WORK PROGRAMME FOR THE EU
REFERENCE LABORATORY FOR MOLLUSC DISEASES

2016-2017

I. LEGAL FUNCTIONS AND DUTIES

The functions and duties of the EU Reference Laboratory for Mollusc Diseases are given in Annex IV, Part 1 of the Council Directive 2006/88/EC.

II. OBJECTIVES FOR THE PERIOD JANUARY 2016 – DECEMBER 2017

The following table presents equivalences between activities we have identified and operational objectives proposed in the Commission Work Programme for 2016 and 2017 on financing contribution to the EURLs

OPERATIONAL OBJECTIVES		ACTIVITIES
1	To ensure the development and use of high quality analytical methods across the EU-RL framework	I-4), I-5), I-6) II-1), II-2), II-3), II-4) III-1), III-2), III-3), III-4), III-6) VI-1), VI-2), VI-3)
2	To maintain appropriate level of proficiency testing ensuring efficiency of control analysis methods	I-1), I-2), I-3), I-4), I-5) III-5)
3	To ensure the availability of scientific and technical assistance provided by the EU-RLs	I-3), I-4), I-5), I-6) II-5) III-1), III-2), III-3), III-4), III-5), III-6) IV-1), IV-2), IV-3), IV-4) V-1) VI-3)
4	To ensure a sound and efficient management of EU-RL funding cycle	VII-1)

Main activities	Sub - activities	Description	Objectives	Expected output	Performance indicators	
I. Coordinate the methods employed for diagnosing diseases	I-1) Organise an Inter-Laboratory Comparison (ILC) test for the detection of some mollusc pathogens by histology and cytology (2016-2017)	In 2016 - Preparation and checking of the required material - Establishing the reference results - Planning the sending and reading of slides - Sending of material In 2017: - Analysing results - Preparation of the report of the ILC - Presentation of the results of the ILC during the 2017 Annual Meeting	To test the competency of participants regarding the detection of some mollusc pathogens by histology and cytology	Each participant is expected to have more than 60% of good results	AH.PT.1 AH.PT.2 AH.PT.4	1 30 60%
	I-2) Organise an ILC for the detection of the *Bonamia ostreae*/*Marteilia refringens* by Real Time PCR (2017)	In 2017: -Preparation and checking of the required material -Establishing reference results -Sending samples -Analysing results -Preparation of the report of the ILC	To test the competency of participants regarding the detection of *Bonamia ostreae*/*Marteilia refringens* by Real Time PCR	About 250 samples are to be prepared Each participant is expected to have more than 60% of good results	AH.PT.1 AH.PT.2 AH.PT.4	1 250 60%
	I-3) Additional training for the detection of mollusc pathogens by histology Organise satisfaction surveys	Based on results obtained during the 2014-ILC-01, specific training (in the EURL and/or by distance using mScope® software) will be proposed to NRLs in 2016 and 2017 Satisfaction surveys are sent after the training sessions to participants	To improve the capacity of some NRLs to detect listed mollusc diseases by histology Evaluate the interest of NRLs in the training sessions and improve the organisation of these sessions	All the NRLs should obtain more than 60% of good results in the next ILC test based on histology the EURL	AH.PT.5 =AH.NRL.5 AH.PT.6 AH.NRL.6	At least one specific training organised at distance or in the EURL Minimum expected success rate at the next ILC test based on histology : 60% 75% of positive feedback (>85%)

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	I-4) Supply available reference reagents and material to the NRLs on request or when a new diagnostic method is available	Reference material provided to laboratories working on mollusc diseases usually consists of H&E stained histological sections and paraffin blocks when available, bacterial strains as well as DNA suspensions. Considering the increased use of PCR and Real Time PCR in NRLs, the EURL has decided to prepare and monitor under quality management a stock of plasmidic DNA suspensions to be used as positive controls in these PCR assays	To supply reference material to the NRLs	All the NRLs should have: - reference histological slides for the listed mollusc pathogens - positive controls available for the detection of listed pathogens by PCR	AH.PT.2	New samples collected through experiments, field studies or confirmatory diagnosis are regularly included in our collections. At least 300 samples are included every year.
	I-5) Maintain and update the library and collections of mollusc pathogens (including scanned slides)	The collections of histological material, bacterial strains, ethanol fixed tissue, DNA suspensions are continuously checked and enriched with new reference material from field studies or from outbreaks. In 2016 and 2017, material will be requested from OIE reference laboratories for listed diseases which are exotic to Europe. Indeed, considering the recent development of molecular tools for the detection of these pathogens, material presently available in the collection of the EURL does not allow providing controls for these PCR tools.	To enrich the different collections	These collections allow the EURL to send reference material to the NRLs on request These collections allow the EURL to prepare material for ILC tests These collections are used for specific or general training especially in histology. These collections are used to develop and validate diagnostic techniques	AH.PT.2 AH.PT.QI	Per year the EURL sends a minimum of 50 samples The EURL is able to send in less than 3 weeks PCR positive controls for the detection of listed pathogens

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	I-6) Provide the National Reference Laboratories with Standard Operating Procedures (SOPs) for each reference technique (available on the EURL website)	The EURL has written SOPs for molecular biology and histopathology diagnosis. These SOPs are regularly updated following technical improvement or regulation evolution. These SOPs are available under pdf format through the EURL website (http://www.eurl-mollusc.eu/)	To facilitate cooperation between laboratories for the harmonisation of SOPs. To help NRLs in writing the documentation of their Quality Management system.	Updated SOPs for reference methods for the detection of listed mollusc pathogens are available for use by NRLs	AH.PT.3	The EURL will update SOPs already available and if necessary will write new ones (if new reference methods are available or new epidemiological situation occurs)
II- Assist NRLs in the diagnosis of disease outbreaks	II-1)Validation of a multiplex Taqman® PCR assay for the detection of *Bonamia* sp. and *Marteilia refringens*	Infections with *Bonamia* sp. and *Marteilia refringens* are listed endemic diseases affecting flat oysters *Ostrea edulis*. In order to detect both parasites in one step, the EURL has developed a multiplex Taqman® assay including an internal control	To finalize the validation of this PCR assay To establish a SOP for this PCR assay To transfer of the technique to interested NRLs	Availability of a new multiplex tool for the detection of *Marteilia refringens and Bonamia sp.* in flat oysters	AH.ANA .1 AH.ANA .2	One new available method for NRLs Validation of the method
	II-2) Development of a multiplex Taqman® PCR assay and *in situ* hybridization (ISH) assay for the detection of *Bonamia ostreae* and *B. exitiosa*	Infections with *Bonamia ostreae* and *Bonamia exitiosa* are listed diseases affecting flat oysters *Ostrea edulis* in Europe sometimes in same oyster population and even in same oysters. In order to detect both parasites in one step, the EURL would like to develop a multiplex Taqman® assay as well as an ISH assay	To test these assays on characterized samples To determine the limit of detection of the techniques To initiate validation of these techniques	Validation of the Taqman multiplex assay and *in situ* hybridization (ISH) assay	AH.ANA .1	Produce new available PCR and ISH methods for NRLs
	II-3) Development of RT-PCR assay for the detection of parasite RNA (to assess parasite viability)	The detection of parasite DNA does not necessarily reveals the presence of live and infectious parasites. In order to detect live parasites (*Bonamia ostreae*/*Marteilia refringens*) only, the EURL would like to develop a PCR tool targeting parasite RNA enabling to assess live parasites load.	To design primers targeting parasite RNA To optimize PCR conditions for the detection of parasite RNA from oyster tissue and water samples	Development of a RT-PCR tool for the detection of live parasites	AH.ANA .1	Develop a new RT PCR tool for NRLs

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	II-4) Comparison of testing samples and pools by PCR for the detection of some listed mollusc pathogens	Testing samples by pool allows saving time but might contribute to decrease the sensitivity of PCR assays. The EURL plans to initiate a comparison between testing samples versus pools for the detection of some mollusc pathogens including Marteilia refringens and Bonamia sp. by Real Time PCR.	To evaluate the possibility to pool samples for the detection of some mollusc pathogens	Provide recommendations regarding pooling samples for the detection of mollusc pathogens	AH.PT.3 and AH.ANA .1	Provide recommendations regarding pooling samples for the detection of some listed mollusc pathogens
	II-5) Identify and characterise mollusc pathogen isolates on NRLs' request	The EURL regularly receive samples from NRLs for investigations and diagnosis	To assist NRLs for screening or confirmation diagnosis	Confirm detection of listed or emergent pathogens		Per year the EURL receive a minimum of 30 samples
III- To facilitate the initial and further training	III-1) Organise and prepare Annual Meetings of the NRLs for mollusc diseases (2016 and 2017) Organize satisfaction survey after the meeting	The EURL will organise the 2016 and 2017 Annual Meetings for NRLs of mollusc diseases in March or April 2016 and 2017. Organize a satisfaction survey for the annual meeting	To share information on - the epidemiological situation of EU countries regarding mollusc diseases - diagnosis of mollusc diseases - other mollusc diseases affecting third countries To evaluate the interest of NRLs in proposed topics To take into account potential negative feedback to organise next annual meeting	Establish the situation of EU regarding mollusc diseases in 2015 and 2016 Training of colleagues from NRLs in mollusc pathology Establish annual meeting agendas which fit with NRLs wishes	AH.NRL. 1 AH.NRL. 2 &3	One to two representatives from each NRLs for mollusc diseases are expected to attend the 2016 and 2017 annual meetings (18 to 36 participants) Attendance rate at least 70% of NRL's 75% of positive feedback (>85%) Agenda of next Annual meetings will take into account potential negative feedback
	III-2) Organise a Technical Workshop (2017) on the diagnosis of some mollusc diseases	The EURL will organise a Technical Workshop combined with the 2017 Annual Meeting. This workshop will include practical sessions which will focus on different topics related to mollusc diseases	To train colleagues from NRLs on the diagnosis of mollusc diseases	Improve diagnostic of mollusc diseases at the EU level	AH.NRL. 5 and 6	One Technical Workshop will be organised in 2017 Attendance rate at least 70% of NRL's

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	III-3) To collect material for and produce a report on the Annual Meetings and Technical Workshop of NRLs for mollusc diseases	The EURL will produce reports of these events including summaries of presentations, discussions and a CDRom of the presentations. These reports circulate among NRLs before their dissemination.	To disseminate information and discussions exchanged during these events	Produce and disseminate reports in two months after the annual meetings		
	III-4) Welcome colleagues from NRLs for specific training or visiting NRL for training	Technical staff of the NRLs and other laboratories involved in the diagnosis of mollusc pathogens is regularly welcome for specific training in the EURL. During these periods, the staff of the EURL helps trainees to improve their practice in mollusc disease diagnosis procedures. If necessary and asked by NRLs, staff from the EURL can also visit NRLs for specific training.	To train colleagues from NRLs To improve level of diagnosis and surveillance of mollusc diseases at the EU level	Training of colleagues from NRLs in mollusc pathology	AH.NRL. 4	The EURL regularly welcomes colleagues from NRLs for specific training. At least one colleague should visit us for training in 2016 and 2017
	III-5) Update mScope®, a tool enabling access to scanned histological slides.	Slides from the EURL collections and from previous ILC based on histo cytopathology are scanned and can be accessible by NRLs through the EURL website thanks to mScope® software. This tool enables training at distance and self testing. The EURL regularly updates information and material available under mScope®	To give NRLs free access to histological slide collections To train colleagues from NRLs in histo cytopathology	Improve results in the ILCs based on histo-cytology Improve the level of diagnosis for mollusc diseases at the EU level	AH.NRL. 5	mScope® is available at any time by the NRLs through the EURL website At least one at distance training session will be organised in 2016 and 2017
	III-6) Update the internet site of the EU Reference Laboratory (http://www.eurl-mollusc.eu/).	The website is very useful to disseminate information on the main activities of the EURL. This website needs to be regularly updated (reports of last meetings, new updated SOPs, ILC registration results, reports...)	To share updated information on mollusc pathology and EURL activities with other laboratories involved in similar activities especially NRLs	Communication, dissemination of information related to EURL activities		EURL website updated at least once a year

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IV- To have trained personnel available for emergency situations and to assist the commission, EFSA, ...	IV-1) Acquire and maintain competency in screening and confirmatory diagnosis techniques	Organisation of annual competency tests for histo-cytopathology and PCR assays	To have qualified staff available to identify and characterize mollusc pathogen isolates on NRLs' request.	To send results in time (within 3 weeks) especially for first attempt tests and in emergency situation	AH. COM. 1 AH.COM.2	At least one person available for histo cytopathology and one person for real time PCR assays Answer any request related to identification and characterization of mollusc pathogens within 3 weeks
	IV-2) Acquire and maintain competency in surveillance and epidemiology of mollusc diseases	Maintain expertise in mollusc pathology by being involved in several European regional projects	To have qualified staff available to assist the Commission in case of specific requests	To answer any specific request related to mollusc disease surveillance	AH. COM. 1	At least one person available for surveillance and mollusc pathology and one person for epidemiology
	IV-3) Acquire and maintain competency in quality management according to CEN ISO 17025	Regular internal and external training on the CEN ISO 17025	To have qualified staff available to assist NRLs to build their quality management system	To visit and assist NRLs if necessary to establish their quality management system or for audit	AH. COM. 1	At least one person available for quality management according to CEN ISO 17025

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	IV-4) Attend international meetings and conferences	Attendance and participation in International meetings and conferences contribute to maintain competency and to improve our knowledge in specific fields	To participate in international conference on aquatic diseases and shellfish pathology such as: the 108th Annual Meeting. Las Vegas, Nevada, USA. February 22 - 26, 2016. the 18th EAFP meeting (September 2017, Belfast, United Kingdom)	To improve our knowledge on aquatic diseases and more particularly on parasitic diseases and on animal health surveillance To present results recently obtained by the EURL	AH.R&D.1	Participation in 2 International conferences per year. At least one person will participate and present results obtained by the EURL in each of these conferences
V- Exchange with competent laboratories in third countries	V-1) Collaboration with colleagues from competent laboratories in third country	The EURL collaborates with colleagues from laboratories involved in investigation and surveillance of mollusc diseases including VIMS (Virginia, U.S.A.), CSIRO (Victoria, Australia) and DFO (MPO, Canada) The EURL is involved in the microcell working group (people working on parasites of the genera *Bonamia* and *Mikrocytos*) The EURL is involved in the working group on Paramyxean parasites (including parasites of the genus *Marteilia*) The EURL regularly welcomes colleagues from third countries	To exchange information related to mollusc health situation in the world especially on listed pathogens exotic to EU To welcome for training staff from laboratories in third countries	To be recognized as a key laboratory on mollusc diseases diagnosis in the world	AH.OIE.1	Collaboration with colleagues from at least 3 different laboratories from third countries

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VI- Design and conduct epidemiological or experimental studies on the listed and emerging diseases in order to improve diagnosis and surveillance at a European level.	VI-1) Determine the geographic distribution and better characterize some pathogens such as *Vibrio aestuarianus or V. splendidus* in mussels	Since 2012, abnormal mortality events of adults *Crassostrea gigas* have occurred in several oyster producing areas in France. Other European countries including Ireland have reported similar events. However, the distribution of *V. aestuarianus* is not well established and needs to be investigated at the European level. More recently, *Vibrio splendidus* has been detected in the context of mortality of mussels in France and Italy. These results raise questions regarding the characterization of these bacterial strains at the European level.	- NRLs have been trained to detect the bacteria during the last Technical Workshop and will be tested regarding their competency to detect bacteria (ILC). These activities should help NRLs to detect bacteria including *Vibrio aestuarianus* and *V. splendidus* during mortality events. In such cases, the EURL will contribute to characterize bacterial strains eventually detected during mortality outbreaks	To have European colleagues ready to detect these bacteria in order to establish their geographic distribution and to get material to better characterize them	AH. R&D.1	Collaboration with some colleagues from NRLs
	VI-2) Evaluate the presence of parasites such as *Bonamia ostreae*/*Marteilia refringens* in the water column/sediment	Surveillance of mollusc diseases currently target molluscs. However parasites may spend some time in the water column and/or sediment during their transmission from infected to non infected molluscs.	The purpose of this work is to test the presence of listed pathogens outside their hosts and to evaluate the relevance of testing water/sediment versus hosts in surveillance programme.	To improve our knowledge regarding the cycle of listed pathogens such as *Bonamia ostreae*/*Marteilia refringens* To identify the best samples to be targeted for diagnosis and surveillance purposes	AH. R&D.1	Improvement of our understanding of parasite life cycle and of the surveillance of mollusc diseases in Europe

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	VI-3) Set readable rules for laboratory diagnostic tests to better inform about the involvement of some listed mollusc pathogens in a mortality event	Different diagnostic laboratory tests are undertaken when a mortality event occurs in mollusc population. Result interpretation is often conducted for each individual diagnostic test, but a global interpretation of all tests' results is often lacking. This could inform about the level of involvement of some listed mollusc pathogens in the observed mortality event.	The objective is to identify associations between different diagnostics test results obtained during the same mortality event.	To define interpretation rules to inform about the level of involvement of some listed mollusc pathogens in an observed mortality event.	AH. R&D.1	Define interpretation rules regarding the level of involvement of some listed mollusc pathogens in an observed mortality event
VII- Administrative support	VII-1) Ifremer administrative support to the coordination of the EURL activities	Contribution to establish financial provisional budget Establishment of the financial report Preparation and following of the orders	To contribute to the administrative coordination of the activities of the EURL			