TITLE: RAPID DETECTION OF *Listeria monocytogenes* IN CHICKEN MEAT PROCESSING USING CULTURE ENRICHMENT COMBINED WITH REAL-TIME PCR

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ABSTRACT:

The bacteriological diagnosis of *L. monocytogenes* is laborious and time-consuming (7-10 days), making it difficult to make fast decisions in chicken slaughterhouses that dispatch many batches of chicken meat daily and require agility in the detection of this pathogen in the industrial environment. Real-time PCR (q-PCR) technologies have become powerful diagnostic tools for the analysis of microorganisms in food and can potentially meet the requirements of the industry. Validation of q-PCR based methods for pathogen detection in food is essential if such new technologies are to be adopted on a large scale by the food-testing industry. Real time PCR (q-PCR) is a molecular tool that can meet this demand. In this context, we aimed to evaluate the application of a q-PCR assay in the detection of *L. monocytogenes* in pre-enrichment Fraser broth (CF). The DNA of the bacterial colonies and positive and negative controls were subjected to q-PCR, in triplicate, for the identification of one fragment of the hly gene specific for *L. monocytogenes*. Amplification conditions were adjusted to 45 cycles at 95 °C for 10 min, 95 °C for 15 sec, and 60 °C for 1 min. A limit of detection of 1.34 cfu of *L. monocytogenes*/mL was achieved. A total of 100 knife and board swab samples from chilled/frozen chicken carcasses and chicken meat sections were tested using bacteriological analysis and q-PCR. 5 mL aliquots of all CF (100) were used for DNA extraction by the commercial Dneasy blood and tissue kit (Qiagen®) and 40 ng were submitted to q-PCR analysis. Bacteriological cultures identified *L. monocytogenes* in 14% of samples (14/100) while q-PCR increased detection to 20% (20/100). The q-PCR assay showed a sensitivity of 64%, specificity of 88% and accuracy of 89% when compared to the standard bacteriological method. These high values indicate that this method has the potential to be used as an alternative to the standard method for food quality assurance, providing the rapid detection of *L. monocytogenes* in food products.

Keywords: gene *hlyA*, q-PCR, chicken slaughterhouse

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