



# Are all *L. monocytogenes* strains of health concern?

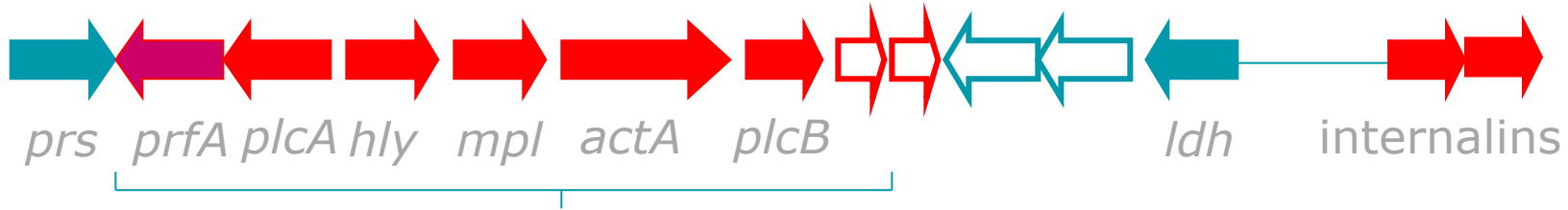
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*Institute for Milk Hygiene, Vetmeduni  
Vienna, Austria; Listeria WG member*

Stakeholder meeting, 19-20 Sep 2017

# IMPORTANT MOLECULAR TRAITS FOR *LISTERIA* VIRULENCE

LIPI-1



LIPI-2: in *Listeria ivanovii* (Dominguez-Bernal et al., 2006)

LIPI-3: streptolysin S (Molloy et al., 2011) clinical lineage 1 isolates

LIPI-4: a cellobiose-family phosphotransferase system (Maury et al., 2016)

**Activity 3:** the comparison of isolates from different compartments along the food chain, and in humans using Whole Genome Sequencing

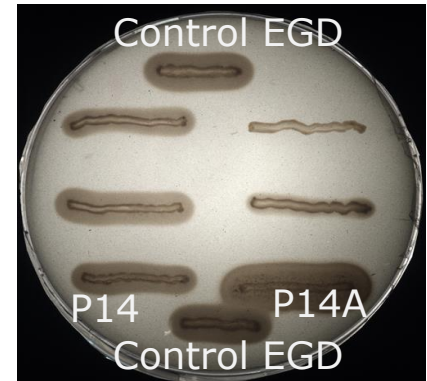
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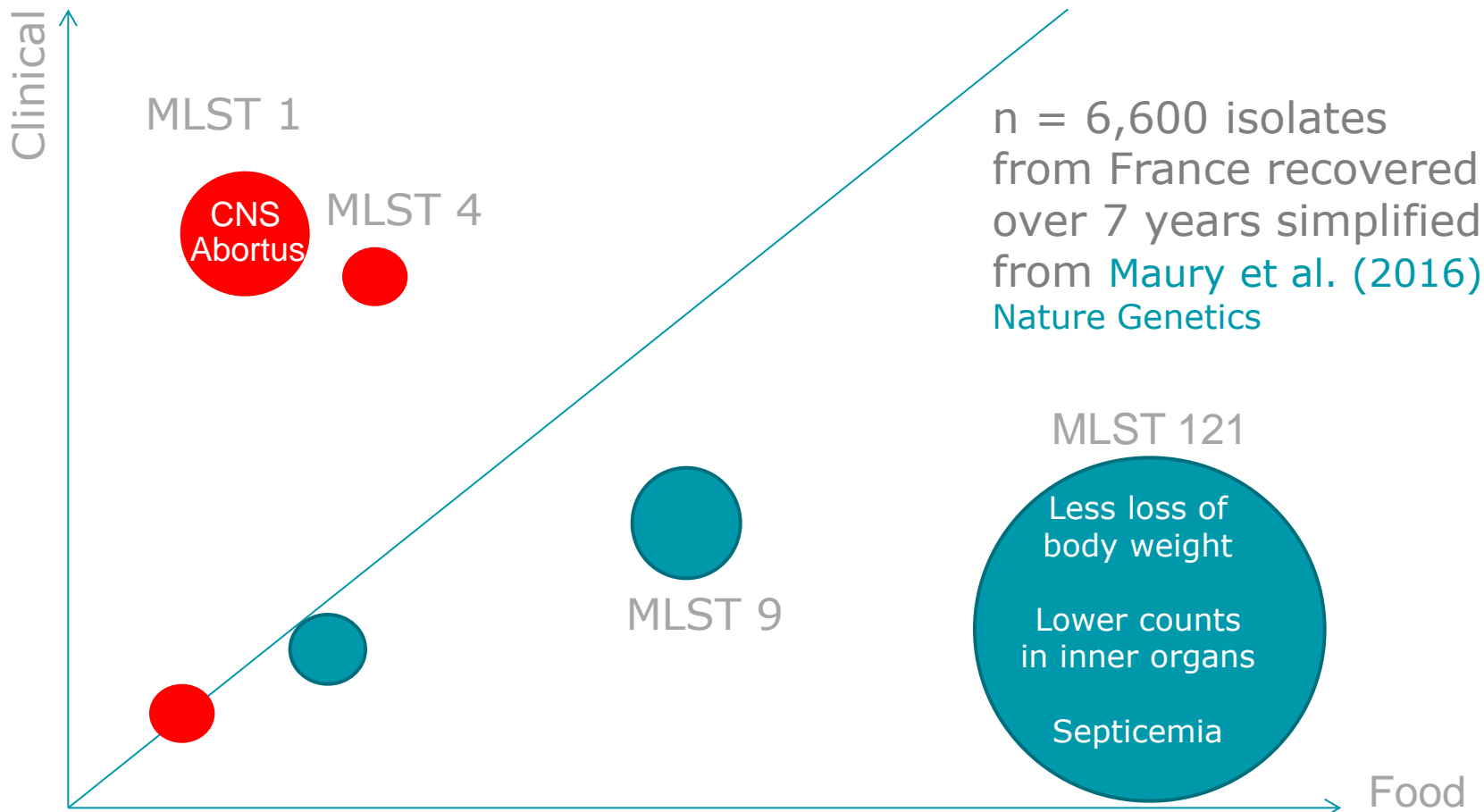
More than 80% of virulence markers (n=115) were present in >95% of strains of lineage I and II

# L. MONOCYTOGENES: VIRULENCE VARIABILITY

- Evidence: Only three (1/2a, 1/2b and 4b) out of 13 serovars cause disease regularly
- Virulence variability shown in different models (cell culture, chick embryos, gerbils...)
- Difficulty to close the gap between virulence (ability to spread a trait/bug in a population) and pathogenicity (ability to cause disease)
- Loss of virulence occurs naturally in  $\sim 0.1\%$  of isolates mostly due to point mutations in gene regulators or genes (Maury et al., 2017)



# L. MONOCYTOGENES: VIRULENCE VARIABILITY



## L. MONOCYTOGENES MLST 121: MOLECULAR FEATURES

- .. possess a transposon mediated tolerance mechanisms against QATS
- .. possess a stress survival islet-2 (Imo0464/Imo0465, acquired from *Listeria innocua*)
- .. other factors outcompeting *L. monocytogenes* non-MLST 121?
- .. are impaired with regard to virulence (*inlA*, *actA*?)

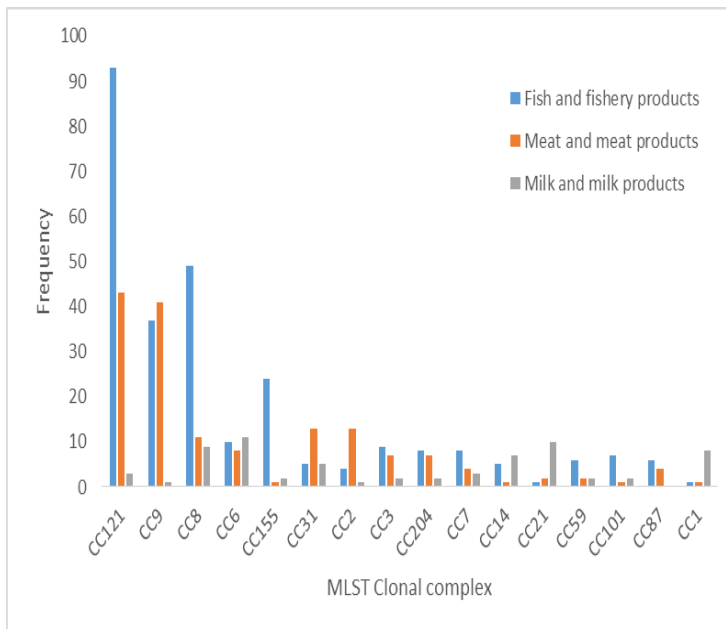
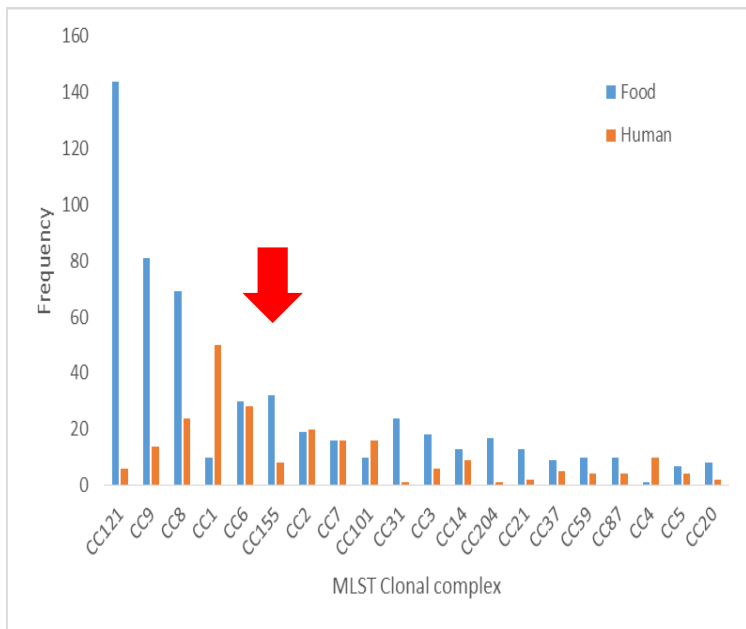
# KNOWN MUTATIONS IN *L. MONOCYTOGENES*

Source	Mutation-type	AA position	Genetic lineage/serotype	
Human, Food	<i>inlA</i>	PMSC (type 2)	I (1/2b)	
Human		PMSC (type 18)	I (4b)	
n. s.		PMSC (type 16, 17)	170, 253	I (1/2b)
Food		PMSC (type 8, 10)	460, 677	II (1/2a)
Human, Food, FPE		PMSC (type 5, 7)	189, 562	II (1/2a, 3a)
n. s.		PMSC (type 15)	77	II (1/2a)
Food		PMSC (type 9)	519	II (1/2c)
Human		PMSC (type 14)	539	II (1/2c, 3c)
Human, Food, FPE		PMSC (type 3)	700	II (1/2a, 3a; 3c)
Food, FPE		PMSC (type 4)	9	II (1/2a, 3a; 1/2c, 3c)
Human, Food		PMSC (type 1)	606	I (1/2b, 4b)+II (1/2a, 3a)
Human, Food		PMSC (type 6)	492	I (1/2b, 4b)+II (1/2a, 3a)
Human, Food, FPE		PMSC (type 12)	576	I (4b)+II (1/2c, 3c)
Food		PMSC (type 11)	685	I (1/2b)+II (1/2c)
Seafood		PMSC (type 13)	527	n. s.
Food (dairy products)		<i>inlB</i> -substitution (9bp)	LRR-region	II (1/2a)
Food (dairy products)	<i>plcA</i> -substitution (12bp)	17, 119, 262	II (1/2a)	
Human	<i>brtA</i> -deletion (188bp)	79	II (1/2c)	
Bovine placenta (abortion)	<i>prfA</i> -deletion	701	II (1/2a, 3a)	
Pet food	<i>prfA</i> -deletion (1kb)	n.s.	II (1/2a)	
Human, Food	<i>actA</i> -deletion (105bp)	n.s.	I (4a, 4b)	

Phosphatidylinositol phospholipase C (PI-PLC); Amino acid (AA); Premature stop codon (PMSC); Leucine-rich repeat region (LRR); n.s. not specified; Source: Burall et al. (2014); Hain et al. (2012); Roche et al. (2005); Rupp et al. (2015); Schwartz et al. (2012); Temoin et al. (2008); Van Stelten and Nightingale (2008); Van Stelten et al. (2010)

# L. MONOCYTOGENES: VIRULENCE VARIABILITY

n = 1,143 isolates; 2 years isolation time frame



**Activity 3:** the comparison of isolates from different compartments along the food chain, and in humans using Whole Genome Sequencing

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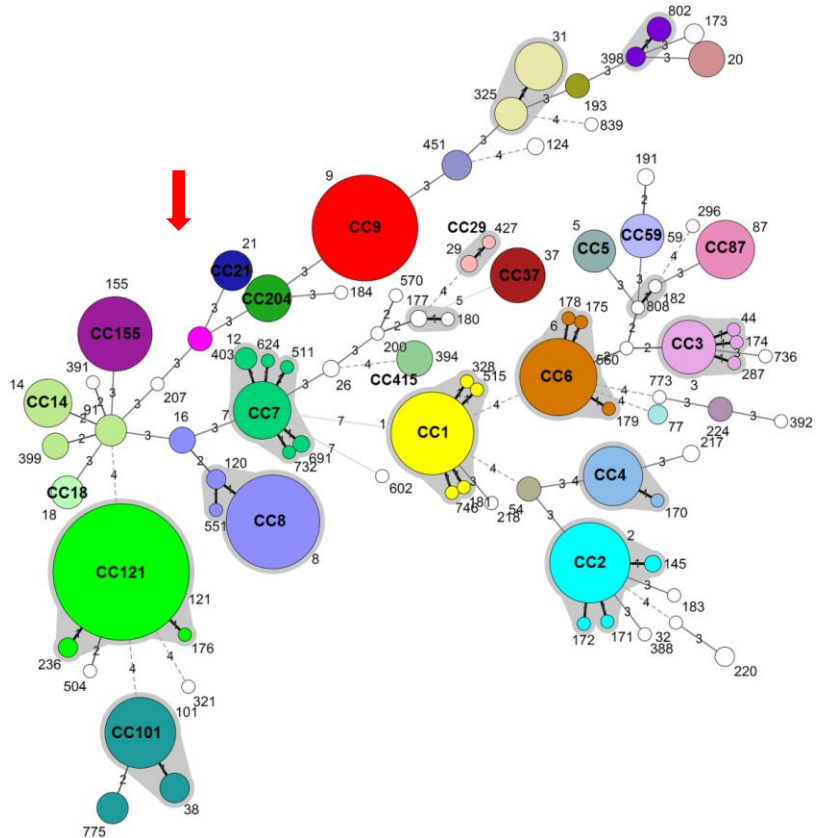
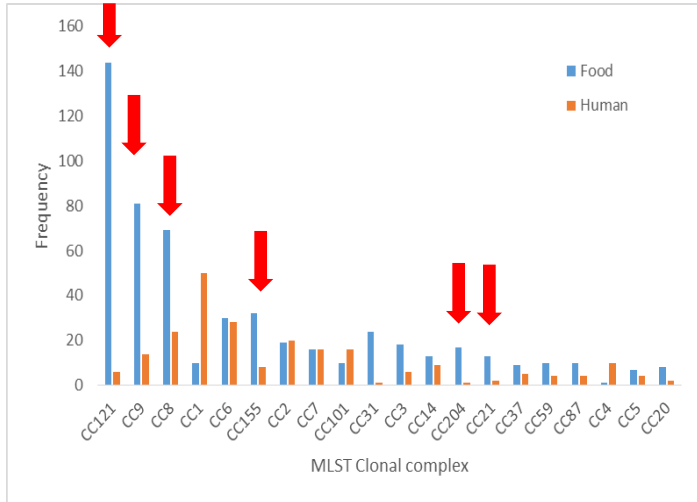
**EXTERNAL SCIENTIFIC REPORT**

Closing gaps for performing a risk assessment on *Listeria monocytogenes* in ready-to-eat (RTE) foods: activity 3, the comparison of isolates from different compartments along the food chain, and from humans using whole genome sequencing (WGS) analysis

Key Author: Neilson<sup>1</sup>, Jones<sup>2</sup>, Spigoni<sup>3</sup>, Krasakova<sup>4</sup>, Kralj<sup>5</sup>, Tomaszewski<sup>6</sup>, Aho<sup>7</sup>, Rasmussen<sup>8</sup>, Corine<sup>9</sup>, Saito<sup>10</sup>, Rasmussen<sup>11</sup>, Lamm<sup>12</sup>, Corbett<sup>13</sup>, Stapanian<sup>14</sup>, O'Brien<sup>15</sup>, Paredes<sup>16</sup>, Pineda<sup>17</sup>, Kralj<sup>18</sup>, Rasmussen<sup>19</sup>

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# VIRULENCE AND POPULATION STRUCTURE



**Activity 3:** the comparison of isolates from different compartments along the food chain, and in humans using Whole Genome Sequencing

- OC/EPSA/BIOCONTAM/2014/01-CT1
- 7/10/2014-7/10/2016

**EXTERNAL SCIENTIFIC REPORT**

40000456, 12 December 2014  
4000020000, 06 July 2015 (12)

**Closing gaps for performing a risk assessment on *Listeria monocytogenes* in ready-to-eat (RTE) foods: activity 3, the comparison of isolates from different compartments along the food chain, and from humans using whole genome sequencing (WGS) analysis**

Eva Heller-Nelso<sup>1</sup>, James T. Spillner<sup>2</sup>, Anindita K. Ghosh<sup>3</sup>, Keith Giger<sup>4</sup>, Tim Dallwitz<sup>5</sup>, Anna Kozlov<sup>6</sup>, Louise Hogg<sup>7</sup>, Sigrun Knapp<sup>8</sup>, Luciana Giamberini<sup>9</sup>, Silvana Basso<sup>10</sup>, Ovidia Kozlov<sup>11</sup>, Francisco Perez Baeza<sup>12</sup>, Ken Walker<sup>13</sup>, Norval Stratton<sup>14</sup>

<sup>1</sup>Novartis Institute for Biomedical Research, Basel, Switzerland; <sup>2</sup>Health Protection Agency, London, UK; <sup>3</sup>Health Protection Agency, London, UK; <sup>4</sup>Health Protection Agency, London, UK; <sup>5</sup>Health Protection Agency, London, UK; <sup>6</sup>Health Protection Agency, London, UK; <sup>7</sup>Health Protection Agency, London, UK; <sup>8</sup>Health Protection Agency, London, UK; <sup>9</sup>Health Protection Agency, London, UK; <sup>10</sup>Health Protection Agency, London, UK; <sup>11</sup>Health Protection Agency, London, UK; <sup>12</sup>Health Protection Agency, London, UK; <sup>13</sup>Health Protection Agency, London, UK; <sup>14</sup>Health Protection Agency, London, UK

n=1,143 isolates recovered over 2 years



# DETECTABILITY AND VIRULENCE

- Hypovirulent strains are underdetected (Gracieux et al., 2003; detection media e.g., antimicrobials added)
- Detection may be limited by the natural microbiota or by other *Listeria* spp. (Cornu et al., 2002; Zitz et al., 2011; Keys et al., 2013; Dailey et al., 2014)
- Strain competition within *L. monocytogenes* is one of the factors related to bias during enrichment (Gorski et al., 2006; Zilelidou et al., 2016b)

# DETECTABILITY AND VIRULENCE

- Outcompetition could not be correlated with the serotype (Gorski et al., 2006; Zilelidou et al., 2016b)
- Lineage dependent detection of strains during enrichment (Bruhn et al., 2005) and a competitive advantage of serotype 1/2a strains over serotype 4b in biofilm formation were reported (Pan et al., 2009)

# SOURCE ATTRIBUTION OF *L. MONOCYTOGENES*

based on 7 locus MLST

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EXTERNAL SCIENTIFIC REPORT

EFSA-14-001

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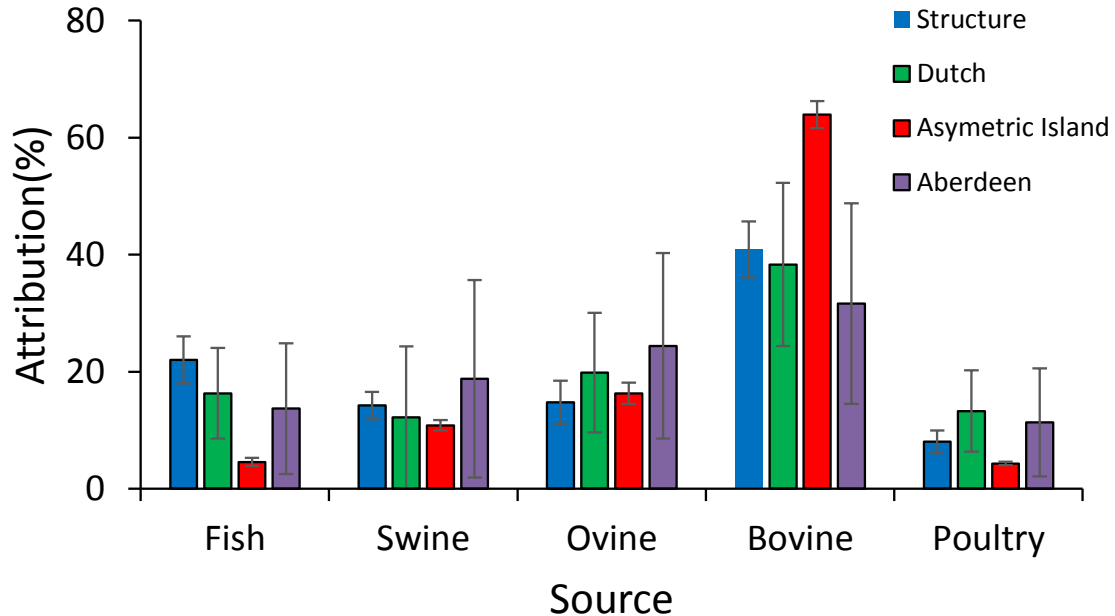
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# CONCLUSIONS

- In principal, all *L. monocytogenes* isolates are of health concern, but the impact to genotypes to the burden of diseases differs
- Sequencing of isolates has shown that most virulence markers are present in most strains-adaptation through genetic mobile elements
- In an increasingly vulnerable population, also the impact of low virulent clones on the burden of disease will increase (albeit not as massive as for high virulent clones)...
- Detectability of low-virulent clones is an actual problem