

Pathogenic microorganisms

– no thanks: Use of new sequencing techniques in risk assessment and HACCP

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“A food business operator has **primary legal responsibility** for ensuring **food safety**”

– Regulation (EC) No 178/2002 General Food Law

Our vision

Creating the future of dairy to bring **health** and inspiration to the world, naturally

Arla is participating



Microbiological Food Safety Task Force
Expert group “The Use of NGS:
Translation into practice”

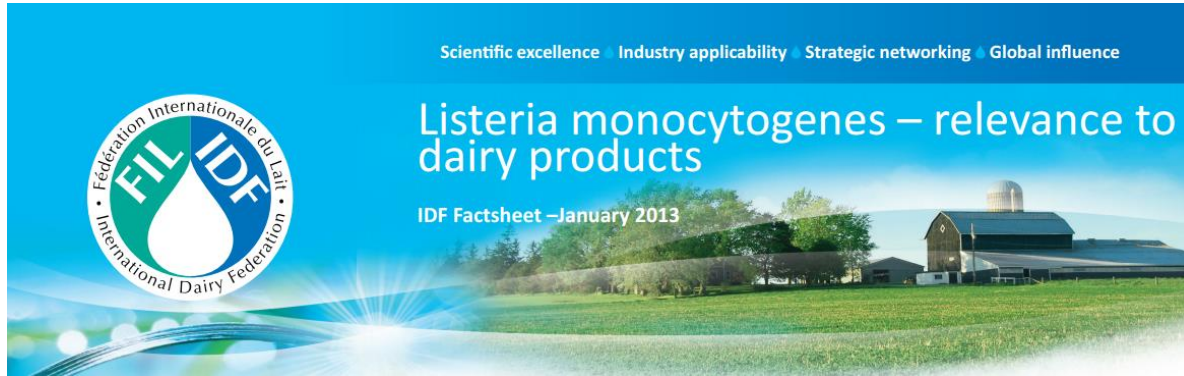
<http://ils.eu/task-forces/food-safety/microbiological-food-safety/>

Nestlé, Unilever, Danone,
Fuji Oil, Mérieux, PepsiCo,
Mars, Mondelēz, **Arla**

FAO, FDA, CDC, TNO,
DTU, U. Nantes, U. Malaya,
Münster U.H., Tokyo U.



Whole genome sequencing (WGS) applications in *Listeria monocytogenes* risk management



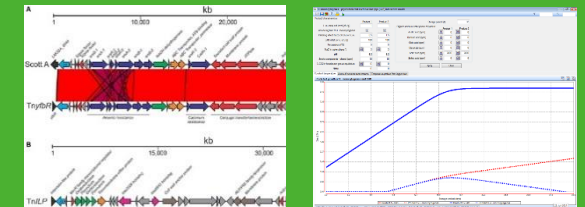
<https://www.fil-idf.org/wp-content/uploads/2016/04/Factsheet-SCMH-Listeria-Monocytogenes.pdf>

Facts about Listeria monocytogenes

- Causing listeriosis, >90% hospitalization, 20% death rate
- Sensitive groups: young, old, pregnant, immunodeficient (YOPI)
- Dairy products can become contaminated at many stages along the food chain
- A particular concern where the product can support the growth of the organism

Genomic and phenotypic characterization and their applications in risk assessment

- Product isolates
- Growth potential
- Virulence or persistence
- **Product matrices**
- **Predictive modeling**

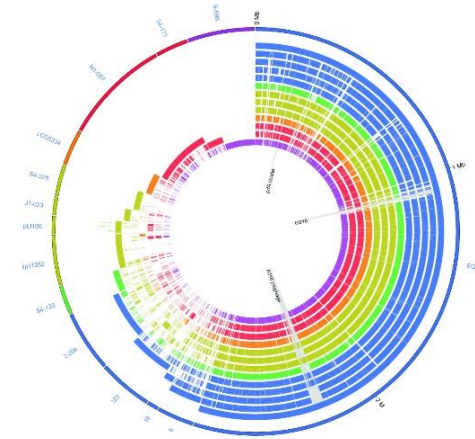


Pathogen Source Tracking

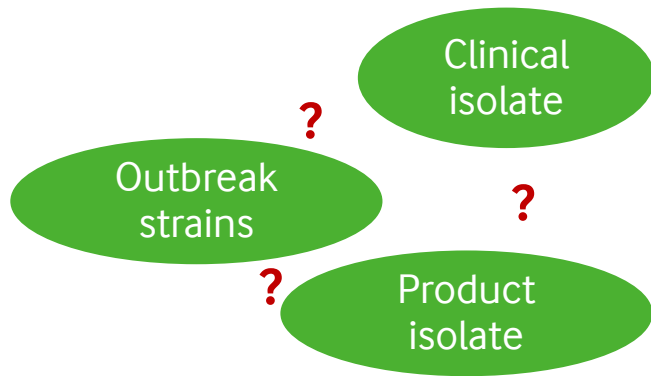
WGS applications in risk management: pathogen source tracking

Following a food safety incident, tracking contamination resource/reservoir of the causative pathogenic isolate(s) becomes one of the main tasks for microbiologists.

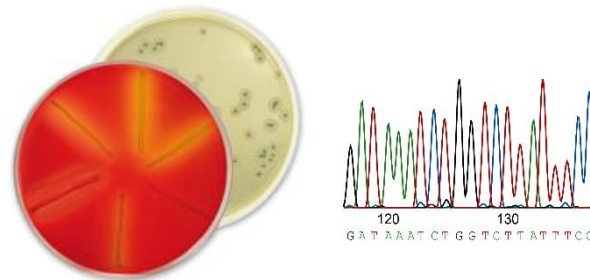
- Farm-to-Fork milk chain
- Dairy companies in Global supply chain
- Outbreaks and historical events



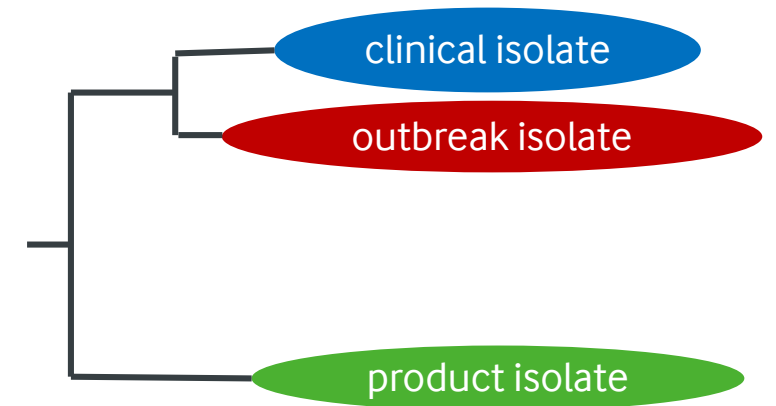
Whole genome alignment



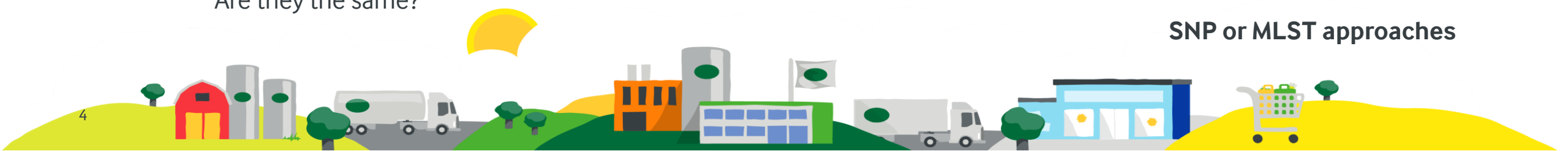
Are they the same?



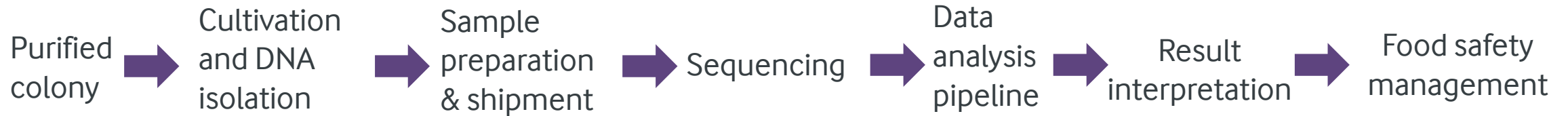
Current identification techniques can only reach genus or species level



SNP or MLST approaches

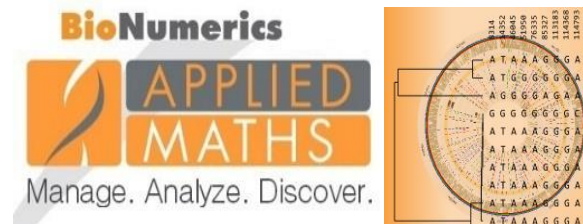


Commercial pipelines and collaborations



- **in a few days**
- **3.000** DKK/isolate

- Standardized sample preparation
- Local or nearby sequencing facility
- Data analysis pipeline and databases are available in house
- Result interpretation is verified



Genome analysis module

Partners needed

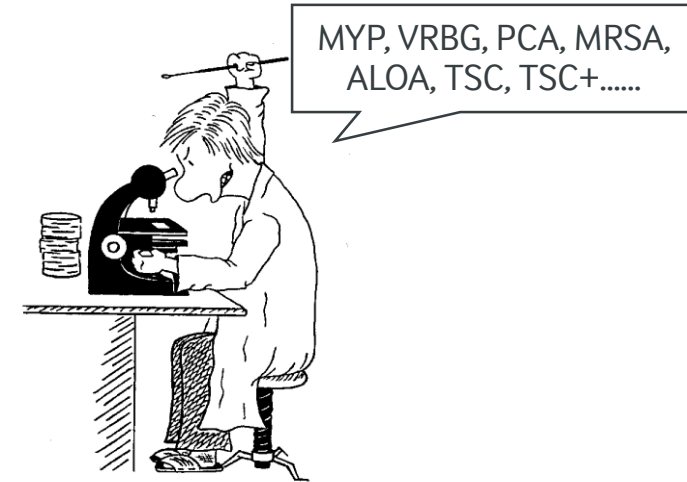


Cultivation-independent investigation

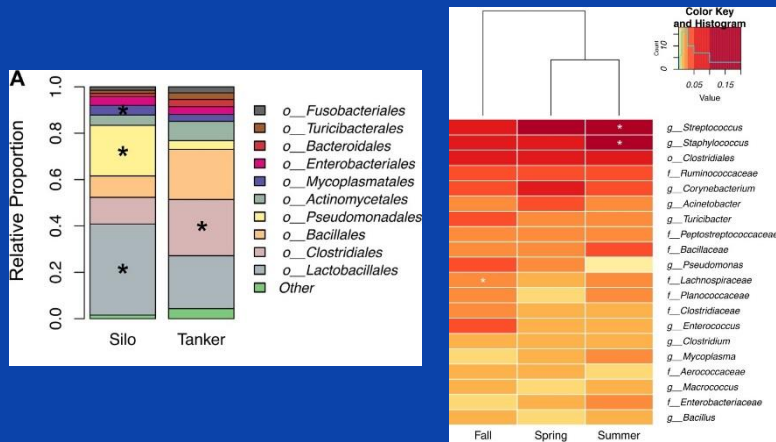


From its humble beginnings as simple meat extract to the advanced science of diagnostic media, the agar plate has always been the workhorse of the microbiologist.

- History of the agar plate¹



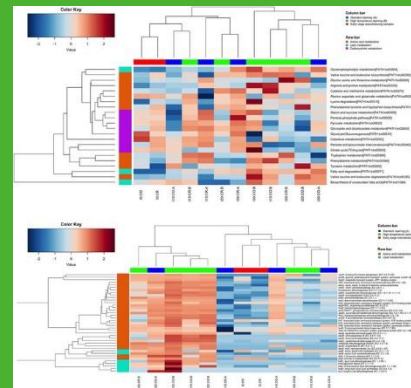
Core and seasonal **Microbiota** of raw bovine **milk** in **tanker trucks** and **silos**²



Metatranscriptomics

reveals temperature-driven functional changes in microbiome impacting

cheese maturation rate³



¹ <http://www.labnews.co.uk/features/history-of-the-agar-plate-01-11-2005/>

² Kable ME, et al. (2016) mBio 7.

³ De Filippis F, et al (2016). Scientific Reports 6: 21871.

Mapping microbial contamination sources inside the plant



Mapping microbial contamination sources inside the brewery as presented in Bokulich et al. .
Increasing color intensity of each surface indicates an increasing relative degree of microbial contamination from that source type.

Bokulich NA, et al. (2015) Mapping microbial ecosystems and spoilage-gene flow in breweries highlights patterns of contamination and resistance. eLife 4: e04634.

Discussion

Can information from NGS be used within an industrial setting to intelligently optimize our assessment of microbial risks and verification of our HACCP plans?

- Food safety management
 - risk assessment
 - pathogen source tracking
 - crisis management
 - control strategy
- HACCP plans
- Routine analysis
- Specification management
- Quality constancy
- Fermentation and culture



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