

## ISO Validation for Pathogen Detection in Food and Environmental Samples Utilizing the Hygiena™ foodproof® *Listeria* genus plus *Listeria monocytogenes* Multiplex PCR Assay

Due to the extreme risk of illness from foods contaminated with *Listeria monocytogenes*, many Ready-to-Eat products and food production environments are highly monitored utilizing *Listeria* spp. as an indicator. However, currently available PCR assays require a separate secondary test to identify *L. monocytogenes*, which causes delays for corrective actions and increases laboratory labor.



### Validation Methods

- The study comprises 50 inclusivity and 30 exclusivity strains per target, sensitivity, relative level of detection (RLOD) and collaborative studies.
- Validation of the alternative method was performed and compared to the cultural reference method for:
  - Composite Foods/Ready-to-Eat/Reheat
  - Meat Products
  - Milk and Dairy Products
  - Vegetables
  - Seafood and Fishery Products
  - Environmental Samples
- Samples were enriched in:
  - Half-Fraser Broth 1:10 at 30°C ± 1°C for 24 h - 46 h
  - Actero™ *Listeria* Enrichment Broth 1:7 at 36°C ± 1°C for 20 -24 h.
- Following incubation, DNA extraction was performed with foodproof® StarPrep Two Kit (bulk or 8-strip), then lysates were analyzed by real-time PCR.



### Validation Results

- The validation study (NordVal No. 054) indicated that the alternative method performs equally compared to reference method EN ISO 11290-1:2017 and fulfills the validation criteria according to EN ISO 16140-2:2016.
- The specificity studies yielded:
  - 100% inclusivity of the 50 target strains
  - 100% exclusivity of the 30 non-target strains
- Additionally, the acceptability limits for the sensitivity and the Relative LOD studies for all categories and enrichment protocols were met.

### Industry Significance

- The validation of the foodproof® *Listeria* multiplex assay provides many industries with the ability to quickly screen for *Listeria* and then immediately identify if corrective actions need to occur for *Listeria monocytogenes* in a single test.
- The foodproof® assays have a wide range of extraction options and compatible instruments like the ABI7500 and BAX® Q7 Systems.
- Ability to utilize a single assay improves operational efficiencies throughout food production and environmental monitoring, as well as high throughput testing laboratories.