

Same-Day Detection Method of Salmonella Spp., Shigella Spp. and Listeria Monocytogenes with Fluorescence-Based Triplex Real-Time PCR

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Abstract : Faster detection and characterization of pathogens are the basis of the avoid from foodborne pathogens. Salmonella spp., Shigella spp. and Listeria monocytogenes are common foodborne bacteria that are among the most life-threatening. It is important to rapid and accurate detection of these pathogens to prevent food poisoning and outbreaks or to manage food chains. The present work promise to develop a sensitive, species specific and reliable PCR based detection system for simultaneous detection of Salmonella spp., Shigella spp. and Listeria monocytogenes. For this purpose, three genes were picked out, ompC for Salmonella spp., ipaH for Shigella spp. and hlyA for L. monocytogenes. After short pre-enrichment of milk was passed through a vacuum filter and bacterial DNA was extracted using commercially available kit GIDAGEN®(Turkey, İstanbul). Detection of amplicons was verified by examination of the melting temperature (T_m) that are 72° C, 78° C, 82° C for Salmonella spp., Shigella spp. and L. monocytogenes, respectively. The method specificity was checked against a group of bacteria strains, and also carried out sensitivity test resulting in under 10² CFU mL⁻¹ of milk for each bacteria strain. Our results show that the fluorescence based triplex qPCR method can be used routinely to detect Salmonella spp., Shigella spp. and L. monocytogenes during the milk processing procedures in order to reduce cost, time of analysis and the risk of foodborne disease outbreaks.

Keywords : evagreen, food-born bacteria, pathogen detection, real-time pcr

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