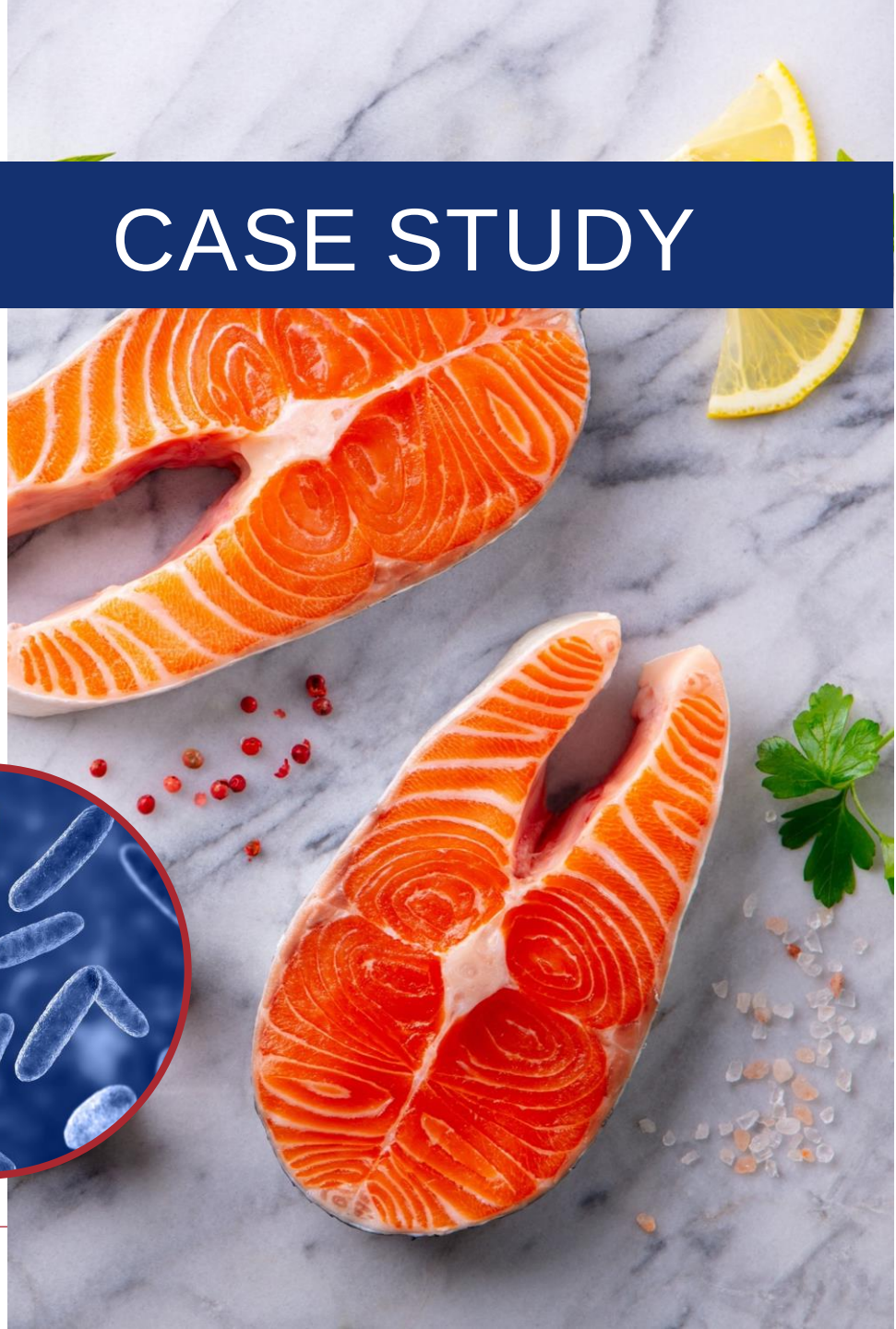
CASE STUDY

Identifying and Preventing Foodborne Dangers With Microfluidics Technology



FOODBORNE ILLNESS IDENTIFICATION AND PREVENTION

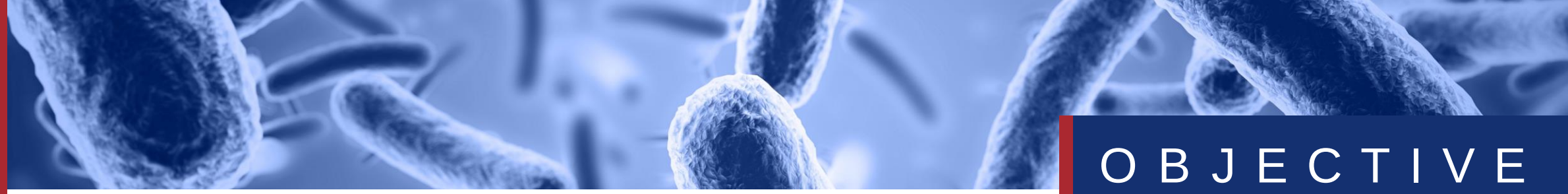
CONTENT

STUDY 1 Insights in ***Listeria monocytogenes*** for better surveillance and contamination control

STUDY 2 Distribution and virulence of ***Salmonella*** in floodwaters

STUDY 3 Detection of the 30 most common clonal complexes found in ***Listeria monocytogenes*** enabling food safety measures

STUDY 1



OBJECTIVE

Identification by high-throughput real-time PCR of 30 major circulating *Listeria monocytogenes* clonal complexes in Europe

Felix, B. et al. *Microbiology Spectrum* (2023)

Foodborne illness caused by the bacterium *Listeria monocytogenes* can typically be classified into a set of major clonal complexes that account for the majority of outbreaks; the number of different complexes poses a challenge to food safety efforts. To address the need for a rapid and reliable method to identify the 30 major clonal complexes, scientists at ANSES Paris developed a high-throughput qPCR assay using the Biomark™ system that can accurately identify these complexes.

KEY TAKEAWAYS

- An assay was developed on the Biomark system, then optimized for conventional multiplex qPCR. A comparison of the assays demonstrated complementarity of the two methods.
- The two assays are of great interest to the food industry and public agencies for tracking *L. monocytogenes* food contamination and can be used for frontline identification of *L. monocytogenes* isolates prior to whole genome sequencing.
- Better surveillance reveals bacterial distribution in food, identifies strains with the greatest health risk and provides additional information to anticipate emerging variants.

Felix, B. et al. "Identification by high-throughput real-time PCR of 30 major circulating *Listeria monocytogenes* clonal complexes in Europe." *Microbiology Spectrum* 11 (2023): e0395422.

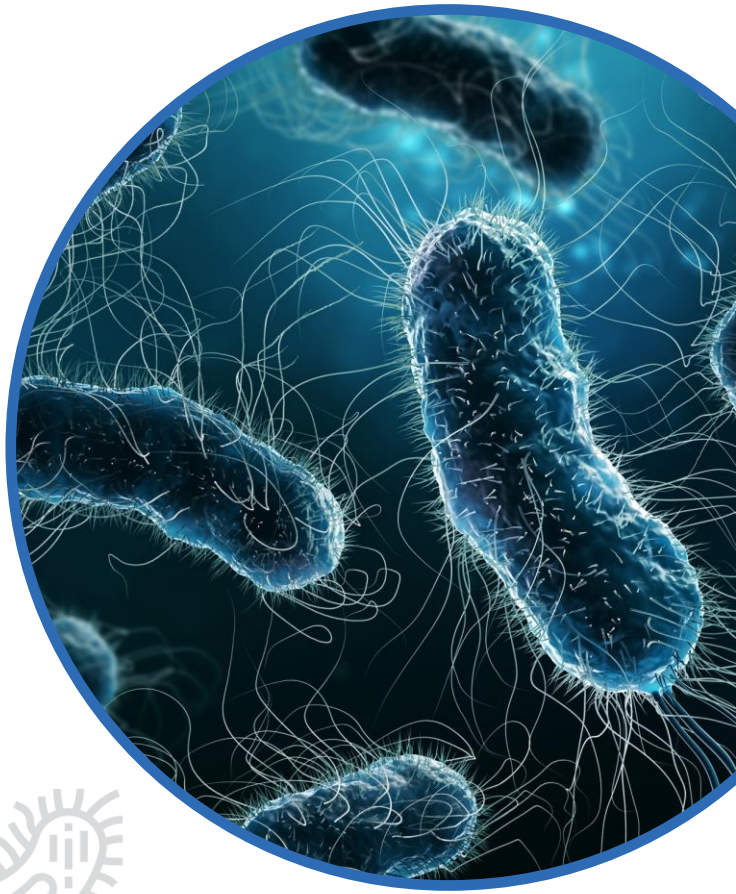
Utilization of Standard BioTools™ products

- Biomark system
- 48.48 Dynamic Array™ IFCs (integrated fluidic circuits)
- Standard BioTools Real-Time PCR Analysis Software

Learn more about the Biomark™ X9 System for High-Throughput Genomics >

BACKGROUND

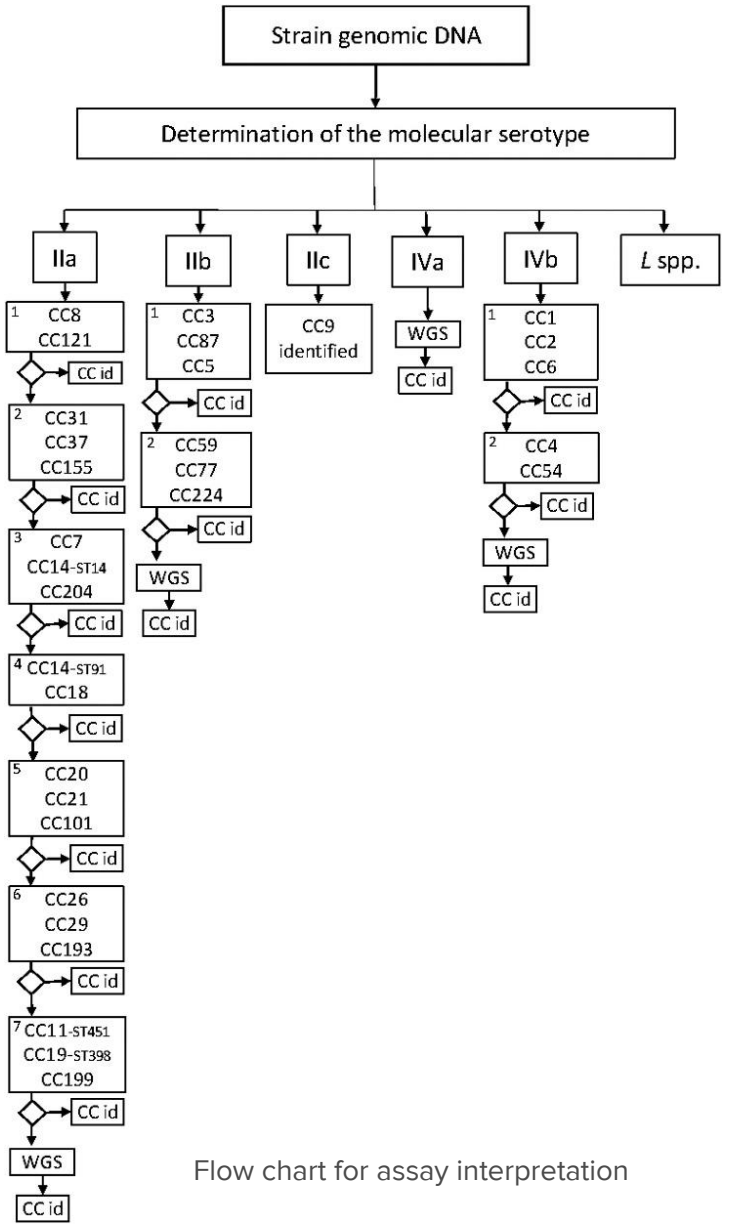
Listeriosis is a serious foodborne illness caused by *Listeria monocytogenes* bacteria. *L. monocytogenes* food contamination can occur from raw plant or animal food matter or from food-processing environments. With its ability to grow at low temperatures, form biofilms and persist in processing plants, *L. monocytogenes* poses a significant challenge to food safety. Current detection methods are time consuming and costly. qPCR offers a rapid and cost-effective approach with high specificity, sensitivity and reliability.



Felix, B. et al. “Identification by high-throughput real-time PCR of 30 major circulating *Listeria monocytogenes* clonal complexes in Europe.” *Microbiology Spectrum* 11 (2023): e0395422.

RESULTS

- The high-throughput real-time PCR assay provides accurate identification of the 30 most abundant clonal complexes found worldwide to improve management of health risks associated with *L. monocytogenes* with better surveillance and contamination control.
- Overall performance of the assay was consistent and remained unaffected by variations in three different extraction methods, in which all primer and probe sets were effective in amplifying their respective targets.

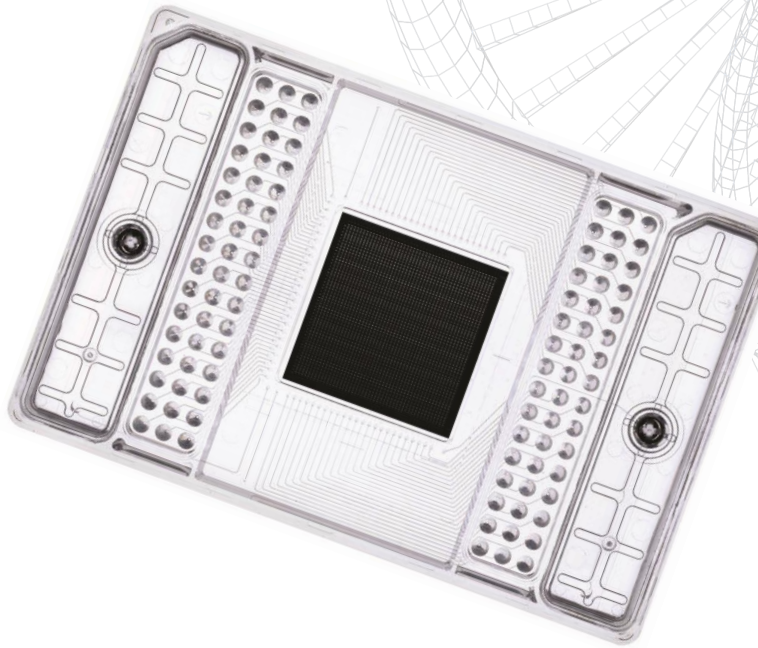


Flow chart for assay interpretation

Felix, B. et al. "Identification by high-throughput real-time PCR of 30 major circulating *Listeria monocytogenes* clonal complexes in Europe." *Microbiology Spectrum* 11 (2023): e0395422.

STUDY DESIGN

The Biomark system was chosen for development of a rapid and reliable pathogen detection assay. The assay was designed from a broad panel of 3,342 *L. monocytogenes* genomes. Sensitivity and specificity were tested on 597 sequenced strains. Performance was evaluated by typing 526 strains collected during surveillance activities. The final assay analyzed 46 strains against 40 real-time PCR arrays in a single experiment. The assay was then optimized for conventional multiplex real-time PCR.



48.48 Dynamic Array IFC

Learn more about IFCs >

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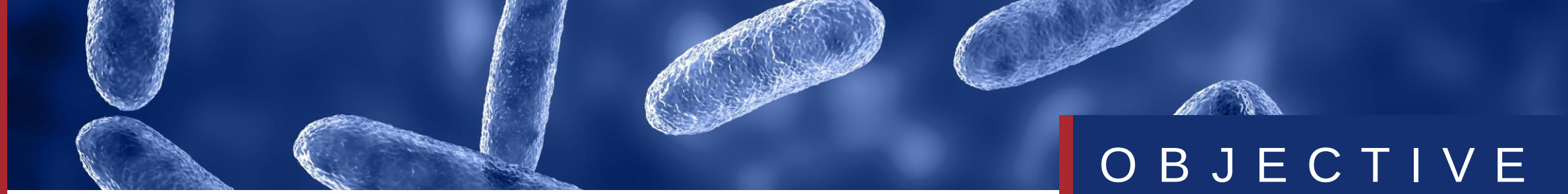
STUDY 2



OBJECTIVE

Distribution and antibiotic resistance profiles of *Salmonella enterica* in rural areas of North Carolina after Hurricane Florence in 2018

Mao, Y. et al. *GeoHealth* (2021)

A waterborne and zoonotic pathogen that is also the cause of the most foodborne bacterial outbreaks in the US, *Salmonella* is a major threat to public health. And the growing frequency and magnitude of changing environmental conditions can introduce pathogens to new areas, such as bodies of water or farmland. Researchers at the University of Illinois investigated the distribution and antibiotic resistance profiles of *Salmonella enterica* in floodwaters from rural North Carolina after Hurricane Florence using microfluidics-based qPCR to ensure proper flood management and protect exposed herds.

KEY TAKEAWAYS

- Antibiotic resistance genes (ARGs) were prevalent in the *S. enterica* isolates, while the antibiotic resistance *ttrC* gene was increased in some but not all flooded areas.
- Microfluidics-based qPCR enables accurate quantification of DNA in water with increased throughput and sensitivity and reduces costs associated with these types of studies.
- qPCR successfully detected *Salmonella enterica* distribution in addition to the prevalence of virulence gene *ttrC* in unflooded and flooded areas.

Utilization of Standard BioTools products

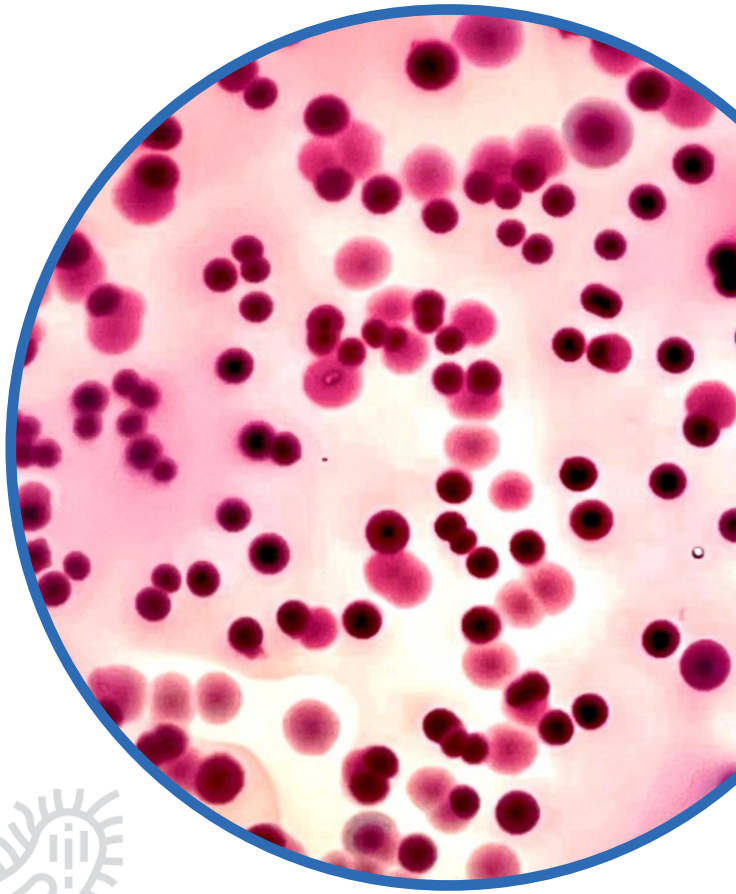
- Biomark system
- 96.96 Dynamic Array IFCs
- Standard BioTools Real-Time PCR Analysis Software

Learn more about the Biomark X9 System >

Mao, Y. et al. "Distribution and antibiotic resistance profiles of *Salmonella enterica* in rural areas of North Carolina after Hurricane Florence in 2018." *GeoHealth* 5 (2021): e2020GH000294.

BACKGROUND

An increase in extreme weather events and climate fluctuations means environmental conditions are likely to change, which affects the metabolism and proliferation of microorganisms. Pathogenic bacteria such as *Salmonella enterica* could subsequently be introduced into new areas, impacting agriculture, infecting livestock, disrupting supply chains and contaminating facilities. Rapid and accurate detection and evaluation of the presence and type of microorganisms in a flooded area could help with environmental management of the area and correct treatment of livestock.



Mao, Y. et al. "Distribution and antibiotic resistance profiles of *Salmonella enterica* in rural areas of North Carolina after Hurricane Florence in 2018." *GeoHealth* 5 (2021): e2020GH000294.

RESULTS

- The study aimed to understand how *Salmonella* might spread during a flooding event and evaluate the presence of ARGs in these bacterial populations.
- Findings suggest that the spread of *Salmonella* and antibiotic resistance can increase after a flood event. This knowledge can help reduce pathogen exposure in herds and determine proper treatments, protecting the food supply chain.
- The virulent *ttrC* gene was detected in 23 out of 25 locations, with wider and higher presence in flooded water versus unflooded water, but antibiotic resistance profiles suggest prevalence in the region regardless of flooding.

Mao, Y. et al. "Distribution and antibiotic resistance profiles of *Salmonella enterica* in rural areas of North Carolina after Hurricane Florence in 2018." *GeoHealth* 5 (2021): e2020GH000294.

STUDY DESIGN

Water samples were collected from rural areas of North Carolina after Hurricane Florence in 2018. Microfluidics-based qPCR was used to quantify the *S. enterica ttrC* gene for each sample and investigate the antibiotic resistance profiles of the *Salmonella* isolates.

Quantification of ARGs in *Salmonella* Isolates
36 aminoglycoside resistance genes
25 macrolide-lincosamide-streptogramin B (MLSB) resistance genes
34 beta-lactam resistance genes
Total 95 sequences of 75 ARGs and 1 16S rRNA

Learn more about IFCs >

96.96 Dynamic Array IFC

Mao, Y. et al. "Distribution and antibiotic resistance profiles of *Salmonella enterica* in rural areas of North Carolina after Hurricane Florence in 2018." *GeoHealth* 5 (2021): e2020GH000294.

STUDY 3

OBJECTIVE

Genetic population structure of *Listeria monocytogenes* strains isolated from salmon and trout sectors in France

Brauge, T. et al. *Heliyon* (2023)

Listeria monocytogenes can cause serious foodborne illness, threatening consumer health. Smoked salmon and smoked trout are examples of foods that do not have to be cooked and thus have the potential to be easily contaminated with pathogenic bacteria. Researchers at ANSES's Laboratory for Food Safety assessed the genetic diversity of circulating bacterial strains among salmon and trout products and in processing facilities to evaluate risk associated with *L. monocytogenes* infection and improve food safety.

KEY TAKEAWAYS

- Findings enabled contamination source tracing and suggested implementation of specific measures to reduce contamination risks and target control interventions more precisely.
- High-throughput qPCR using microfluidics enabled the simultaneous detection and distribution analysis of common *L. monocytogenes* clonal complexes, significantly reducing the cost of experiments and genetic analysis.
- Monitoring epidemiological trends and tracking the evolution of clonal complexes over time help manage risk assessment, predict the emergence of new strains and develop proactive measures to prevent the spread of virulent strains.

Utilization of Standard BioTools products

- Biomark system
- 48.48 Dynamic Array IFCs
- Standard BioTools Real-Time PCR Analysis Software

Learn more about the Biomark X9 System >

Brauge, T. et al. “Genetic population structure of *Listeria monocytogenes* strains isolated from salmon and trout sectors in France.” *Heliyon* 9 (2023): e18154.

BACKGROUND

Listeriosis, a serious foodborne illness caused by *Listeria monocytogenes*, has a lethality rate of 20–30% and impacts at-risk populations such as the elderly, pregnant women, newborns and immunocompromised individuals. Given that listeriosis is primarily contracted from the ingestion of contaminated food, with many outbreaks due to seafood contamination, early detection is essential to control distribution of contaminated products. Additionally, *L. monocytogenes* can survive in extreme conditions, including a wide range of pH values and cold temperatures, resulting in difficulties with elimination in processing facilities.

Brauge, T. et al. “Genetic population structure of *Listeria monocytogenes* strains isolated from salmon and trout sectors in France.” *Heliyon* 9 (2023): e18154.

RESULTS

- Thirteen clonal complexes and one sequence type with variable distribution in salmon and trout samples were identified.
- There were three predominant clonal complexes and one that was detected for the first time in these environments. Less than 0.6% of the isolates belonged to hypervirulent clonal complexes.
- Results detailed the population diversity of the bacteria in the salmon and trout industries.

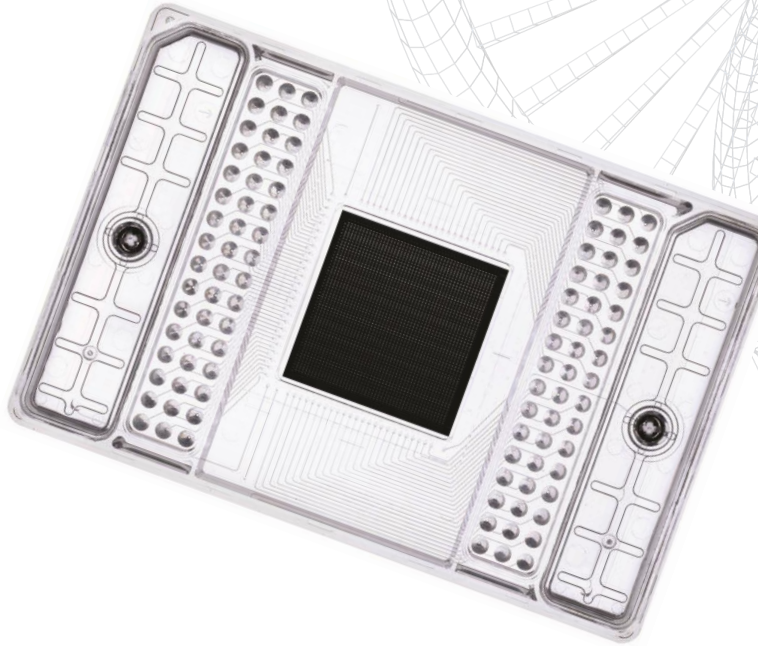
Brauge, T. et al. “Genetic population structure of *Listeria monocytogenes* strains isolated from salmon and trout sectors in France.” *Heliyon* 9 (2023): e18154.

STUDY DESIGN

The genetic structure of 698 strains of *L. monocytogenes* isolated from salmon and trout food products and processing facilities between 2006 and 2017 in France was analyzed. The team identified and evaluated bacterial serogroup, lineage and clonal complexes determined by multilocus sequence typing and high-throughput real-time PCR.

Lineage	Serogroup	Sample Type
I – 22 strains	IIb – 12 strains	Salmon food – 6 strains
		Trout food – 6 strains
	IVb – 10 strains	Trout food – 10 strains
II – 676 strains	IIa – 667 strains	Salmon food – 640 strains
		Salmon processing – 23 strains
		Trout food – 4 strains
	IIc – 9 strains	Trout food – 9 strains

Learn more about IFCs >



48.48 Dynamic Array IFC

Brauge, T. et al. "Genetic population structure of *Listeria monocytogenes* strains isolated from salmon and trout sectors in France." *Heliyon* 9 (2023): e18154.

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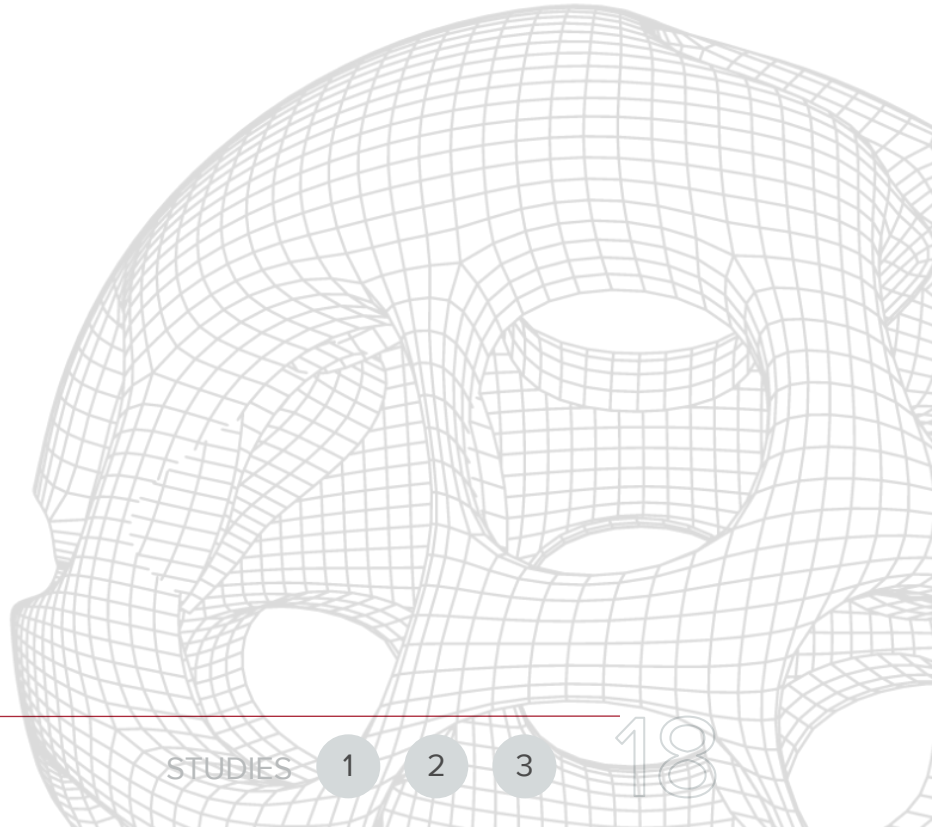

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