Bacteriophage LAMP Salmonella detection

Novel Same-Day method for viable *Salmonella* Enteritidis detection in chicken meat combining phage amplification and LAMP

Alexandre Lamas [1*], Sílvio B. Santos [2,3], Alejandro Garrido-Maestu [4]

1. Department of Analytical Chemistry, Nutrition and Bromatology, University of Santiago de Compostela, Lugo, 27002, Spain.
2. Centre of Biological Engineering, University of Minho, 4710-057 Braga, Portugal.
3. LABBELS—Associate Laboratory, Braga/Guimarães, Portugal
4. Food Quality and Safety Research Group, International Iberian Nanotechnology Laboratory, Av. Mestre José Veiga s/n, Braga, 4715-330, Portugal.

Correspondence:
Alejandro Garrido-Maestu: alejandro.garrido@inl.int

Salmonella enterica is a major foodborne pathogen worldwide. Poultry products, especially eggs and meat, are the main responsible for human salmonellosis cases. Culture-based methods require at least 3 days to detect *Salmonella* positive samples. To facilitate food chain processes and provide a rapid response to food outbreaks, a simple and rapid detection method is necessary. For this purpose, nucleic acid amplification-based techniques are a potential solution. Loop-Mediated isothermal AMPlification (LAMP) has emerged as an alternative to qPCR due to the simple equipment necessary to perform the analysis while allowing the detection of living cells when combined with bacteriophages. The aim of this work was to develop a same-day protocol based in the combination of LAMP and a *Salmonella* phage (vB-SenS_PVP-SE2) to detect viable *Salmonella* Enteritidis cells in chicken meat. Specific LAMP primers were designed to target the capsid and endolysin genes of *Salmonella* phage vB-SenS_PVP-SE2. Two different detection strategies were developed: real-time fluorescence; and colorimetric (naked-eye detection). The LAMP method developed could detect down to 0.2 fg/µL of pure phage DNA and concentrations of viral particles in buffered peptone water (BPW) of 10^2 pfu/mL. After optimization in spiked chicken samples, a 3 h sample pre-enrichment diluted 1/10 in BPW before phage addition to the samples followed by a co-incubation (with phage) of 4 h was established. The proposed method could determine the presence of *S.* Enteritidis in less than 8 h including sample processing, DNA isolation and LAMP analysis with a LOD_{50} of 1.5 cfu/25g and a LOD_{95} of 6.6 cfu/25g, both by fluorescence and naked-eye observation. The results were in close concordance with the reference method for *Salmonella spp.*, the ISO 6579-1:2017. The described method represents a promising alternative for the rapid detection of *Salmonella* in the food chain.