qPCR workshop Leipzig 7-9th March 2005

This workshop is aimed at giving participants a deeper understanding of real-time quantitative PCR and its applications. The courses are intended for persons interested in, or starting out with the technology.

The course covers all aspects in qPCR, from sample preparation to data analysis and is held during 3 days. The course is approximately 50% hands-on as is limited to 15 participants, resulting in very interactive teaching and everybody given the opportunity to try the instrumentation.

After the course participants will be able to plan and perform qPCR experiments themselves, as well as interpret and analyze data.

Preliminary Schedule for TATAA Biocenter qPCR workshop Leipzig 7-9th March 2005

Day 1-Basic qPCR course

09.00-10.00 **Basic PCR and qPCR theory and applications**
-Amplification and detection
-Detection chemistries
-Selected applications

10.00-10.30 **Primer and probe design and considerations**
-What does primer design affect?
-What are primer dimers?
-How do we minimize formation of primer dimers?
-Design of Molecular Beacons and TaqMan probes

10.45-12.00 **Demonstration of setting up qPCR experiment**
-Display of various instrument platforms
-Demonstration of qPCR software

12.00-13.00 **Lunch**

13.00-13.45 **qPCR experiment by participants**
-practical considerations when preparing PCR reactions
-programming qPCR machines

13.45-14.30 **Optimization of qPCR protocols**
-What parameters can/should be optimized?
-A optimization strategy

14.30-15.30 **Data analysis**
-How does qPCR software process the data?
-How are standard curves used and created?
-How are melt curves used?
-Principle of quantification using standard curves
-Principle of relative quantification

15.45-16.15 Analysis of performed qPCR experiments

16.15-16.30 **Discussion and Q&A**
16.30 End of qPCR course day 1.

Day 2-Advanced qPCR course- Quantification, Normalization and experimental design

09.00-09.50 **qPCR quantification strategies**
-standard curves
-relative quantification
-how to compensate for inhibition in biological samples

09.50-10.15 **Normalization of qPCR data**
-What levels of normalization can be used?
-How to choose a good reference gene?

10.30-11.45 **Experiment comparing different quantification strategies**
-relative and standard curve quantification

11.45-12.45 **Lunch**

12.45-14.15 **Quantification calculation examples**
-what effect will efficiency have on quantification
-quantification methods, and equations

14.30-16.15 **Analysis of experimental data**
-differences in quantifications strategies
-pros and cons of different methods

16.15-16.30 **Discussion and Q&A**

16.30 End of qPCR course day 2

Day 3-Advanced qPCR course- Sample Preparation and reverse transcription

09.00-10.00 **Principle of RT and different RT priming strategies**
-Pros and cons of different methods

10.00-10.45 **Principle of RNA and DNA extraction**
-How it works
-Available methods and products suitable for qPCR
-Practical considerations

11.00-11.45 **Reverse transcription experiment using different priming methods**
-Oligo(dt)
-Random Hexamers
-Gene specific primers

11.45-12.45 **Lunch**

12.45-13.30 **qPCR experiment evaluating RT using the generated cDNA**
-Is there a best RT priming method?

13.40-14.30 **Quality Control in qPCR using Kinetic Outlier Detection**

14.45-15.30 **SNP detection. Multiplexing possibilities and problems**.

15.30-16.15 **Analysis of experimental data**
-Which priming method is best?
-How should experiments be planned to take RT priming into consideration?

16.15-16.30 **Probes and Dyes**

16.30-16.45 **Discussion and Q&A**

16.45 End of qPCR course day 3

Lunch, coffee and fruit is included in the course fee. The course is focused on practical issues for qPCR and are partly hands-on, performed by the course participants in the lab (marked in blue).