

ON-rep-seq as a rapid and cost-effective alternative to whole-genome sequencing for species-level identification and strain-level discrimination of *Listeria monocytogenes* contamination in a salmon processing plant

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Abstract

Identification, source tracking, and surveillance of food pathogens is a crucial factor for the food-producing industry. Over the last decade, the techniques used for this have moved from conventional enrichment methods, through species-specific detection by PCR to sequencing-based methods, whole-genome sequencing (WGS) being the ultimate method. However, using WGS requires the right infrastructure, high computational power, and bioinformatics expertise. Therefore, there is a need for faster, more cost-effective, and more user-friendly methods. A newly developed method, ON-rep-seq, combines the classical rep-PCR method with nanopore sequencing, resulting in a highly discriminating set of sequences that can be used for species identification and also strain discrimination. This study is essentially a real industry case from a salmon processing plant. Twenty *Listeria monocytogenes* isolates were analyzed both by ON-rep-seq and WGS to identify and differentiate putative *L. monocytogenes* from a routine sampling of processing equipment and products, and finally, compare the strain-level discriminatory power of ON-rep-seq to different analyzing levels delivered from the WGS data. The analyses revealed that among the isolates tested there were three different strains. The isolates of the most frequently detected strain (n=15) were all detected in the problematic area in the processing plant. The strain level discrimination done by ON-rep-seq was in full accordance with the interpretation of WGS data. Our findings also demonstrate that ON-rep-seq may serve as a primary screening method alternative to WGS for identification and strain-level differentiation for surveillance of potential pathogens in a food-producing environment.

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