

Genomic characterization of *Listeria monocytogenes*

Gabriella Centorotola

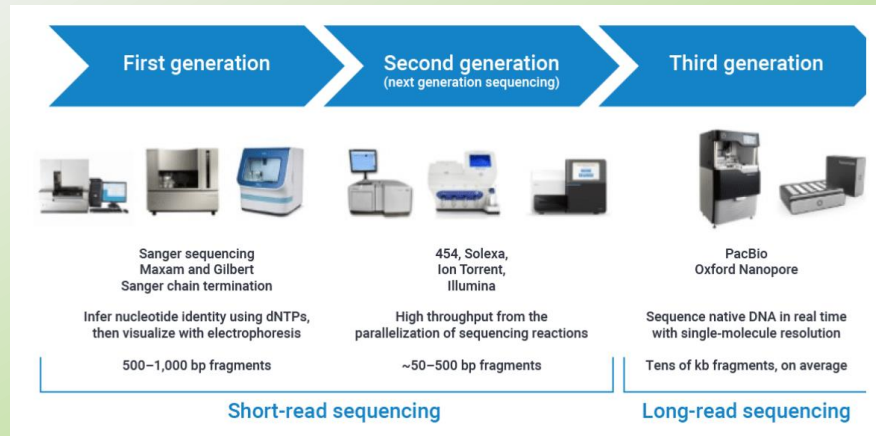
**Food Hygiene Unit
National Reference Laboratory for *Listeria monocytogenes*
Istituto Zooprofilattico Sperimentale Abruzzo e Molise "G. Caporale"**

Introduction

Genomic characterization: a laboratory approach used to study about all the genes in a specific cell type and the way those genes interact with each other and with the environment.

Whole genome sequencing (WGS) provides the most comprehensive data about a given organism, allowing to obtain its **entire genome**.

Using **next generation sequencing (NGS)** can deliver large amounts of data in a short amount of time.



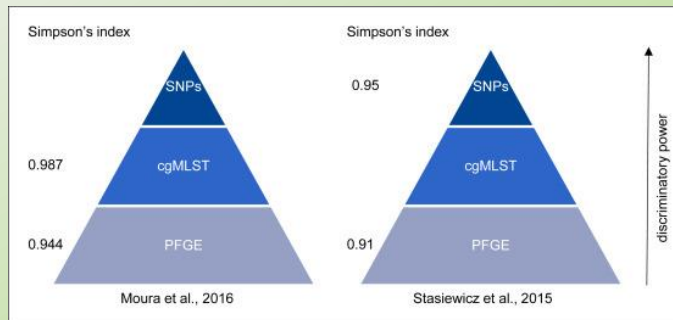
WGS

Nowadays, WGS is the “gold standard” workflow used in public health and food safety to:

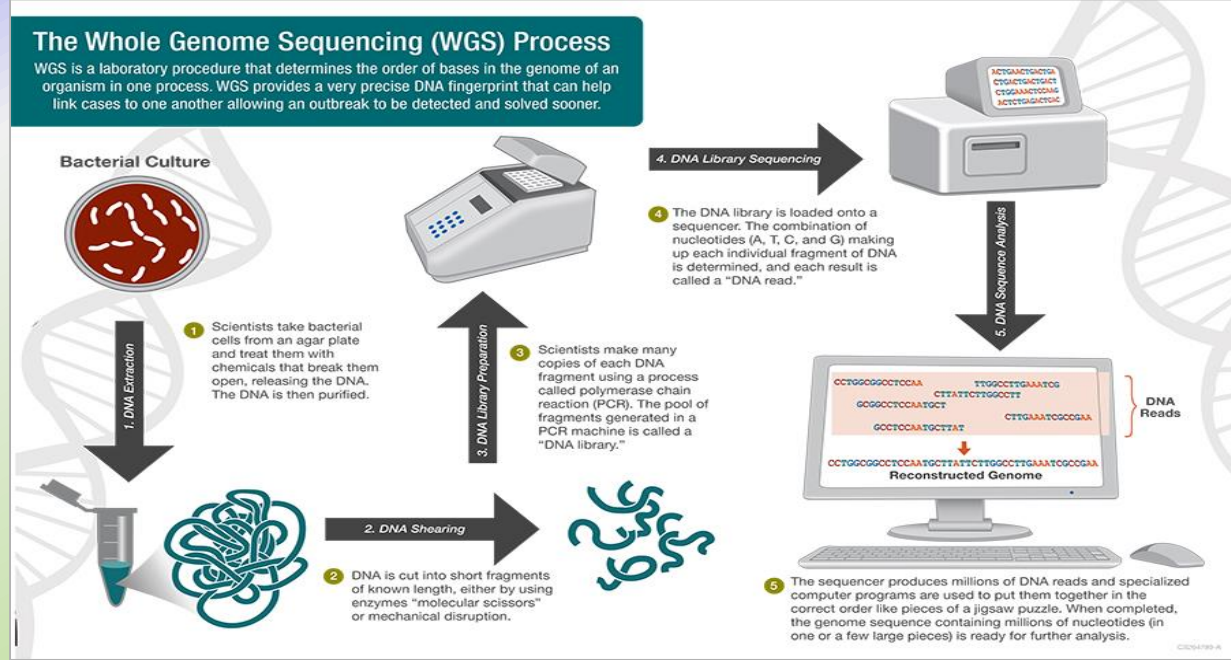
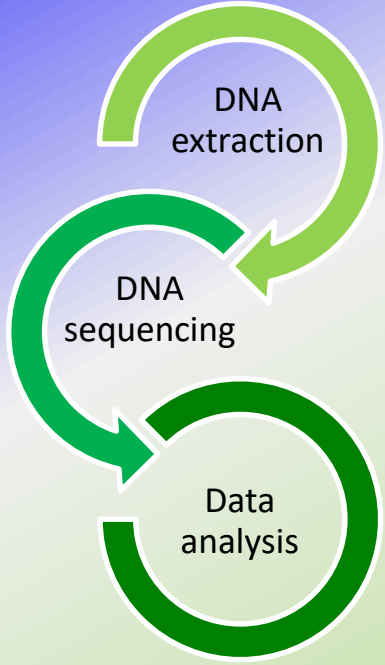
- investigate outbreaks (when they are still contained)
- source attribution for clinical cases
- identify a larger number of clusters (hypothetical outbreaks)

<https://www.cdc.gov/listeria/pdf/whole-genome-sequencing-and-listeria-508c.pdf>

Whole genome sequencing prevents *Listeria* illness



WGS: WET & DRY

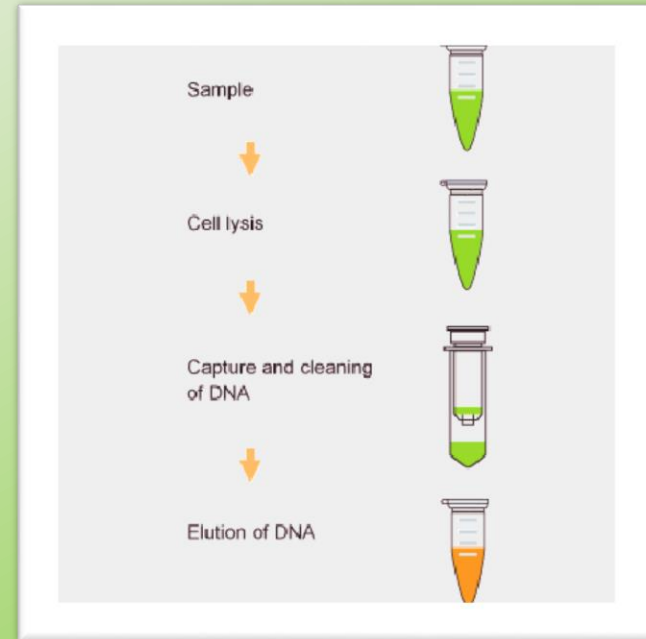


DNA EXTRACTION

Definition: isolation of DNA by breaking the cell membrane and nuclear membrane with the help of chemicals, enzyme or physical disruptions.

The main steps in DNA extraction includes:

1. preparation of biological material
2. cell lysis and separation of nucleic acids from other cell components
3. DNA precipitation
4. wash and DNA elution



DNA extraction: our method for NGS

Columns based DNA extraction

1. In the very first step, the sample is incubated with a **cell lysis buffer** or called a DNA extraction buffer.
2. Along with it, a small amount of **lysozyme** (Gram positive) and **proteinase K** is added to the sample. All the other impurities are removed by centrifugation. DNA remains bounded with silica and other impurities pass through the silica column.
3. Now the DNA can be **washed** twice for improving the purity. The aqueous phase contains the impurities that are discarded by discarding the collection tube.
4. Finally, the DNA is **dissolved** into the AE buffer.



DNA extraction: our method for NGS

Fifth Edition

May 2016

QIAamp® DNA Mini and Blood Mini Handbook

For DNA purification from whole blood, plasma, serum, buffy coat, lymphocytes, dried blood spots (QIAamp DNA Mini Kit only), body fluids, cultured cells, swabs, and tissue (QIAamp DNA Mini Kit only)

Appendix D: Protocols for Bacteria

These protocols have been used successfully for bacteria such as *Escherichia coli*, *Bacillus subtilis*, *Bordetella pertussis* from nasopharyngeal swabs, *Borrelia burgdorferi* from cerebrospinal fluid, and *Legionella pneumophila* from broncho-alveolar lavage. For other bacteria, follow the protocol for Gram-positive bacteria, especially other Gram-positive bacteria, which may be difficult to lyse.

For isolation of bacterial DNA from urine, either follow the protocol for biological fluids, or use the QIAamp Viral RNA Mini Kit. Urine contains numerous unidentified PCR inhibitors. Buffer AVL (included in the QIAamp Viral RNA Mini Kit) is the buffer of choice to destroy these inhibitors.

Some bacteria (particularly Gram-positive bacteria) require pre-incubation with specific enzymes such as lysozyme* or lysostaphin* (e.g., staphylococci) to lyse the rigid multilayered cell wall. In these cases the protocol for Gram-positive bacteria should be used.

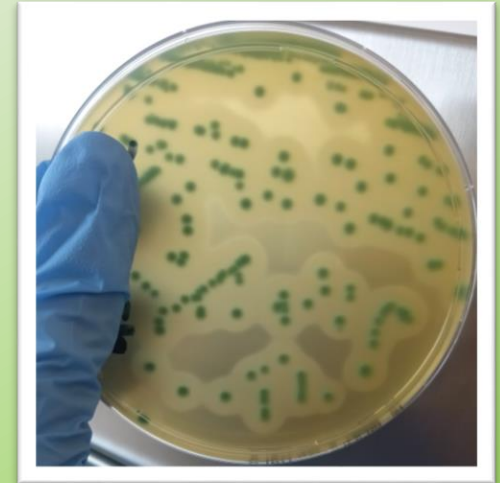
Starting point: **pure culture** from
plate or broth

Incubation with **lysozyme** 37°C 2h
900rpm

Incubation with **proteinase K** 56°C
1h, vortexing every 20 minutes

Extraction of DNA according to the
protocol used for bacteria

Dissolving DNA in AE buffer



ISO requirements for DNA

Standards About us News Taking part Store Q EN

ISO / > 07.100 > 07.100.30

ISO/FDIS 23418

Microbiology of the food chain – Whole genome sequencing for typing and genomic characterization of foodborne bacteria – General requirements and guidance

ISO/CD 23418 (E)

Table A.1 Guidance for development of quality metrics for short and long-read sequencing technologies			
Process	Concern	Guidance	
		Short read technology	Long read technology
DNA extraction	cross-contamination; sample integrity	Broth cultures should be started from a single colony of the isolate being tested.	
		DNA integrity is critical, particularly for long read technologies. Care should be taken to avoid fragmentation of genomic DNA during preparation and storage (e.g. through freeze/thawing)	
DNA quality	Presence of impurities that may negatively impact library construction	Optical density (OD _{260/280}) ratio should be 1.75 - 2.05 and (OD _{260/230}) ratio should be 2.0-2.2	
	low molecular weight DNA may negatively impact library construction	Extraction methods for genomic DNA should be adapted to sequencing platform being used; DNA integrity can be checked on agarose gel or via capillary electrophoresis with appropriate size standards	
DNA quantity	Insufficient input of genomic DNA may result in substandard sequence library	Input DNA quantity should be carefully determined using a DNA-specific, intercalating dye-based fluorescence quantification method prior to further dilution. Minimum quantity needed will be dependent on library kit/sequencing technology used. If modified, this should be supported by validation.	
DNA fragmentation	Sub-optimal fragmentation can result in reduced library yield/reduced coverage	Size distribution of sheared DNA samples should be checked using capillary gel electrophoresis based systems.	

DNA quality

Quality check:

➤ A260 / A280 ratio

It determines the degree of protein purification. **A 260/280 ratio of ~1.8 is generally accepted as “pure” for DNA**; a ratio of ~2.0 is generally accepted as “pure” for RNA. If the ratio is appreciably lower, it may indicate the presence of protein, phenol or other contaminants that absorb strongly at or near 280 nm. Inaccurate ratios may also be encountered at very low concentrations (< 10 ng/μl) of nucleic acids.

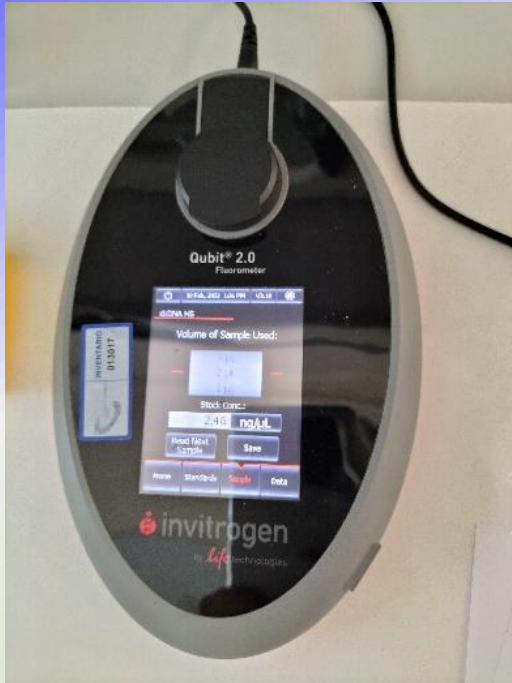
➤ A260 / A230 ratio

Determines the degree of purification of reagent residues. **Expected 260/230 values are commonly in the range of 2.0-2.2**. If the ratio is appreciably lower than expected, it may indicate the presence of contaminants which absorbance at 230 nm.
e.g. EDTA, carbohydrates, guanidine HCL and phenol all have absorbance near 230 nm.



DNA quantity

Fluorometric analysis



The fluorometric measurement of nucleic acids is based upon the use of fluorogenic dyes that bind selectively to DNA or RNA. The signal is measured by fluorometers. Sample is excited with filtered light and the emitted light (at the emission wavelength) is recorded by a detector.

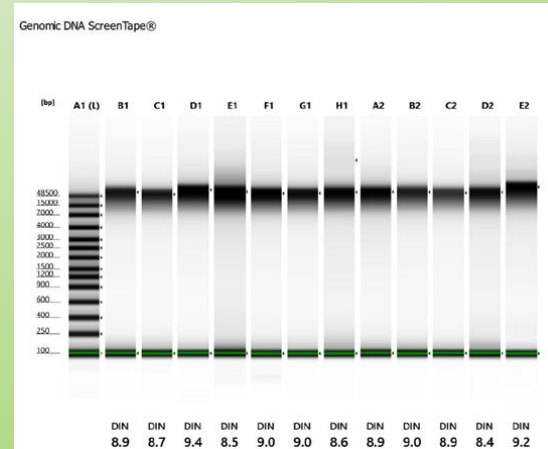
The assay is highly selective for double-stranded DNA (dsDNA) and is accurate for initial sample concentrations from 10 pg/μL to 100 ng/μL. The signal is stable for 3 hours.

DNA integrity

DNA integrity

TapeStation Analysis Software assesses the integrity of gDNA samples and assigns the DNA. Integrity Number (**DIN**), a numerical measure of the gDNA integrity. This value ranges from 1 to 10, where 1 indicates highly degraded gDNA and 10 represents highly intact gDNA.

In our lab **we accept DNA having DIN>7.**



DNA extraction: work in progress

QIAamp 96 DNA QIAcube HT Kit

The QIAamp 96 DNA QIAcube HT Kit combines the selective binding properties of a silica-based membrane with a high-throughput 96-well format, and is designed for fully automated, simultaneous processing of 24–96 samples on the QIAcube HT instrument.

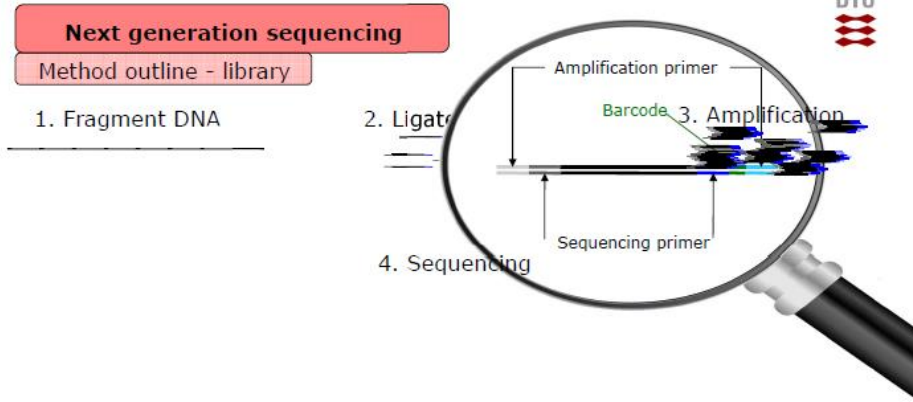
Gram + bacteria require lysis steps with lysozyme and proteinase K.

Vacuum based to binds DNA to membrane and later to elute DNA.

- A minimum of 24 samples is required
- A260/A230 is often < 1,7

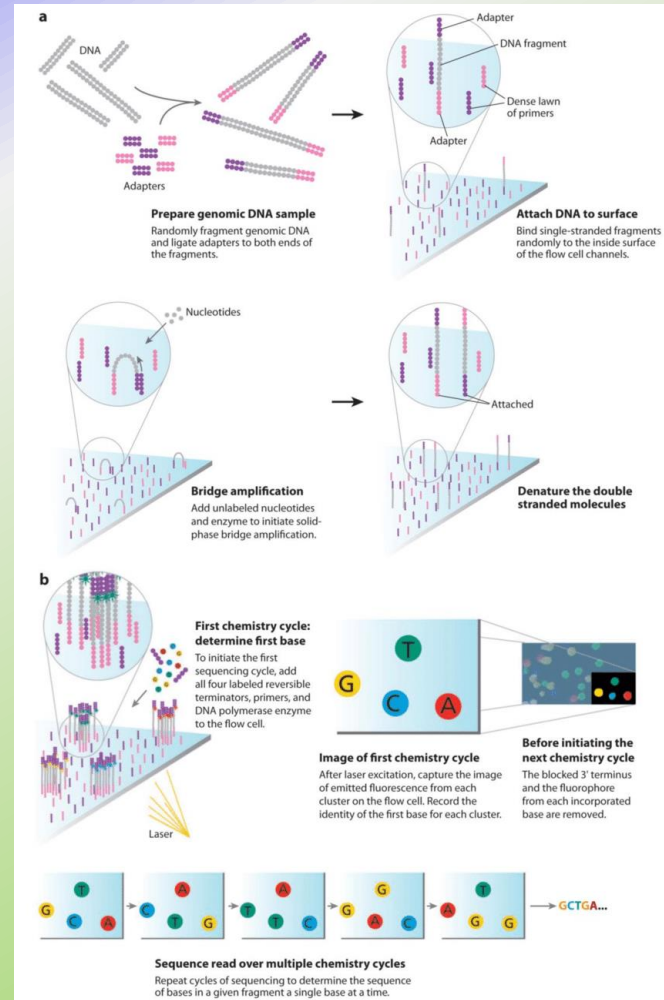


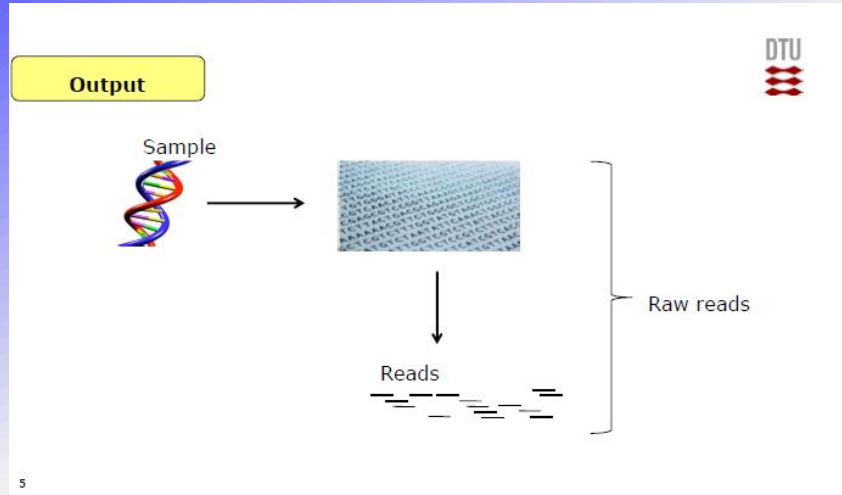
DNA sequencing



The principle of **Illumina** sequencing process: (A) DNA is converted into an Illumina adapter library and amplified by "bridge amplification" on the surface of the flow cell. (B) Amplified molecules are sequenced by the cycle reversible termination chemistry.

DOI: [10.13140/RG.2.2.29564.08327](https://doi.org/10.13140/RG.2.2.29564.08327)

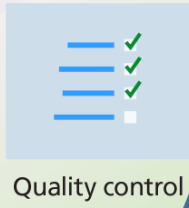




5



Sequencing



Fastq example:

```
@FCC0CD5ACXX:1:1101:1103:2048#ACCGT/1
ACNGTGTTTTTAGTTATGTTTGTGTAAGTTGGGTTTTTTGTACCCATAGCCCAACAAGCCGCGCTTTATGGCGGTTTTTTGTGCTGAAAAGTGGGCGCA
+
BP^ccceggcogihliighiifhifhfdgfhf^efghhfhhegiiliiiiihihinhgeecddccaccWTT^acc[ab_~]`[ _b^`BBBBBBB
@FCC0CD5ACXX:1:1101:1165:2058#ACGTT/1
ACGTTAGCAGAATCGCTTCTGTTGCTTTTTCCACCTGCGACAGACGACCGGACACCGGTTGGGAGATGTTGGGCGAGAATATCGGGCGACGCTGGGAC
+
bb_eceefeggehdagfghhifghighffhifhchghfdhiihfgdceba`a^aacc^V]^baccaccXaaX^bbcccaac[_X]j[a[aaX]
@FCC0CD5ACXX:1:1101:1135:2082#AGCGT/1
AGCGTGACAAACATTTTATTGGCCCGGTTTTATCCAGCTTGAATGCTGACGAAAGAAGATGATGTTGACGACGATGGAGAGAACAATCAGCACGAGATT
+
bbbeeeefggfghiighiigiiiiiiifgfigeghiihhfeffhhfgh_fhgddgegeaceacbdcbcc^aa]^`_bbjbbccccbac_a^bc
@FCC0CD5ACXX:1:1101:1239:2083#AGCGT/1
-----
```

FASTQ format is a text-based format to store both a nucleotide sequence and its quality scores.



Fasta example:

Header/ID: `seq|218682476|ref|NC_011748.1| Escherichia coli 55888 chromosome, complete genome`

Sequence:

```
GTCTGATAGCAGCTTCTGAACTGGTTACCTGCCGTGAGTAAATTAATTTTATTGACTTAGGTCATAA
ATACTTTAACCAATATAGGCATAGCGCACAGACAGATATAAATACAGAGTACACAACATCCATGAAACG
CATTAGCACCACCATTACCACCACCATCACCATTACCACAGGTAACGGTGCGGGCTGACGGGTACAGGAA
ACACAGAAAAAAGCCCGCACCTGACAGTGCGGGCTTTTTTTTCGACCAAAAGGTAACGAGGTAACAACCAT
GCGAGTGTGAAAGTTCGGCGGTACATCAGTGGCAAATGCAGAAACGTTTTCTGCGTGTGCCGATATTCTG
GAAAGCAATGCCAGGCAGGGGCGAGTGGCCACCCTCTCTGCCCCCGCCAAAAATCACCAACCACCTGG
TGGCGATGATTGAAAAACCATTAGCGGCCAGGATGCTTTACCCAATATCAGCGATGCCGAACGATTTT
TGCCGAACTTTTGACGGGACTCGCCGCCGCCAGCCGGGGTTCCCGCTGGCGCAATTGAAAACTTTCGTC
GATCAGGAATTTGCCCAATAAAACATGTCCTGCATGGCATTAGTTTGTGGGGCAGTGCCCGGATAGCA
```

FASTA format is a text-based format to represent nucleotide sequences (single-letter codes).

FASTA is the main format used to easily manipulate sequences by bioinformatics tools.


Table 1 Quality control threshold guidelines for enterica pathogens collected for GenomeTrakr

From: [Optimizing open data to support one health: best practices to ensure interoperability of genomic data from bacterial pathogens](#)

Quality metric	<i>Salmonella enterica</i>	<i>Listeria monocytogenes</i>	<i>Escherichia coli</i>	<i>Shigella sp.</i>	<i>Campylobacter jejuni</i>	<i>Vibrio parahaemolyticus</i>
Average read quality Q score for R1 and R2	> = 30	> = 30	> = 30	> = 30	> = 30	> = 30
Average coverage	> = 30X	> = 20X	> = 40X	> = 40X	> = 20X	> = 40X
De novo assembly: Seq. length (Mbp)	~ 4.3–5.2	~ 2.7–3.2	~ 4.5–5.9	~ 4.0–5.0	~ 1.5–1.9	~ 4.8–5.5
De novo assembly: no. contigs	<=300	<=300	<=500	<=650	<=300	<=300

Timme R.E. et al. **Optimizing open data to support one health: best practices to ensure interoperability of genomic data from bacterial pathogens.** *One Health Outlook* 2, 20 (2020). <https://doi.org/10.1186/s42522-020-00026-3>

Multilocus sequence typing (MLST)

 [alexandra.moura@pasteur.fr] Listeria PasteurMLST submission closed -

20 settembre 2022 15:34

BIGSdb_20220919144659_2003020_65744

Da:

A:

Cc:

Submission status

=====

ID: BIGSdb_20220919144659_2003020_65744

Data type: genomes

Date submitted: 2022-09-19

Last updated: 2022-09-20

Status: closed

Submitter: Francesco Pomilio (listeria@izs.it), Istituto Zooprofilattico Sperimentale dell'Abruzzo e del Molise "G. Caporale

Curator: Alexandra Moura (alexandra.moura@pasteur.fr), Institut Pasteur

Outcome: accepted - data uploaded

Correspondence

=====

Alexandra Moura (2022-09-20 15:33:02+02):

isolate id: 97399

new MLST type: ST3100

new cgMLST type: CT12370

Alexandra Moura (2022-09-20 15:34:21+02):

Thank you for your submission.

Frontiers in [Microbiology](#)

ORIGINAL RESEARCH
published: 27 June 2022
doi: 10.3389/fmicb.2022.930895



Atypical Serogroup IVb-v1 of *Listeria monocytogenes* Assigned to New ST2801, Widely Spread and Persistent in the Environment of a Pork-Meat Producing Plant of Central Italy

OPEN ACCESS

Fabrizia Guidi^{1*}, Cinzia Lorenzetti¹, Gabriella Centorotola², Marina Torresi², Cesare Cammà², Alexandra Chiaverini², Francesco Pomilio² and Giuliana Blasi¹

Core genome MLST (cgMLST)

Core genome MLST (cgMLST) is an extension of the MLST concept to more than seven conserved genes of the species.

- ✓ It consist of a fixed set of conserved genome-wide genes and are usually species specific.
- ✓ All public and stable cgMLST schemes are curated by species experts.
- ✓ It's a gene by gene approach and doesn't require reference genome.

Final result, as for MLST, is the creation of a systematic allele numbering system corresponding to all loci present in the specie's schema.

For *Listeria* the cgMLST is defined on **1748 loci**. Cut off $\geq 95\%$ of loci (minimum of 1660 loci are needed).

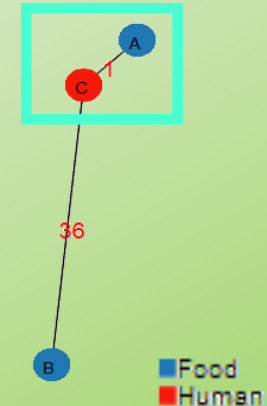
cgMLST schemes may be developed and locally implemented using commercial softwares such as **BioNumerics** (Applied Maths, Sint-Martens-Latem, Belgium), SeqSphere+ (Ridom, Münster, Germany), **BIGSdb** (Pasteur database, France), Center for Genomic Epidemiology (DTU, Denmark) or **in house platform** like GenPat.

cgMLST clustering: MST

Minimum spanning tree (MST) is a graph that connects all the strains together, with the **minimum** possible total edge weight. The minimum spanning tree algorithm is known to be capable of detecting **clusters (≤ 7 allelic differences)**.

It allows the analysis of sequence-based typing methods that generate allelic profiles and their associated epidemiological data (**metadata**).

It provides a graphical visualization of core genomic relationships between bacterial isolates.



Single nucleotide polymorphism (SNP)

- **Single-nucleotide polymorphism (SNP)** is a substitution of a single nucleotide at a specific position in the genome.
- If more than 1% of a population does not carry the same nucleotide at a specific position in the DNA sequence, then **this variation can be classified as a SNP**.

Example:

Isolate 1: AC**G**TTTACC

Isolate 2: AC**C**TTTGCC

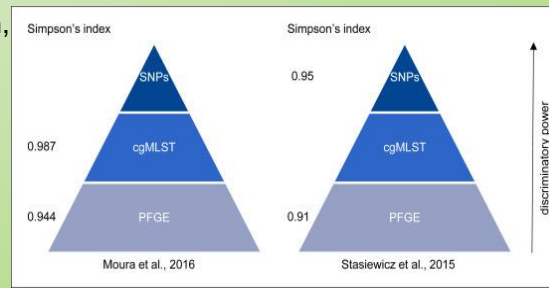
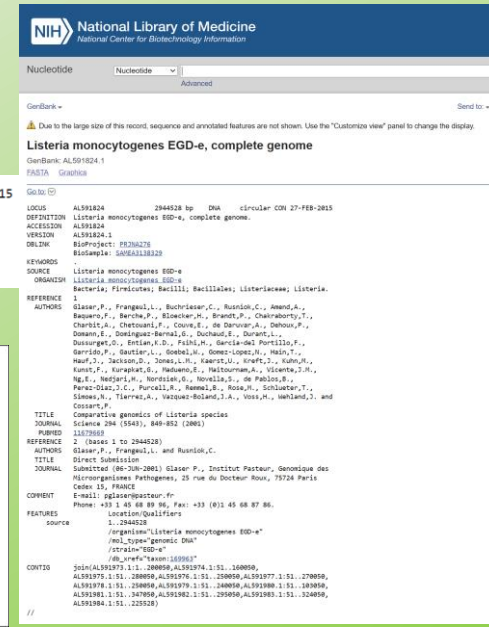
Isolate 3: AC**C**TTAGCC

- **SNP genotyping** is the measurement of genetic variations of SNPs between members of a species.
- **It usually requires a reference genome.**
- It can be used to:

- discriminate the genetic relatedness in a bacterial population,
- trace the evolutionary origin of a bacterial species.

```

LOCUS      AL591824                2944528 bp    DNA    circular CON 27-FEB-2015
DEFINITION Listeria monocytogenes EGD-e, complete genome.
ACCESSION  AL591824
VERSION   AL591824.1
DBLINK    BioProject: PRJNA276
          BioSample: SAMEA9138329
    
```

NLM National Library of Medicine
National Center for Biotechnology Information

Nucleotide

GenBank: AL591824.1

FASTA Graphics

Go to

LOCUS AL591824 2944528 bp DNA circular CON 27-FEB-2015
DEFINITION Listeria monocytogenes EGD-e, complete genome.
ACCESSION AL591824
VERSION AL591824.1
DBLINK Bioproject: PRJNA276
 Biosample: SAMEA9138329

KEYWORDS SOURCE
SOURCE Listeria monocytogenes EGD-e
ORIGIN Listeria monocytogenes EGD-e
 Bacteria; Firmicutes; Bacilli; Bacillales; Listeriaceae; Listeria.

REFERENCE
AUTHORS Glaser,P., Frangeul,L., Buchrieser,C., Rusinol,C., Amend,A.,
Baqueiro,F., Barche,P., Blocker,H., Brandt,P., Chairakooty,T.,
Charif,R., Chetani,S.P., Coumle,L., de Darovar,A., Debou,P.,
Domane,F., Dominguez-Bernal,J., Duchaud,E., Duront,L.,
Dussinger,O., Ertlan,V.D., Falck,P., Garcia-del Portillo,P.,
Garrido,P., Gascler,L., Gebel,A., Gomez-Lopez,M., Hain,T.,
Kauf,J., Jackson,D., Jones,L.H., Kaerst,U., Kraft,T., Kuhn,M.,
Kuntz,F., Kurapkin,G., Madico,E., Hattoum,A., Vicente,J.H.,
Ng,E., Neider,J., Norislan,D., Novilla,S., de Pablo,S.,
Parat-Ouali,C., Purcell,H., Remeis,L., Risse,J., Schlotter,T.,
Simeas,S., Tierney,A., Vazquez-Solano,J.A., Voss,H., Weiland,J. and
Cossart,P.

TITLE Comparative genomics of Listeria species
JOURNAL Science 294 (5643), 840-852 (2005)
PMID 15170168
REFERENCE 2 (bases 1 to 2944528)
AUTHORS Glaser,P., Frangeul,L. and Rusinol,C.
TITLE Direct Submission
JOURNAL Submitted (06-200-2002) Glaser P., Institut Pasteur, Genomique des
Microorganismes Pathogenes, 25 rue du Docteur Roux, 75724 Paris
CENOS 15, FRANCE
COMMENT E-mail: genap@pasteur.fr
 Location:Qualifiers
 Phone: +33 1 45 68 89 96, Fax: +33 (0)1 45 68 87 86.
FEATURES
source /organism="Listeria monocytogenes EGD-e"
 /mol_type="genomic DNA"
 /FTZdb="EGD-e"
 /db_xref="taxon:109963"

CONTIGS 205(AL591977..11..208050,AL591974..1..51..208050,
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AL591981..1..51..247050,AL591982..1..51..230050,AL591983..1..51..324050,
AL591984..1..51..225250)

Database for *in silico* results: BigsdB Pasteur

Welcome to **BIGSDb-Pasteur**, the genomically strain taxonomy and nomenclature platform of Institut Pasteur, powered by the **BIGSDb schemas** developed at Oxford University. This web platform hosts collections of curated, open or private databases of bacterial isolates, genomes and genotypes based on multilocus sequence typing (MLST), whole genome based typing and supplementary schemes (in particular, antimicrobial resistance or virulence genes).

Nomenclatures in **BIGSDb-Pasteur** are universally accessible for referencing and comparing strains, allowing global integration of epidemiological investigations of bacterial pathogens of public health importance, population biology research and surveillance activities in the contexts of One Health and Global Health (see the [BIGSDb-Pasteur policy](#)).

[Edit on GitHub](#)

Databases hosted on this site

Search: Index: Public Filter: All

- Klebsiella pneumoniae**
- Listeria monocytogenes**
- Bordetella**
- Corynebacterium diphtheriae**
- Elizabethkingia**
- Escherichia coli**
- Kingella kingae**
- Leptospira**
- Mycobacterium abscessus**
- Staphylococcus epidermidis**

Listeria monocytogenes

Alleles & profiles database
This typing database contains nomenclatures - allele definitions that provide an identifier for every unique allele sequence, and MLST profiles that index each unique combination of alleles with a unique type ID.

Isolates & genomes database
This table database consists of isolate records containing provenance and phenotype information linked to molecular typing information. These records may also include genome assemblies.

Data submission
Submission of data | Template for Listeria isolates
We appreciate if you can recognize our efforts in the acknowledgments section of your publications.
We thank the Institut Pasteur teams for the creation and maintenance of BIGSDb-Pasteur databases at [BIGSDb-Pasteur.fr](#).

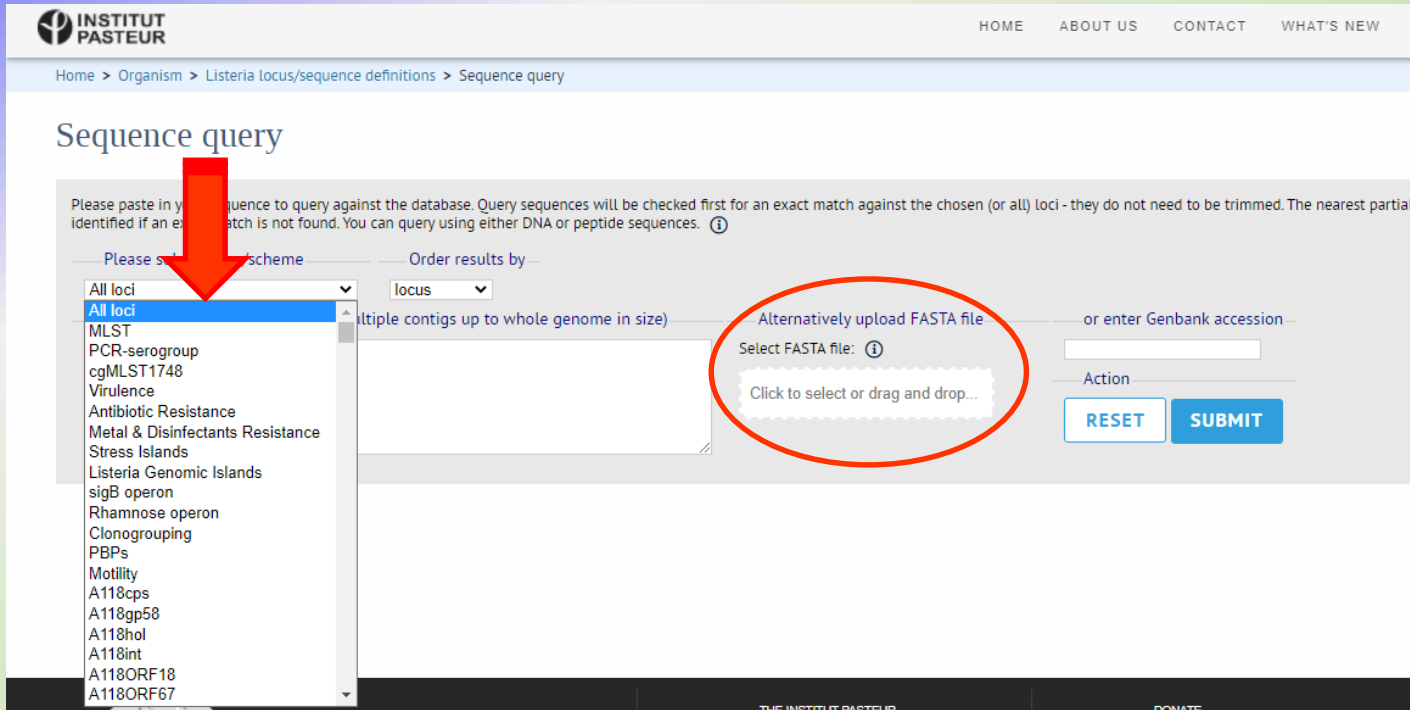
Curators access
Site Curator is performed on a voluntary basis and is based on a community effort.
Sequences & profiles | Isolates

Other Information
BIGSDb manual | RESTful API access link | Nomenclature updates | Guidelines for cgMLST typing | Evaluation of de novo assemblers performance | Primers used for amplification and sequencing MLST
Sequencing collection strains reviewed by Jane Hease and Mark Achtman (Hease et al. 2011, PMID 24274458) | [info_pasteur_type_updates](#)

<https://bigsdB.pasteur.fr/>

<https://bigsdB.pasteur.fr/listeria/>

BigsdB Pasteur: *in silico* MLST, cgMLST... and not only



INSTITUT PASTEUR HOME ABOUT US CONTACT WHAT'S NEW

Home > Organism > Listeria locus/sequence definitions > Sequence query

Sequence query

Please paste in your sequence to query against the database. Query sequences will be checked first for an exact match against the chosen (or all) loci - they do not need to be trimmed. The nearest partial identified if an exact match is not found. You can query using either DNA or peptide sequences. ⓘ

Please select a scheme Order results by

All loci locus

- All loci
- MLST
- PCR-serogroup
- cgMLST1748
- Virulence
- Antibiotic Resistance
- Metal & Disinfectants Resistance
- Stress Islands
- Listeria Genomic Islands
- sigB operon
- Rhamnose operon
- Clonogrouping
- PBPs
- Motility
- A118cps
- A118gp58
- A118hol
- A118int
- A118ORF18
- A118ORF67

Multiple contigs up to whole genome (in size)

Alternatively upload FASTA file or enter Genbank accession

Select FASTA file: ⓘ

Click to select or drag and drop...

Action

RESET **SUBMIT**

BigsdB Pasteur: *in silico* MLST

7 exact matches found.

Locus	Allele	Length	Contig	Start position	End position	Flags
abcZ (lmo2752)	2	537	NODE_2_length_347922_cov_8.428441	160005	160541	
bgIA (lmo0319)	1	399	NODE_11_length_108064_cov_8.553613	107027	107425	
cat (lmo2785)	11	486	NODE_2_length_347922_cov_8.428441	203620	204105	
dapE (lmo0265)	3	462	NODE_11_length_108064_cov_8.553613	49623	50084	
dat (lmo1619)	3	471	NODE_3_length_342314_cov_6.438290	148638	149108	
ldh (lmo0210)	1	453	NODE_8_length_149619_cov_8.590603	16766	17218	
lhkA (lmo1508)	7	480	NODE_3_length_342314_cov_6.438290	26979	27458	

Only exact matches are shown above. If a locus does not have an exact match, try querying specifically against that locus to find the closest match.






MLST



Matching profile



ST: 5
 CC: CC5

Bigbdb Pasteur: *in silico* cgMLST


← → ↻ 🏠 https://bigbdb.pasteur.fr/cgi-bin/bigbdb/bigbdb.pl?db=pubmlst_listeria_ 🔒    🔍 Cerca

Locus	Allele	Length	Contig	Start position	End position	Flags
lmo0002	3	1146	NODE_2_length_518264_cov_91.699089	283351	284496	
lmo0003	2	1344	NODE_2_length_518264_cov_91.699089	284604	285947	
lmo0005	3	1113	NODE_2_length_518264_cov_91.699089	286352	287464	
lmo0006	3	1941	NODE_2_length_518264_cov_91.699089	287513	289453	
lmo0007	2	2529	NODE_2_length_518264_cov_91.699089	289548	292076	
lmo0009	3	519	NODE_2_length_518264_cov_91.699089	293740	294258	
lmo0010	3	969	NODE_2_length_518264_cov_91.699089	294400	295368	
lmo0011	3	972	NODE_2_length_518264_cov_91.699089	295325	296296	
lmo0012	3	1080	NODE_2_length_518264_cov_91.699089	296277	297356	

⌵

cgMLST1748

 **Matching profiles**

Closest profile: cg-15712
Fields: cg: 15712
Mismatches: 8
Loci matched: 1740/1748 (99.5%)

BigsdB Pasteur: *in silico* analysis

[Home](#) > [Organism](#) > [Listeria locus/sequence definitions](#) > [Sequence query](#)

Sequence query

Please paste in your sequence to query against the database. Query sequences will be checked first for an exact match against the chosen (or all) loci - they do not need to be trimmed. The nearest part identified if an exact match is not found. You can query using either DNA or peptide sequences. [?](#)

Please select locus/scheme

Order results by

Metal & Disinfectants Resistance

locus

Enter query sequence (single or multiple contigs up to whole genome in size)

Alternatively upload FASTA file

or enter Genbank accession

Select FASTA file: [?](#)

Click to select or drag and drop...

Action

RESET

SUBMIT

Uploaded file: C1.fasta

1 exact match found.

Locus	Allele Length	Contig	Start position	End position	Flags
Tn6188_qac (ermC)	2 372	NODE_3_length_482505_cov_42.105601	320579	320950	



IZS
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/

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LABORATORY FOR
LISTERIA
MONOCYTOGENES

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NATIONAL REFERENCE
CENTRE FOR
*WHOLE GENOME
SEQUENCING OF
MICROBIAL PATHOGENS:
DATABASE AND
BIOINFORMATIC
ANALYSIS*

BioinfoDB - LNR *Listeria_monocytogenes*. Per visualizzare il Motivo Attivo clicca [qui](#).

5368 Elementi

Codice I	Specie	Host	Matrice	Data Prelievo	Punto Prelievo	Clonal Complex	Sequence Type	Richiedente
2022.TE.32894.1.2	<i>Listeria monocytogenes</i>			23/03/2021		CC3	3	
2022.TE.32893.1.2	<i>Listeria monocytogenes</i>			23/03/2021		CC3	3	
2022.TE.32892.1.2	<i>Listeria monocytogenes</i>			01/03/2021		CC3	3	
2022.TE.31043.1.2	<i>Listeria monocytogenes</i>	SUINO	CARASSA	01/07/2013		CC9	9	
2022.TE.31042.1.2	<i>Listeria monocytogenes</i>	SUINO	CARNE A PEZZI	22/02/2013		CC9	9	
2022.TE.31041.1.2	<i>Listeria monocytogenes</i>	SUINO	CARNE A PEZZI	22/02/2013		CC9	9	
2022.TE.31037.1.2	<i>Listeria monocytogenes</i>	SUINO	CARASSA	18/02/2013		CC9	9	
2022.TE.31035.1.2	<i>Listeria monocytogenes</i>	SUINO	CARASSA	18/02/2013		CC9	9	
2022.TE.31033.1.2	<i>Listeria monocytogenes</i>	SUINO	CARASSA	01/07/2013		CC9	9	
2022.TE.31001.1.2	<i>Listeria monocytogenes</i>	SUINO	COPPA	05/11/2021		CC2	2	
2022.TE.30999.1.2	<i>Listeria monocytogenes</i>	SUINO	COPPA	05/11/2021		CC2	2	
2022.TE.30998.1.2	<i>Listeria monocytogenes</i>	SUINO	COPPA	05/11/2021		CC2	2	
2022.TE.30997.1.2	<i>Listeria monocytogenes</i>	SUINO	COPPA	05/11/2021		CC2	2	
2022.TE.30994.1.2	<i>Listeria monocytogenes</i>		TAMPONE AMBIENTALE (SPONGE BAGS)	01/12/2021		CC2	2	
2022.TE.28684.1.2	<i>Listeria monocytogenes</i>	UOMO	SANGUE	26/02/2022		CC29	29	
2022.TE.28683.1.2	<i>Listeria monocytogenes</i>	UOMO	SANGUE	13/02/2022		CC429	429	
2022.TE.28681.1.2	<i>Listeria monocytogenes</i>	UOMO	SANGUE	10/02/2022		CC429	429	
2022.TE.28680.1.2	<i>Listeria monocytogenes</i>	BOVINO	FORMAGGIO A PASTA FILATA	01/02/2022		CC2	2	
2022.TE.28678.1.2	<i>Listeria monocytogenes</i>	LATTUGA	LATTUGHE E SIMILI (CRESCIONE, DOLCET...	03/02/2022		CC6	6	
2022.TE.28677.1.2	<i>Listeria monocytogenes</i>	POLLO	INVOLTINI	14/09/2021		CC475	504	
2022.TE.28675.1.2	<i>Listeria monocytogenes</i>	POLLO	INVOLTINI	14/09/2021		CC475	504	
2022.TE.28674.1.2	<i>Listeria monocytogenes</i>	BOVINO	CARNE MACINATA	07/09/2021		CC1	1	
2022.TE.28673.1.2	<i>Listeria monocytogenes</i>	BOVINO	CARNE MACINATA	07/09/2021		CC1	1	
2022.TE.28672.1.2	<i>Listeria monocytogenes</i>	SUINO	SALSICIA	30/06/2021		CC9	9	
2022.TE.28671.1.2	<i>Listeria monocytogenes</i>	POLPO	MOLLUSCHI COTTI	25/05/2021		CC8	8	
2022.TE.28670.1.2	<i>Listeria monocytogenes</i>	POLPO	MOLLUSCHI COTTI	25/05/2021		CC8	8	
2022.TE.28669.1.2	<i>Listeria monocytogenes</i>	TONNO	PESCE AFFUMICATO	10/02/2021		CC7	7	
2022.TE.25575.1.2	<i>Listeria monocytogenes</i>	TARTARUGA MARINA	CERVELLO	07/07/2021		CC7	7	
2022.TE.21810.1.2	<i>Listeria monocytogenes</i>		TAMPONE ATTREZZI	21/04/2021		CC177	177	
2022.TE.21809.1.2	<i>Listeria monocytogenes</i>		TAMPONE ATTREZZI	21/04/2021		CC177	177	
2022.TE.21808.1.2	<i>Listeria monocytogenes</i>		TAMPONE ATTREZZI	09/09/2020		CC121	121	
2022.TE.21807.1.2	<i>Listeria monocytogenes</i>		TAMPONE ATTREZZI	09/09/2020		CC121	121	
2022.TE.21806.1.2	<i>Listeria monocytogenes</i>		TAMPONE ATTREZZI	09/09/2020		CC121	121	
2022.TE.21805.1.2	<i>Listeria monocytogenes</i>		TAMPONE ATTREZZI	09/09/2020		CC121	121	
2022.TE.21804.1.2	<i>Listeria monocytogenes</i>		TAMPONE ATTREZZI	18/04/2017		CC121	121	

BioinfoDB - LNR Listeria_monocytogenes. Per visualizzare il Motivo Attivo clicca [qui](#).

5357 Elementi

Specie	Host	Matrice	Clonal Complex ↑	Sequence Type
<input checked="" type="checkbox"/> Listeria monocytogenes	SUINO	SALAME	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	LUPO	CERVELLO	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	SUINO	SALAME	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	SUINO	SALAME	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	SUINO	SALAME	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	SUINO	SALAME	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	SUINO	SALAME	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	SUINO	SALAME	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	SUINO	TAMPONE AMBIENTALE (SPONGE BAGS)	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	SUINO	SALAME	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	SUINO	SALAME	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	BOVINO	CARNE MACINATA	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	BOVINO	CARNE MACINATA	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	MERLUZZO	PREPARAZIONI ALIMENTARI MISTE	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	MERLUZZO	PREPARAZIONI ALIMENTARI MISTE	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	MERLUZZO	PREPARAZIONI ALIMENTARI MISTE	CC1	1
<input checked="" type="checkbox"/> Listeria monocytogenes	UOMO	TUTTI	CC1	1

cgMLST Clustering: GenPat IZSAM

Lancia Accertamenti Singoli

Motivo CSV Campioni

Tipologia Accertamento * Metodo * Accertamento di Input Metodo di Input

DS Esami CSV per Dati dei Risultati [Template di esempio File CSV Dati dei Risultati](#) [WIKI su come inserire i dati nel file CSV dei Risultati](#)

Tipologia Specie Tipologia Schema

— Descrizione Template

In questa pagina si possono lanciare gli accertamenti sui campioni selezionati utilizzando il tool NexFlow.
Trovate una spiegazione su come lanciare le varie analisi nella pagina Wiki al seguente [link](#).

BioInfoDB - LNR Listeria_monocytogenes. Per visualizzare il Motivo Attivo clicca qui.

Schede dati Controlla Lanci Pipeline

Utente	Campioni	Data Richiesta	Ora Richiesta	Data Fine	Ora Fine	Tipologia Analisi	Stato	Tipologia Esito
Gabriella Centonosta	2022.TE.33234.1.8.2022.TE.33234.1...	11/05/2022	10:52:43	11/05/2022	10:53:38	LANCIA ACCERTAMENTI SINGOLI	TERMINATO	Q SUCCESSO

ACCERTAMENTOPIPELINE | METODOINPUT
IMC_clustering | grapefree

Dati di base

Utente: Gabriella Centonosta

Campioni
 2022.TE.33234.1.8.2022.TE.33234.1.1.2022.TE.33234.1.17.2022.TE.28674.1.2.2022.TE.28674.1.2.2022.TE.19548.1.2.2022.TE.19547.1.2.2022.TE.19549.1.2.2022.TE.19548.1.1.2022.TE.33234.1.6.2022.TE.33908.1.5.2022.TE.33234.1.7.2022.TE.33234.1.16.2022.TE.33234.1.1.2022.TE.33234.1.21.2022.TE.33234.1.24.2022.TE.33234.1.22

Ora Richiesta: 10:52:43
Ora Fine: 10:53:38
Timestamp: 20220511_105248005
Stato: TERMINATO

Parametri Input
 @pipeline@format: "lanciassestessianaliconextflow" ; csvCorCedioCampioni: "true" ; listaCampioni: "2022.TE.33234.1.8.2022.TE.33234.1.3.2022.TE.33234.1.3.2022.TE.33234.1.17.2022.TE.28674.1.2.2022.TE.19548.1.2.2022.TE.19547.1.2.2022.TE.19549.1.2.2022.TE.19548.1.2.2022.TE.19549.1.2.2022.TE.19548.1.2.2022.TE.33234.1.6.2022.TE.33908.1.5.2022.TE.33234.1.7.2022.TE.33234.1.16.2022.TE.33234.1.1.2022.TE.33234.1.21.2022.TE.33234.1.22" ; motivo: "na" ; role: "LaboratoriLncs" ; stringaQID: "na" ; tipologiaAccertamento: "IMC_clustering" ; metodo: "grapefree" ; input: "na" ; output: "na" ; reference: "na" ; nomeRisultatoReference: "na" ; nomeRisultatoCampioni: "na" ; tuttoAcc: "false" ; data: "na" ; dataBase: "na" ; tipologia: "na" ; tipo: "na" ;

Tipologia Esito
 Q SUCCESSO

METODO/INPUT
 grapefree

Importa Analisi

Opzioni

Reference Segmentato
 INPUT
 47f_cgMST_chewbbaca

ACCERTAMENTO/PIPELINE
 IMC_clustering

Descrizione Esito
 Caricamento riuscito
 Alberto MGS con GrapeTree - Minimum Spanning Tree in formato newick
 Alberto MGS con GenPAT GIG - Neighbor Joining in formato newick
[Caricamento eseguito](#)
 Metadati
 Mancanti Lanci
 Log di GRASP

Descrizione Esito
 Caricamento riuscito

chewBBACA version: 2.8.4
 Authors: Mickael Silva, Pedro Cerqueira, Rafael Mamede
 Github: <https://github.com/B-UMMI/chewBBACA>
 Wiki: <https://github.com/B-UMMI/chewBBACA/wiki>
 Tutorial: https://github.com/B-UMMI/chewBBACA_tutorial
 Contacts: imm-bioinfo@medicina.ulisboa.pt

```
=====
chewBBACA - ExtractCgMST
=====
Started at: 2022-05-09T11:47:28
```

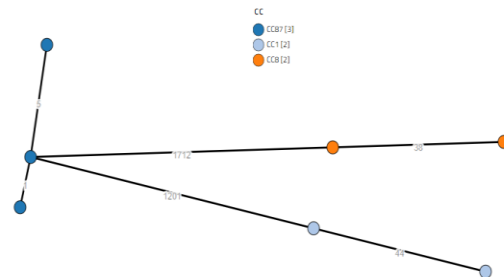
```
Masking missing data...done.
Building presence and absence matrix...done.
Determining genes in the core genome...done.
Determining missing data per genome...done.
```

Core genome composed of 1740/1748 genes.

Finished at: 2022-05-09T11:47:29
 Took 0m 1s.

Plattaforma GenPat, GrapeTree

- Inputs/Outputs
- Tree Layout
- Diagrama di flusso
- Metadati
- Node Style
- Branch Style
- Rendering
- Context Menu



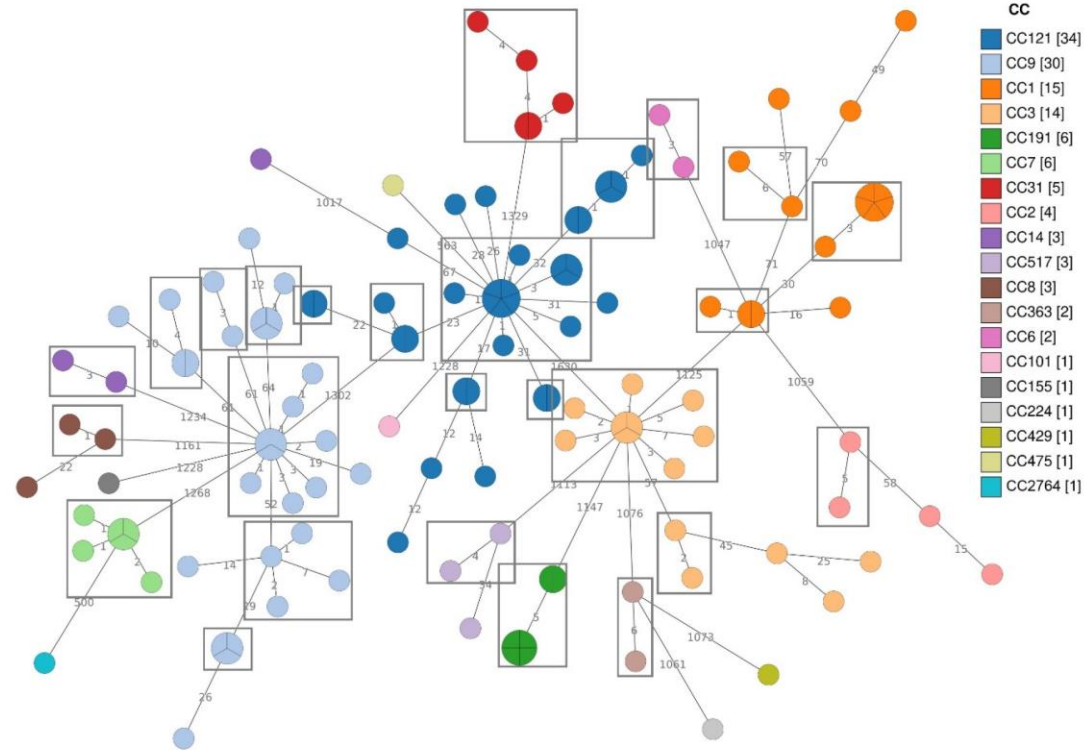


Figure 1. Minimum Spanning Tree (MST) based on the cgMLST profiles of 133 *Lm* strains, coloured according to CCs.

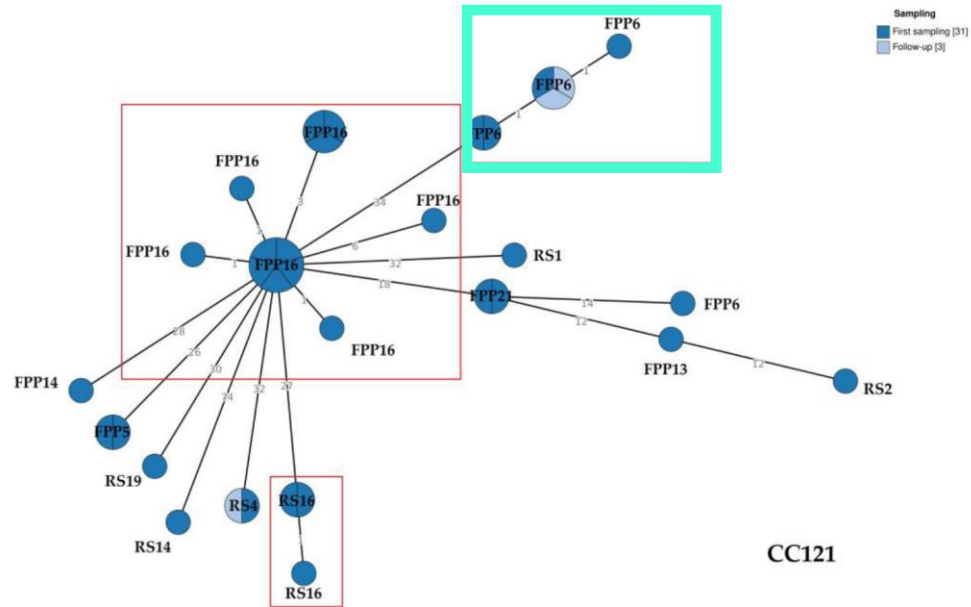


Figure S1. Minimum Spanning Tree (MST) based on the cgMLST profiles of CC121 *Lm* strains coloured according to sampling session; the cgMLST clusters containing more than two strains are highlighted in red.

cgMLST Clustering: GenPat IZSAM

Lancia Accertamenti Singoli

Motivo CSV Campioni

Tipologia Accertamento * **Metodo ***

Accertamento di Input Metodo di Input

DS Esami CSV per Dati dei Risultati [Template di esempio File CSV Dati dei Risultati](#) [WIKI su come inserire i dati nel file CSV dei Risultati](#)

Seleziona Reference da NCBI Seleziona Reference dai Campioni

CSV del Reference [Esempio CSV dei Reference](#) [WIKI su come compilare il CSV dei Reference](#)

Descrizione Template

In questa pagina si possono lanciare gli accertamenti sui campioni selezionati utilizzando il tool NexFlow.
Trovate una spiegazione su come lanciare le varie analisi nella pagina Wiki al seguente [link](#).

Lancia template

SNPs clustering: GenPat IZSAM

Dati di base

Utente *
Gabriella Centorotola

Data Richiesta
13/10/2022

Data Richiesta
13/10/2022

Tipologia Analisi
LANCIA ACCERTAMENTI SINGOLI

Parametri Input
{"tipologiaTemplate":"lanclastepsanalisiconnexflow","csvConCodiciCampioni":"true","listacampioni":"2022.TE.5784.1.2,2022.TE.5780.1.2,2022.TE.5775.1.2","motivo":null,"role":"LaboratorioListeria","stringaDS":null,"tipologiaAccertamento":"9MC_clustering","metodo":"cfsan","kingdom":null,"ospite":null,"referenceString":null,"esameRisultatoReference":"60676949","esameRisultatoCampione":null,"fullOutput":"false","blast_database":null,"tipologiaAnalisiSNP3":"","lunghezzaakmer":"21","taxid":"","coverage":"","identity":"","tipologiaSchema":"","tipologiaSpecie":"","timestamp":"20221013_120504767","accertamento":"1PP_trimming","metodoinput":"trimmomatic","pathFilecsrisultati":"","pathFilecsvreference":"","csvrisultati":"","csvrisultatireference":""}

Tipologia Esito
Successo

METODO/INPUT
cfsan

Importa Analisi

Ospite

Reference Segmentato

Note

Campioni
2022.TE.5784.1.2,2022.TE.5780.1.2,2022.TE.5775.1.2

Ora Richiesta
12:05:04

Ora Fine
12:22:49

Timestamp
20221013_120504767

Stato *
TERMINATO

Accertamento
9MC_clustering

Descrizione Esito
[Cartella Risultati](#)
[Albero ML con GenPAT GIS](#)
[Maximum Likelihood in formato newick](#)
[SNP Matrix](#)
[Warnings](#)
 Cartella Analisi (Per visualizzare il link cliccare prima sull'icona della matita in alto a destra.)
[Cartella Analisi](#)

Reference
AL591824.1 - Listeria monocytogenes

Input
1PP_trimming_trimmomatic



Article

Intensive Environmental Surveillance Plan for *Listeria monocytogenes* in Food Producing Plants and Retail Stores of Central Italy: Prevalence and Genetic Diversity

Gabriella Centorotola ¹, Fabrizia Guidi ^{2,*}, Guglielmo D'Aurizio ³, Romolo Salini ⁴, Marco Di Domenico ⁵, Donatella Ottaviani ², Annalisa Petruzzelli ², Stefano Fisichella ², Anna Duranti ², Franco Tonucci ², Vicdalia Aniela Acciari ¹, Marina Torresi ¹, Francesco Pomilio ¹ and Giuliana Blasi ²

frontiers
in Cellular and Infection Microbiology

BRIEF RESEARCH REPORT
published: 20 October 2021
doi: 10.3389/fcimb.2021.765540



Article

First Report on the Finding of *Listeria monocytogenes* ST121 Strain in a Dolphin Brain

Yann Sévellec ¹, Marina Torresi ², Benjamin Félix ¹, Federica Palma ¹, Gabriella Centorotola ², Stefano Bilei ³, Matteo Senese ³, Giuliana Terracciano ³, Jean-Charles Leblanc ¹, Francesco Pomilio ² and Sophie Roussel ^{1,*}

Hyper-Virulent *Listeria monocytogenes* Strains Associated With Respiratory Infections in Central Italy

Fabrizia Guidi ^{1*}, Alexandra Chiaverini ^{2†}, Antonella Repetto ^{3†}, Cinzia Lorenzetti ^{1†}, Gabriella Centorotola ^{2†}, Viviana Bazzucchi ¹, Barbara Palombo ^{1†}, Antonietta Gattuso ⁴, Francesco Pomilio ^{2†} and Giuliana Blasi ^{1†}





Article

Hypo- and Hyper-Virulent *Listeria monocytogenes* Clones Persisting in Two Different Food Processing Plants of Central Italy

Fabrizia Guidi ^{1,2,*}, Massimiliano Orsini ³, Alexandra Chiaverini ⁴, Marina Torresi ⁴, Patrizia Centorame ⁴, Vicdalia Aniela Acciari ⁴, Romolo Salini ⁵, Barbara Palombo ¹, Giorgio Brandi ², Giulia Amagliani ², Giuditta Fiorella Schiavano ⁶, Francesca Romana Massacci ¹, Stefano Fisichella ¹, Marco Di Domenico ⁷, Massimo Ancora ⁷, Adriano Di Pasquale ⁷, Anna Duranti ¹, Cesare Cammà ⁷, Francesco Pomilio ⁴ and Giuliana Blasi ¹

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ORIGINAL RESEARCH
published: 04 November 2021
doi: 10.3389/fmicb.2021.750065



Phylogenetic Analysis and Genome-Wide Association Study Applied to an Italian *Listeria monocytogenes* Outbreak

Alexandra Chiaverini^{1*}, Fabrizia Guidi², Marina Torresi¹, Vicdalia Aniela Acciari¹, Gabriella Centorotola¹, Alessandra Cornacchia¹, Patrizia Centorame¹, Cristina Marfoggia¹, Giuliana Blasi², Marco Di Domenico³, Giacomo Migliorati¹, Sophie Roussel⁴, Francesco Pomilio¹ and Yann Sevellec⁴

Genetic diversity of *Listeria monocytogenes* strains contaminating food and food producing environment as single based sample in Italy (retrospective study)

Vicdalia Aniela Acciari^{a,b,*1}, Anna Ruolo^{a,b,1}, Marina Torresi^{a,b}, Lucilla Ricci^a, Antonella Pompei^{a,b}, Cristina Marfoggia^{a,b}, Francesca Maria Valente^{a,b}, Gabriella Centorotola^{a,b}, Annamaria Conte^a, Romolo Salini^a, Nicola D'Alterio^{a,b}, Giacomo Migliorati^{a,b}, Francesco Pomilio^{a,b}



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Control/Tracking Number: 2022-A-123-NGS

Activity: Abstract

Current Date/Time: 7/28/2022 4:50:54 AM

Whole Genome Sequencing of *Listeria monocytogenes* from Zambian Meat Foods: a Focus on Hyper-virulent Clones

Author Block: G. Centorotola¹, M. W. Ziba², A. Cornacchia¹, A. Chiaverini¹, M. Torresi¹, D. D'Angelantonio¹, M. Scacchia¹, P. Fandamu³, B. Bowa², P. Mangambwa², G. M. Muuka³, F. Pomilio¹;

¹Istituto Zooprofilattico Sperimentale dell'Abruzzo e del Molise "G. Caporale", Teramo, ITALY, ²Ministry of Fisheries and Livestock, Central Veterinary Research Institute, Lusaka, ZAMBIA, ³Ministry of Fisheries and Livestock, Department of Veterinary Services, Lusaka, ZAMBIA.



THANK YOU!

g.centorotola@izs.it