Annex I

Strengthening capacities of influenza laboratory experts: WHO course for National Influenza Centres on Principles of PCR assays development and validation,

St. Petersburg, the Russian Federation, 25-29 May 2015

Scope and purpose:

WHO-recognized National Influenza Centre at the Research Institute of Influenza of the Ministry of Health, the Russian Federation will organize a 5-day training course on Principles of PCR assays development and validation for specialists in PCR diagnostics of influenza and other viral infections from National Influenza Centers as part of an annual WHO program on training of influenza virologists. The aim of the training course is to strengthen NIC capacities and virological surveillance in the WHO European Region and to provide participants with the competencies to perform molecular identification of circulating human and emerging influenza virus strains and validation of molecular assays for seasonal and emerging influenza viruses.

Objectives:

The main objectives of course – to provide participants with the skills to develop and validate the PCR test-kits and to quality control of influenza PCR diagnostics. Upon completion of the course participants expected to:

- Control the quality of PCR diagnostics
- Design new primers and probes for real-time PCR
- Introduce into laboratory practice test kits based on published sequences of primers and probes
- Develop the algorithm for validation of PCR test kits

Audience:

Specialists from National Influenza Centers responsible for PCR diagnostics of influenza.

Language:

Official language of training course will be Russian.

Preliminary program

Monday, 25th of May

1. Introduction: scope and purpose of the course.
2. Basic principles of PCR and RT-PCR.
3. Realtime PCR: variants of detection (SYBR, TaqMan, MGB probes, Molecular Beacons, Scorpions, LNA-probes).
4. Requirements for the PCR test kits (specificity, sensitivity, robustness, accuracy, precision).

Tuesday, 26th of May

1. Test kits for diagnostics of influenza.
2. Influenza Reagent Resource (IRR): registration and familiarization with the system.
3. Situation analysis: interaction with IRR, ordering the test kits from the WHO Collaborating Centre in Atlanta, USA.
4. Use of the WHO CC test kits in the lab: evaluation of accuracy, precision and robustness.

Wednesday, 27th of May

1. Situation analysis: adaptation of test kits using already published sequences of primers and probes.
2. Validation of test kits after change of reagents: evaluation of sensitivity, accuracy, precision and robustness.

Thursday, 28th of May

1. Situation analysis: development of PCR assay *de novo*.
2. Analysis of target sequences (GenBank and GISAID databases, multiple sequence alignment using MAFFT web-server, sequence alignment analysis).
3. Principles of primer and probe design for TaqMan-based real time PCR, primer and probe design instruments: PrimerBLAST, Ugene.
4. Algorithm of the complete PCR assay validation: evaluation of sensitivity, accuracy, precision and robustness.

Friday, 29th of May

1. Exercise – development of the test kit for detection of the XXX-unknown virus using the TaqMan real-time PCR and design of the validation protocol.
2. Conclusion and certificate awarding.