

Each WG will have a leader and a sub-leader, who could designate specific task leaders to increase work efficiency.

### **E.3 Liaison and interaction with other research programmes**

Interactions with the following projects and actions will include the exchange of information, common meetings, invitations to external researchers to join AQUAGAMETE meetings and Workshops, or participation in the organized training courses.

#### **Projects/Actions funded by international agencies**

PRO-EEL. Reproduction of European eel: towards a self-sustained aquaculture. EU Grant Agreement n°: 245257, 2010-2014.

MG4U. Marine Genomics for Users. Coordinator: B. Kloareg. EU Coordination and support action (COST). 2011-2013.

LARVANET. Critical success factors for fish larval production in European Aquaculture: a multidisciplinary network. COST Action FA0801.

AQUAEXCEL. Aquaculture infrastructures for excellence for European fish research. EU Seventh Framework Programme. 2011-2015.

CytoThreat. Fate and effect of cytostatic pharmaceuticals in the environment and the identification of biomarkers for an improved risk assessment on environmental exposure. EU Grant Agreement n°: 265264, 2011-2014.

REPROSEED. REsearch to improve PROduction of SEED of established and emerging bivalve species (Pacific oyster, king scallop, Blue mussel and European clam) in European hatcheries (KBBE-2009-1-2-11).

FRANCE AGRIMER/FEP. Fécondation Artificielle de l'Ombrine (Artificial fertilization of red drum). FAO. 2011-2013.

OPTISPERM. Optimized Sperm Management in Domesticated Animals: Transfer of knowledge through early stage researcher exchanges and trainings. FP7-PEOPLE-2012-ITN (submitted January 2012). *This project is submitted.*

Revitalization of endemic trouts from Neretva River for stock enhancement, conservation and aquaculture purposes. International project (Public Electrocompany of Herceg-Bosna, Mostar, Bosnia and Herzegovina). 2011-2014.

Development of experimental protocols for the breeding of new fish species of interest for aquaculture. Regional Project Co-funded by European Fund for Fisheries and Maribrin s.r.l. (Italy). 2012-2014.

LIFECYCLE. Building a biological knowledge-base on fish lifecycles for competitive, sustainable European aquaculture. EU FP7 2007-2013 (grant agreement no. 222719. 2009-2012.

BONAQUA. Cross border cooperation for the development of health standards in marine aquaculture. Programme POCTEP (Cross border Cooperation between Spain – Portugal). 2011-2013.

PROSPAWN. Implementation of natural spawning for marine fish species in culture – Improving the quality of offspring and animal welfare. EU 7FP. 2010-2012.

ARRAINA. Advanced Research Initiatives for Nutrition & Aquaculture; EU 7FP-KBBE. (2011.1.2-11). 2012-2016.

ECOAQUA. Establishment of cross border cooperation for a sustainable development of an ecological aquaculture system. Programme POCTEP (Cross border Cooperation between Spain – Portugal). 2009-2012.

Development and application of genetic, molecular and cellular biomarkers during fish development and sexual differentiation in the family Batrachoididae. Participants: ICMAN.CSIC (Spain), Universidad de Cádiz (Spain) and Universidad Oriente (Isla Margarita, Venezuela). 2010-2013.

2010-International Research Networking and Collaboration Scheme. Collaboration between the UK, China, Norway and Italy to improve fish health and ensure sustainable aquaculture. (MIUR) CUP I31J10000050001. 2010-2012.

Development of single sex populations of grey mullets (*Mugil cephalus*) to improve production and the enhancement of egg roe for Batarekh / Karasumi production. MERC-CDR grant (M27-066). 2011.

### **Projects funded by local/national COST agencies**

Producción de gonadotropinas recombinantes de lenguado senegalés: aplicaciones para la monitorización y el control de la maduración sexual en acuicultura. Fundación Ramón Areces. Spain. 2010-2013.

AQUAZOA. Aquaporinas testiculares durante la espermatogénesis de peces marinos y la activación y permeabilidad espermática. Spanish Ministry of Science and Innovation, AGL2010-15597/ACU, 2011-2013.

SPERMOT. Mecanismos fisiológicos implicados en la espermiación y en la motilidad espermática en la anguila europea (*Anguilla anguilla*). Spanish Ministry of Science and Innovation, AGL2010-16009, 2010-2013.

Caracterización, cultivo y criopreservación de PGCs y SSCs de peces. Spanish Ministry of Science and Innovation, AGL2009-06994, 2010-2012.

Aplicación de análisis genéticos en la creación de bancos de germoplasma en teleósteos. Fundación Ramón Areces. 2010-2012.

Sperm cryopreservation and detection of spermatological properties in rainbow trout (*Oncorhynchus mykiss*). 2012.

Identification and characterization of common carp (*Cyprinus carpio* L) seminal plasma proteins – proteomics and classical approach. Poland. 2011-2016.

Proteomic analysis of rainbow trout reproductive tract: identification of proteins specific for rainbow trout seminal plasma. Poland. 2011-2016.

Application of flow cytometry for the evaluation of sperm quality of Siberian sturgeon and rainbow trout. Poland. 2011-2016.

Characterization and the role of calcium binding proteins of fish semen. Poland. 2011-2014.

Using an air-stripping method for fish spawning in hatcheries – possible application and effect on gamete quality and broodstock welfare. Poland. 2010-2015.

Identification and characterization of common carp (*Cyprinus carpio* L) seminal plasma proteinases. Poland. 2010-2013.

Biochemical parameters of fish seminal plasma and their relationship with male gametes biological quality – improving the methods in fish sperm quality analysis. Poland. 2011-2014.

CZ.1.05/2.1.00/01.0024 South Bohemian Research Center of Aquaculture and Biodiversity of Hydrocenoses Czech Republic. 2010-2013.

CZ.1.07/2.3.00/20.0024 Strengthening of excellence scientific teams in USB FFPW Czech Republic. 2011-2014.

CZ.1.07/2.3.00/30.0006 The Creation of Postdoc Positions at the University of South Bohemia and the Support of Intersectional Mobility by Expert Stays at the Foreign Leading R&D Institutions. Czech Republic. 2012-2015.

ME10126. Environmental and hormonal induction of spawning, anesthesia, ontogenetic development and rearing of chosen fish species. Czech Republic. 2010-2012.



ME10015. The using of sex reverse and proteomic analysis in frozen sperm of paddelfish for production of caviar. Czech Republic. 2010-2012.

QH82119 Research of sperm and embryos cryopreservation. National Agency for Agricultural Research. Czech Republic. 2008-2012.

P502/10/P426. Fertilization process in sturgeon, function of acrosome and prevention polyspermy. Grant Agency of the Czech Republic. 2010-2012.

502/11/0090. Maturation and ageing of fish spermatozoa: A comparative study between teleostean and chondrosteian fish species as taxonomically distant models. Grant Agency of the Czech Republic. 2011-2015.

IAA 608030801 Diversity of bioenergetics pathways, membrane functions, signaling mechanisms and proteomics of cryopreserved sperm of evolutionary different fish species. Grant Agency of the Czech Republic Academy of Science. 2008-2012.

046/2010/Z Reproduction and genetics of selected model species of teleostean and chondrosteian fish. Grant Agency of the Czech Republic Academy of Science. 2010-2012.

Molekuláris biológiai vizsgálatra alapozott haltenyésztési rendszer prototípus kifejlesztése. GOP-1.1.1-09/1-2010-0141. Hungary. 12 000 000 HUF.

Mesterséges intelligencia interdiszciplináris alkalmazása az állatbiotechnológia-informatika korrelációban KMOP-1.1.1-09/1-2009-0049 Hungary. 55 000 000 HUF.

Network of Biological Resources Centers for domestic animals. Agence Nationale de la Recherche. The objective of this project is to integrate and upgrade Biological Resources Centers (BRCs) storing reproductive and genomic material for domestic animals in France, mammals, birds, fishes and shellfish. *This project is submitted.*

Reproductive, genetic and breeding characteristic of organisms interesting for mariculture. P.I.: Bosko Skaramuca. Ministry of Science, Higher Education and Sport. Croatia. 2007-2012.

Alteraciones en el material genético de espermatozoides: estudio de las consecuencias sobre la progeñie en peces (LE365A11-2). Junta de castilla y León (JCyL). 2011-2013.

Análisis de la cromatina espermática de peces y sus aportaciones al control del desarrollo y la calidad de la descendencia. Spanish Ministry of Science and Innovation (AGL2011-27787). 2012-2014.

OSCILE. Oocyte-somatic cells interactions, lessons from evolution. ANR – French National Research Agency. 2009-2013.

PHYLOFISH. RNASeq-based PHYLOgenomic analysis of gene duplications in teleost FISHes. ANR – French National Research Agency. 2011-2015.

EPISOLE. Epigenetic regulation of development and growth in Senegalese sole (*Solea senegalensis*). Fundação para a Ciencia e Tecnologia. Portugal. 2011-2013.

ULTRAFISH. Ultrasound to enhance the nutritional status of eggs and yolk-feeding larvae: a novel nutritional tool to program growth potential and metabolic pathways in juvenile fish. Fundação para a Ciencia e Tecnologia. Portugal. 2010-2012.

SEPIABREEDS. Improving the reproduction of the European cuttlefish, *Sepia officinalis*(Linnaeus, 1758) in captivity: a multidisciplinary approach. Fundação para a Ciencia e Tecnologia. Portugal. 2012-2014.

CRYOXI. Influence of antioxidant supplements in fish sperm quality: effects on seminal cryopreservation and progeny. Spanish Ministry of Science and Innovation (AGL2011-28810). 2012-1014

FA Polytechnic University of Marche. 2011- 2012.

RICERCA FINALIZZATA 2009 Ministero della Salute – Direzione Generale della Ricerca Scientifica e Tecnologica Code RF-2009-1536185. 2012-2014.

Ministero dello Sviluppo Economico (accordo ICE-MISE-CRUI) Code RF-220. 2012-2014

Domestication of the yellow tang as a model species for the surgeon fishes. Israel Ministry of Agriculture (894-0181-11). 2011.

Improving grey mullet stocking success in Lake Kineret & evaluating survival and growth performance of wild and cultured stocks via genetic markers. Israel Ministry of Agriculture (894-0186-11). 2012.

Artificial spawning of Lake Ohrid Trout for stocking purposes. National Project. Macedonia.

Artificial spawning of the common carp at Lake Prespa. National Project. Macedonia.

Control and spawning of salmonids in the fish farms in Macedonia. National Project. Macedonia.

Forming broodstocks of the endemic fishes in Macedonia for farming (aquaculture) and biodiversity conservation. National Project. Macedonia.

Cryopreservation of spermatozoids of the autohtonous and endemic fish species in Macedonia. National Pilot Project. Macedonia.

Oral application for passive immunisation for the manipulation of growth and reproduction. National fundings involving a mayor topic of fish physiology. Federal Ministry of Food, Agriculture and Consumer Protection. Germany. 2011-2014.

Future fish farming (F3) – Integrative agricultural production systems. Federal Ministry of Education and Research. Germany. 2012-2015.

Broodstock diet in sturgeon. German Foundation for the Environment. Germany. 2012-2014.

Reproduction of Atlantic bluefin tuna (*Thunnus thynnus*) in culture conditions and production of fingerlings for growout. ESPA 2007-2013 "SMEs", Ministry of Education, Lifelong Learning and Religious Affairs, General Secretariat for Research and Technology, Greece (9SMEs2009). 2011-2014.

Development of methods for reproduction and rearing of meagre (*Argyrosomus regius*) as a means for the enhancement of the competitiveness of aquaculture, with the introduction of new species. ESPA 2007-2013 "Cooperation", Ministry of Education, Lifelong Learning and Religious Affairs, General Secretariat for Research and Technology, Greece (ΣΥΝ09-24-424. 2011-2014.

### **Projects developed at NCI**

CRP7 project funded by the Singapore National Research Foundation for increasing seafood production through aquaculture genomics. Singapore. 2012-2016.

National project funded by the Singapore government on marker-assisted selection for improving foodfish stocks. Singapore. 2010-2014.

Genetic resources conservation of aquatic organisms with economic and environmental potential. EMBRAPA. Brazil. 2008-2012.

Preservation of tambaqui *Colossoma macropomum* semen. CNPq. Brazil. 2008-2014.

Development of great scale cryopreservation of tambaqui *Colossoma macropomum* semen: use in the production system and germoplasm bank. CNPq and FAPITEC. Brazil. 2012-2014.

Characterization and preservation of bijupirá *Rachycentron canadum* semen as a support to the formation of germoplasm banks and genetic improvement programs. CNPq and EMBRAPA. Brazil. 2010-2012.

Histopathological assessment of gonadal tissue in fish from eutrophic impoundments, South Africa. Fish Health and Reproduction on South African freshwater fish species.

Viability of male gametes in the sharptooth catfish *Clarias gariepinus* and Mozambique tilapia *Oreochromis mossambicus* exposed to endocrine disrupter chemicals.

Sperm motility parameters of *Oreochromis mossambicus* and *Clarias gariepinus* in a DDT sprayed area in South Africa. Water Research Commission WRC Report No 1674/1/09. South Africa.

Fish Health and Reproduction in selected dams in the North West Province, South Africa. Rand Water and Ecodynamics. South Africa.

Characterization and functional evaluation of antelope epididymal spermatozoa following cryopreservation in South Africa. (Wildlife Biological Resource Centre and the National Zoological Gardens). South Africa.

Reproductive physiology of the leopard, *Panthera pardus*. Agricultural Resource Centre and the Leopard Conservation Project. South Africa.

Study for Regulation of Sperm Function during Fertilization. Grant-in-Aid for Scientific Research (B) from Japan Society for the Promotion of Science (JSPS). FY2009-FY2011. Japan.

Study for Molecular Mechanisms of Sperm Chemotaxis toward Egg. Grant-in-Aid for Scientific Research on Innovative Areas from Ministry of Education, Culture, Sports, Science & Technology in Japan (MEXT). FY2010-FY2011. Japan.

#### **E.4 Gender balance and involvement of early-stage researchers**

This COST Action will respect an appropriate gender balance in all its activities and the Management Committee will place this as a standard item on all its MC agendas. The Action will also be committed to considerably involve early-stage researchers. This item will also be placed as a standard item on all MC agendas.

AQUAGAMETE COST Action involves for the moment a scientific community of 200 persons. The COST action guarantees a well balanced representation of both sexes. It directly involves women in all project activities. Young female researchers are also involved in the project. Early-stage researchers (<35) are very well represented, with a 46.5% of the total.

Women are 58.5% of the total, and have an especially high proportion between the early-stage researchers, being 66.4% of them.

The Management Committee will put a special emphasis on ensuring that a gender-neutral language be used at all levels of communication in the project. In its current structure, 58.5% of the participants in the Action are women, they are actively involved in the work of the action at the level of Management Committee as well as working groups leadership. The percentage of early-stage researchers in the Action is 54%, including the leader of WP4 and will be in charge of organizing STSMs. PhD students will be the main target for this kind of activities.

## F. TIMETABLE

Year 1	1	2	3	4	5	6	7	8	9	10	11	12
WG1	Grey								Grey			
WG2									Grey			
WG3			Grey + bars						Grey			
WG4						Grey + bars			Grey	Grey + bars		
Year 2	13	14	15	16	17	18	19	20	21	22	23	24
WG1									Grey			
WG2									Grey			
WG3			Grey + bars						Grey			
WG4						Grey + bars			Grey	Black	Grey + bars	
Year 3	25	26	27	28	29	30	31	32	33	34	35	36
WG1									Grey			
WG2									Grey			
WG3			Grey + bars						Grey			
WG4						Grey + bars			Grey	Grey + bars		
Year 4	37	38	39	40	41	42	43	44	45	46	47	48
WG1									Grey			
WG2									Grey			
WG3			Grey + bars						Grey			
WG4						Grey + bars			Grey	Black	Grey + bars	

Grey : Coordination meetings

Grey + bars : Training courses

Black : IWBFs

## **G. ECONOMIC DIMENSION**

The following COST countries have actively participated in the preparation of the Action or otherwise indicated their interest: CZ, DE, DK, EL, ES, FR, HR, HU, IL, IT, MK, PL, PT, SI, TR. On the basis of national estimates, the economic dimension of the activities to be carried out under the Action has been estimated at 60 Million € for the total duration of the Action. This estimate is valid under the assumption that all the countries mentioned above but no other countries will participate in the Action. Any departure from this will change the total cost accordingly.

## **H. DISSEMINATION PLAN**

### **H.1 Who?**

The target audiences for the dissemination of the AQUAGAMETE COST Action include several types of European and international stakeholders: Aquaculture companies (especially hatcheries) working with both marine or freshwater species, research groups working in the field, Universities (being an added value for Universities having Aquaculture programmes, especially specific Aquaculture Masters, some of them involved in the Action), Biotechnology and Reproduction companies (including biomedicine and human applications), NGO-s such as angling clubs conducting induced spawning of fish for stock enhancement, European, national and regional levels policy makers and, at least indirectly, general public.

### **H.2 What?**

AQUAGAMETE Action e-mail network

It exists even before the preparation of this Action and will be a fast-communication and coordination system to link all the Action participants.

AQUAGAMETE Action website

The website will have a key role in ensuring the dissemination or exploitation of the results of the Action. It will have several sections with different kinds of information.

In the open-access part of the web site, web visitors will find posts of general information for general public, a list of the published articles into the Action in peer-reviewed scientific and technical Journals (with links to abstracts, contents and main journals), results, documents (Action publications, reports and conclusions), as well as links to participating institutions to increase interaction.

On the same website but in a password-protected area, the Action participants will find posts of working documents, detailed information of coming activities (as training courses, or links to webs of the next IWBF) and a blog open to partners for fast and daily exchange of information.

Other on-line resources

Dropbox (or similar software) will be used to the share information between Action participants and to give the possibility of working online with common files (reports, papers, etc).

Other on-line delivered information (i.e.: COST webs).

Reports and recommendations

Progress reports to COST.

Specific reports published as user-friendly instruction manuals for the industry.

Preparation of recommendations for agency calls (national, EU, bilateral cooperations) following the Action conclusions and trying to avoid overlapping research funded by EU.

In this case summaries and recommendations will be especially important because they will constitute the main targets of AQUAGAMETE Action, both in terms of research coordination and standardization on the application of lab techniques.

Press

Periodic Action newsletters (for scientific audience).

Press releases (for general audience), using the involved institutions systems (i.e.: journalists, university radio/TVs, regional TVs, etc).

## Events

WG4 will be specifically dedicated to the organization of training courses (initially, 3 per year) and coordination meetings, and will help with the organization of STSMs (seminars, tutorials, practical exercises), coordinating with the hosting groups the organization of each of the courses. An initial (open) list of possible training courses has been mentioned in section D.2. An additional objective of WG4 will be organizing the 4th (Faro, Portugal 2013) and 5<sup>th</sup> (Ancona, Italy 2015) IWBFs as a forum for researchers directly involved in the proposal and many others (see section D.2.).

Contributions to other national and international (aquaculture, endocrinology, reproduction) conferences and symposia.

## Academic activities

All kind of open on-line resources can be shown in Aquaculture Masters lectures.

Some of the Action subjects can be used as case study.

Manuals derived from the Action can become study materials for students.

In the case of host institutions, training courses can even be linked with Aquaculture Masters Programs (student attendance, parallel series of talks by experts).

## Publications

Articles and reviews in peer-reviewed scientific and technical Journals.

State-of-the-art reports (i.e. to be shown in international congresses or book writing).

Books of abstracts of the coming Biennial workshops (4<sup>th</sup> and 5<sup>th</sup> IWBFs).

Proceedings of coming 4<sup>th</sup> and 5<sup>th</sup> IWBFs that will be published as special numbers in the Journal of Applied Ichthyology.



### **H.3 How?**

The dissemination methods described in section H.2. will be implemented at different levels:

The Management Committee will decide the subjects of training courses and will compile annual COST reports.

The Executive Group will be the main responsible for ensuring appropriate dissemination of results, checking in each of its meetings the progress of the Action to update the dissemination plan during the course of the Action. They will face the obligation to set up an Action specific website, while the WGs leaders will keep it updated. The Executive Group will receive the Action groups' proposals for the use of funding and select annual activities.

The Working Groups leaders will write WGs progress reports as parts of the annual COST reports. They will coordinate dissemination of results through different ways, especially updating the AQUAGAMETE Action website (information of coming activities, links to IWBFs webs, etc). An open Action blog will be open to all partners-participation for fast and daily exchange of information.

As it has been mentioned before (see section D.2.) WG4 will be centered in the organization of training courses and coordination meetings, as well as the two next IWBFs, respectively.

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