EURL-Fish training course: Real-time PCR for diagnostics and surveillance of Fish Diseases Copenhagen 15th to 17th September 2014

Description of the course "Real-time PCR for diagnostics and surveillance of Fish Diseases", held at the European Union Reference Laboratory (EURL) for Fish diseases, Frederiksberg (15/9-17/9 2014)

Course content:

The 3-day course is primarily based on presentations in combination with theoretical exercises. There will be no practical lab work.

This year the course will focus on the real-time PCR technique used for diagnostics and surveillance of fish diseases.

The first day of the course the topics will focus on the sample preparation and validation of new real-time PCR assays for the lab. At the end of the day, the participants will be split into groups for theoretical exercises.

The second day will focus on the chemistry of real-time PCR as well as primer/probe design. The theoretical exercises will focus on what the participants has learned earlier in the day, including primer/probe design in silico.

The third day the focus will be on the analysis, interpretation and presentation of results, including familiarising the participants with the MIQE guidelines. The day will end with further theoretical exercises as well as general discussion and evaluation of the course.

A social dinner will be organized the second evening. Further details are provided in the invitation letter.

General course objectives

The course aims to provide participants knowledge on how to properly use real-time PCR in the lab for diagnostics and surveillance of fish diseases. This includes knowledge on how to validate new assays for the laboratory, different chemistries used and primer/probe design. To understand and interpret results as well as troubleshooting. To understand the underlying principles of the assays and to critically review them in order to assess pitfalls and to correctly interpret them. The course will be a mix of presentations by the teachers and theoretical exercises by the participants. The course will focus on assays for listed fish diseases.

Learning objectives

The participants that will have fully followed all the course's objectives should be able to:
Validate new real-time PCR techniques for the lab.
Design and evaluate new primer/probe sets.
Understand the basic chemistry behind PCR.
Understand how to analyse and interpret the results. Knowledge of the MIQE guidelines.
Troubleshooting.

The major focus will be on assays for listed fish diseases.
The course will provide a forum where pre-knowledge, experience and examples can be discussed between participants and teachers, and hereby raise the awareness of pitfalls when using the technique.

Intended learning outcomes

To increase the theoretical knowledge of real-time PCR for analysis of listed fish diseases in the lab. The course also aims at providing a forum where (good and bad) experiences with real-time PCR can be discussed among participants and teachers.

The core elements
Presentations of the real-time PCR technique.
Theoretical exercises by the participants.

Assessment
Each day there will be theoretical exercises by the participants that will be followed-up for discussion and evaluation. At the end of the course a questionnaire for course evaluation will be delivered to all participants.

The course material
Power-point presentations as well as the original scientific papers describing useful assays/techniques will be included in the course binder. For the theoretical exercises laptops will be necessary. Unfortunately these cannot be provided by the course responsible and the participants are required to bring their own from home. It might be necessary to download and install (free or trial) software for use in the theoretical exercises, and information of this will be provided before course start.

The course participants
Since course attendants can come from very different experiences, during the general introduction (day 1), researchers and technicians will be asked to introduce themselves, their pre-experience in the laboratory and their expectations to the course. Their starting point will therefore be mixed as some may have limited theoretical or practical experience, while others may be highly experience in some or all disciplines.

Course responsible and teacher:
Susie Sommer Mikkelsen (Molecular Biologist, PhD), National Veterinary institute, DTU, Denmark (susmi@vet.dtu.dk)

Teachers:
Marc Engelsma, Central Veterinary Institute of Wageningen, UR Lelystad, The Netherlands
(marc.engelsma@wur.nl)
Guest teachers (TBA)

Draft programme (subject to changes)

Day 1	Day 2	Day 3
Section 1 Basic PCR	**Section 2 primers and probes**	**Section 3**
9:00-9:30 Registration and presentation of course **9:30-10:00 Presentation of participants** **10:00-10:45 History of PCR** **10:45-11:15 Coffee** **11:15-12:00 Practical recommendations** **(lab setup, instruments, plasticwares, handling of samples)**	**9:00-10:00 Chemistry** **(SYBR green, Taqman, FRET, others)** **10:00-10:30 coffee** **10:30-12:00 Controls and standards** **(melting curves, standard curves, internal controls, reference genes)**	**9:00-9:45 Analysis and thresholds** **9:45-10:30 Cut-offs** **10:30-11:00 coffee** **11:00-12:00 interpretation and presentation of results**
12:00 – 13:00 Lunch	**12:00 – 13:00 Lunch**	**12:00 – 13:00 Lunch**
Section 1 Sample prep and validation	**Section 2**	**Section 4**
13:00-13:45 Sample preparation **Extraction RNA/DNA** **13:45-14:15 Preventing contamination (UNG...)** **14:15-14:45 Coffee** **14:45-15:45 Validation (Efficiency, specificity, sensitivity)** **15:45-17:00 Theoretical exercises RNA/DNA extraction Validation**	**13:00-14:00 Primer/probe design in silico** **14:00 14:30 Coffee** **14:30-16:30 Theoretical exercises** **Primer design in silico**	**13:00-14:30 Theoretical exercises** **Analysis of results** **14:30-15:00 General discussion and course evaluation** **15:00 Cakes and Goodbyes**