European Commission

Summary report

NRL Training Workshop on the transferability of qPCR methods for GMOs into a dPCR format, aspects to be considered for accreditation and the use of conversion factors

JRC Geel

12-13 November 2019

The European Commission's science and knowledge service

Joint Research Centre

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NRL Training Workshop on the transferability of qPCR methods for GMOs into a dPCR format, aspects to be considered for accreditation and the use of conversion factors

12 November 2019

09:00 **Welcome and introduction**

- Welcome
- Introduction to the workshop
- Tour de table (all)
- Introduction into the application of digital PCR

10:30 Coffee break

11:00 **Transferability of qPCR methods into dPCR**

- LGC experiences with dPCR accreditation and recommendations for general use
- Workflow, parameters and outcome of validation of a ddPCR system using a CRM
- ILVO experiences on ddPCR

12:30 Lunch

13:30
- BELAC approved procedure to transfer qPCR methods into a dPCR format
- NIB experiences on transferability and accreditation, also for complex matrices
- Preparing for accreditation: implementation and verification of GMO methods on a ddPCR system
- Validation of a duplex ddPCR method

15:00 Coffee break

15:30
- Difficulties encountered in applying ddPCR for food allergen detection
- Multiplex dPCR questionnaire
- Discussion on verification/validation and accreditation (all)

17:30 Departure to restaurant

Networking dinner in restaurant Stokt 21

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13 November 2019

09:00 **Proper use of conversion factors**

- General principle of using conversion factors
- Determination of conversion factors and their uncertainty

10:30 Coffee break

11:00 Practical examples on the use of conversion factors in dPCR: exercises in small groups

12:30 Lunch

13:30 **Discussion and wrapping up**

- Discussion on use of conversion factors
- Summary
- What did you learn?

15:00 Coffee break

15:30 Visit to the JRC Nucleic Acids Analysis laboratory and Reference Materials Production Building

17:00 End of the training workshop

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Summary

The EURL GMFF organised a training workshop for NRLs on the important issues to be considered when transferring a validated event-specific quantitative PCR (qPCR) method for a GM event into a digital PCR (dPCR) format. The aim was to share the common knowledge in this field, to present a number of successful approaches used in different NRL laboratories and to discuss steps for harmonisation of such approaches within the EU.

A total 23 NRL representatives from 17 EU Member States and 5 JRC colleagues from Geel and Ispra participated to the 2 days event, which was organised at JRC-Geel.

The first day was devoted to an introduction to digital PCR, the crucial parameters and (dis)advantages in comparison to qPCR. The recently published ENGL Guidance on dPCR is considered a good starting point. Validation parameters to be assessed were discussed and different approaches were presented. A number of participants demonstrated how they were successful in obtaining converted dPCR methods for GMOs under their scope of accreditation.

During the second day the principle of using a conversion factor (CF) was explained. These CFs are instrumental in converting the dPCR results, expressed in GM DNA copy number ratio, into corresponding, legally relevant GM mass fractions. They are anchored to the CRMs used as calibration standards for qPCR, making qPCR and dPCR results comparable. The use of these CFs, which have been determined for over 50 CRMs, were then exercised in small groups.

An optional visit to the Nucleic Acids Analysis laboratories and the Reference Materials Processing facilities concluded the workshop.

The event was organised with a good balance of presentations, exercises and discussions on the two topics described, and was highly appreciated by the participants.

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