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Title: New approaches in molecular detection of pathogens in food: combined use of SYBR®Green real-time PCR and 'High Resolution Melting' for verocytotoxin *E. coli* detection

Date and time: 15 June 2012 at 10:00

Place: Ghent University (Department of Applied Mathematics and Computer Science), Auditorium A3, Campus De Sterre, Krijgslaan 281, building S9, 9000 Ghent, Belgium.

Abstract

Escherichia coli is a group of bacteria which has raised a lot of safety concerns in recent years. Five major intestinal pathogenic groups have been recognized amongst which the verocytotoxin or shiga-toxin (stx1 and/or stx2) producing E. coli (VTEC or STEC respectively) have received a lot of attention recently. Indeed, due to the high number of outbreaks related to VTEC strains, the European Food Safety Authority (EFSA) has requested the monitoring of the "top-five" serogroups (O26, O103, O111, O145 and O157) most often encountered in food borne diseases and addressed the need for validated VTEC detection methods. Here we report the development of a set of intercalating dye Real-time PCR methods capable of rapidly detecting the presence of the toxin genes together with intimin (eae) in the case of VTEC, or aggregative protein (aggR), in the case of the O104:H4 strain responsible for the outbreak in Germany in 2011. All reactions were optimized to perform at the same annealing temperature permitting the multiplex application in order to minimize the need of material and to allow for high-throughput analysis. In addition, High Resolution Melting (HRM) analysis allowing the discrimination among strains possessing similar virulence traits was established. The development, application to food samples and the flexibility in use of the methods are thoroughly discussed. Together, these Real-time PCR methods facilitate the detection of VTEC in a new highly efficient way and could represent the basis for developing a simple pathogenic *E. coli* platform.

About the speaker

Dafni-Maria Kagkli is a food microbiologist who graduated from the Agricultural University of Athens in 2001 and got further specialized in microbiology during her PhD at the University College Cork in 2006. She has worked on several projects and different laboratories dealing with bacterial strain typing and food pathogen detection. Till January 2012 she had been working at the Institute for Health and Consumer Protection of the Joint Research Centre of the European Commission, where she and her colleagues developed a method for the detection and grouping of the five major serogroups of *Escherichia coli* by means of real-time PCR and High Resolution Melting (HRM) analysis.

