



Virtual Training 8

Leptospira bigsDB PASTEUR

2024-07-22

Intended Learning Objectives

Leptospira bigsDB PASTEUR basic use

1. How to search in bigsDB Leptospira PASTEUR
2. How to blast a given sequence in bigsDB Leptospira PASTEUR (Ifb1)
3. How to retrieve informations from clustering based on cgMLST

Outline

This session consists of the following elements

1. Introduction to bigsDB Leptospira PASTEUR web interface
2. Introduction to cgMLST Leptospira in bigsDB PASTEUR
3. Practice of lfb1 blast and search for similar isolates

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Audience Q&A Session

① Start presenting to display the audience questions on this slide.

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Leptospira molecular analysis

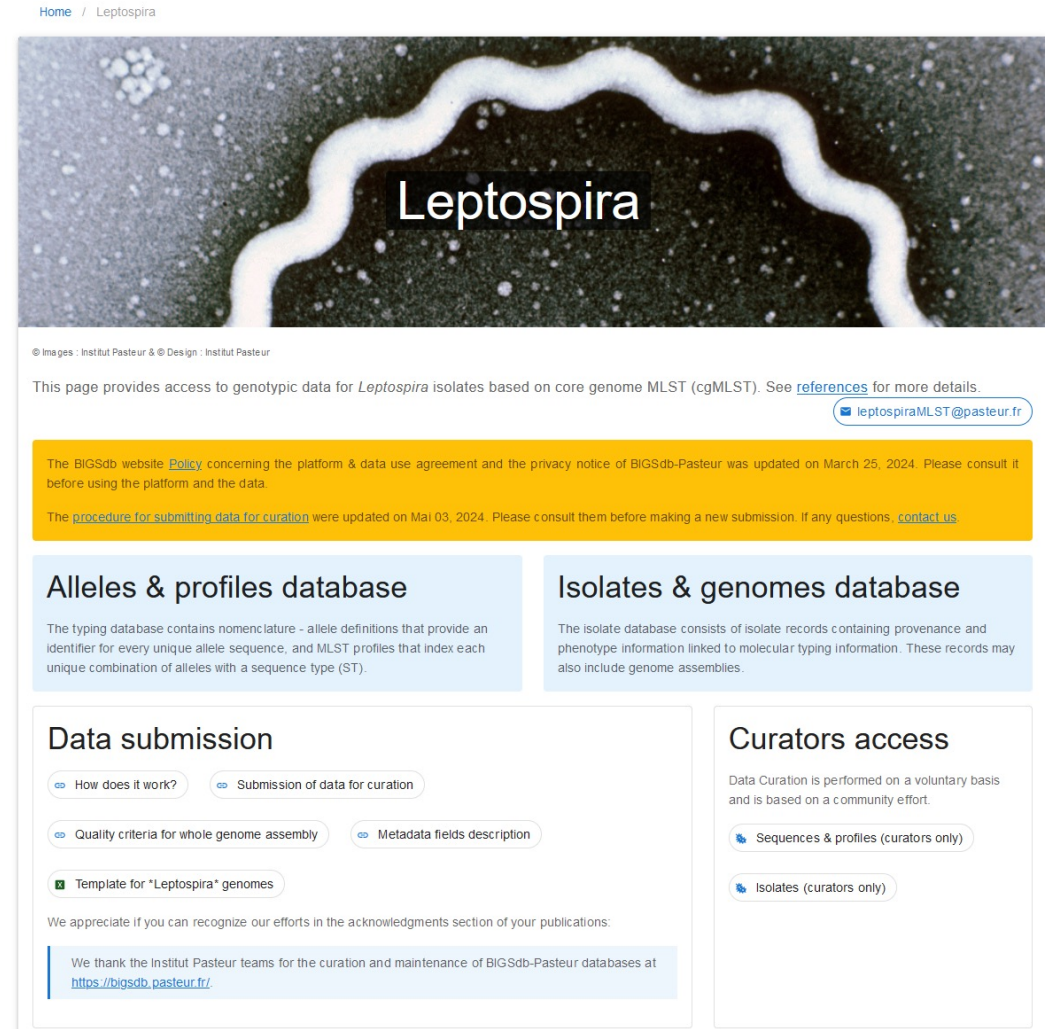
① Start presenting to display the poll results on this slide.

bigsdDB Leptospira web interface


Where : <https://bigsd.db.pasteur.fr/leptospira/>

What : BIGSdb is software designed to store and analyse sequence data for bacterial isolates.

How : it hosts collections of curated, open or private databases of bacterial isolates, genomes and genotypes based on multilocus sequence typing



Home / Leptospira



Leptospira

© Images : Institut Pasteur & © Design : Institut Pasteur

This page provides access to genotypic data for *Leptospira* isolates based on core genome MLST (cgMLST). See [references](#) for more details.

leptospiraMLST@pasteur.fr

The BIGSdb website [Policy](#) concerning the platform & data use agreement and the privacy notice of BIGSdb-Pasteur was updated on March 25, 2024. Please consult it before using the platform and the data.

The [procedure for submitting data for curation](#) were updated on Mai 03, 2024. Please consult them before making a new submission. If any questions, [contact Us](#)

Alleles & profiles database

The typing database contains nomenclature - allele definitions that provide an identifier for every unique allele sequence, and MLST profiles that index each unique combination of alleles with a sequence type (ST).

Isolates & genomes database

The isolate database consists of isolate records containing provenance and phenotype information linked to molecular typing information. These records may also include genome assemblies.

Data submission

- How does it work?
- Submission of data for curation
- Quality criteria for whole genome assembly
- Metadata fields description
- Template for *Leptospira* genomes

We appreciate if you can recognize our efforts in the acknowledgments section of your publications:

We thank the Institut Pasteur teams for the curation and maintenance of BIGSdb-Pasteur databases at <https://bigsd.db.pasteur.fr/>

Curators access

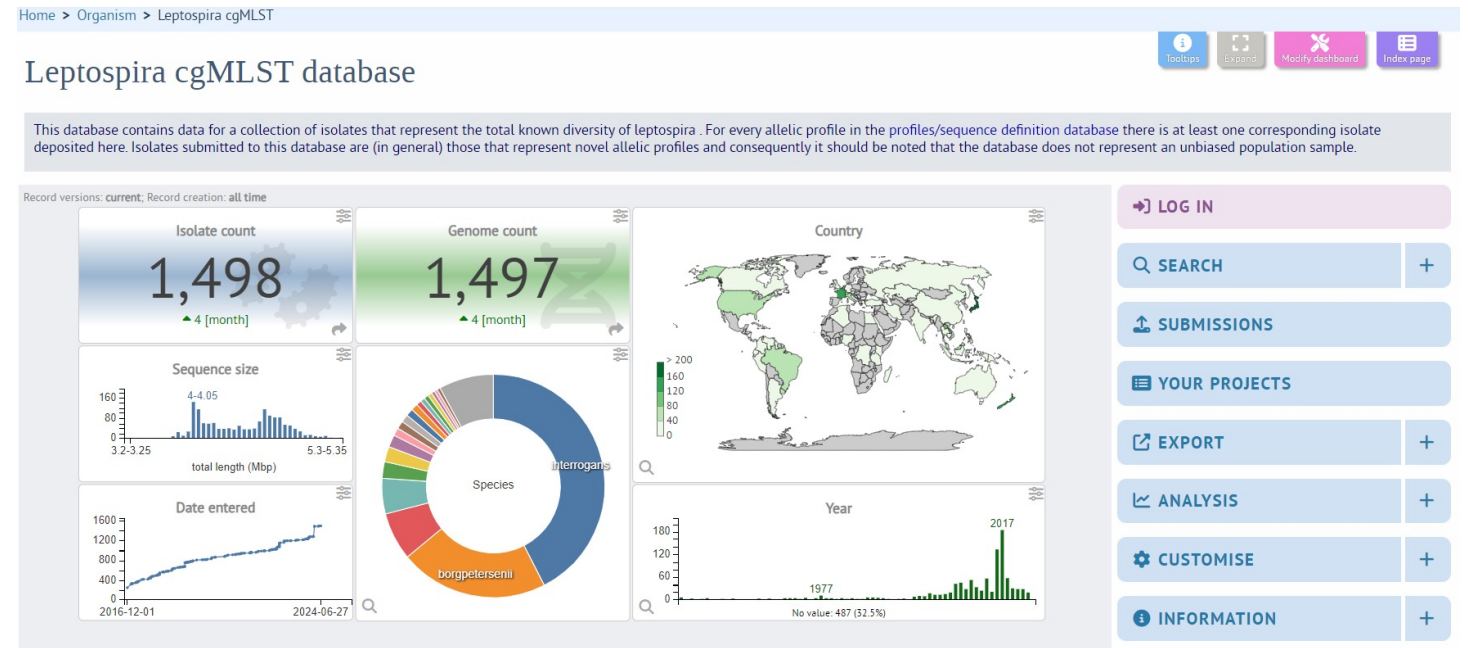
Data Curation is performed on a voluntary basis and is based on a community effort.

- Sequences & profiles (curators only)
- Isolates (curators only)

Leptospira isolate database

To look for available isolates and metadata related to those curated sequence

- Possibility to access to public isolates including related epidemiological data
- Possibility to temporarily submit isolates in private environment (prior publication for example)



Web interface bisgDB Leptospira and search tool



Search for isolate based on « species » and « serogroup » criterium

Study case :

- ✓ species « kirschneri »
- ✓ serogroup « Grippytyphosa »

Output :

List of 29 genomes

Type of metadata available :

World region, host, sample type

Home > Organism > Leptospira cgMLST > Search or browse database

Search or browse database

Enter search criteria or leave blank to browse all records. Modify form parameters to filter or enter a list of values.

Isolate provenance/primary metadata fields: species = kirschneri, serogroup = grippytyphosa

Display/sort options: Order by: id ascending, Display: all records per page

Isolate count: 30, Genome count: 29, Species: kirschneri

30 records returned. Click the hyperlinks for detailed information.

Isolate fields													
id	isolate	aliases	biosample	accession number	clade	species	serogroup	serovar	passage	world region	host	sample type	source lab
78	UT130		SAMN00739395	1049947	P1	kirschneri	Grippytyphosa	Grippytyphosa		South-eastern Asia	human	Unknown	Craig Venter Institute
115	200703020				P1	kirschneri	Grippytyphosa	Grippytyphosa		Western Europe	human	Blood	MaGe
116	200703021				P1	kirschneri	Grippytyphosa	Grippytyphosa		Western Europe	human	Blood	MaGe
117	201402975		SAMEA14271407		P1	kirschneri	Grippytyphosa	Grippytyphosa		Western Europe	human	Blood	MaGe
124	Brem 179		SAMN00739374	GCF_000246295.1	P1	kirschneri	Grippytyphosa	Valbuzzi		Western Europe	horse	Unknown	Craig Venter Institute
142	20021189				P1	kirschneri	Grippytyphosa	Grippytyphosa		Western Europe	human	Blood	MaGe
174	200801774		SAMN02436494		P1	kirschneri	Grippytyphosa	Unknown		Eastern Africa	human	Blood	MaGe
176	201401993				P1	kirschneri	Grippytyphosa	Unknown		Eastern Africa	human	Blood	MaGe
214	RMS2		SAMN02436368	1049946	P1	kirschneri	Grippytyphosa	Grippytyphosa		Northern America	Pig	Unknown	Craig Venter Institute

cgMLST *Leptospira* based on subselection

Several tool to analyse or download informations related to subselection of isolates

Study case :

tool « iTOL » to build cgMLST phylogeny
Subselection k29 kirschneri grippotyphosa isolates

Output

Phylogeny annotated with metadata

Result to come in a few minutes

Analysis tools

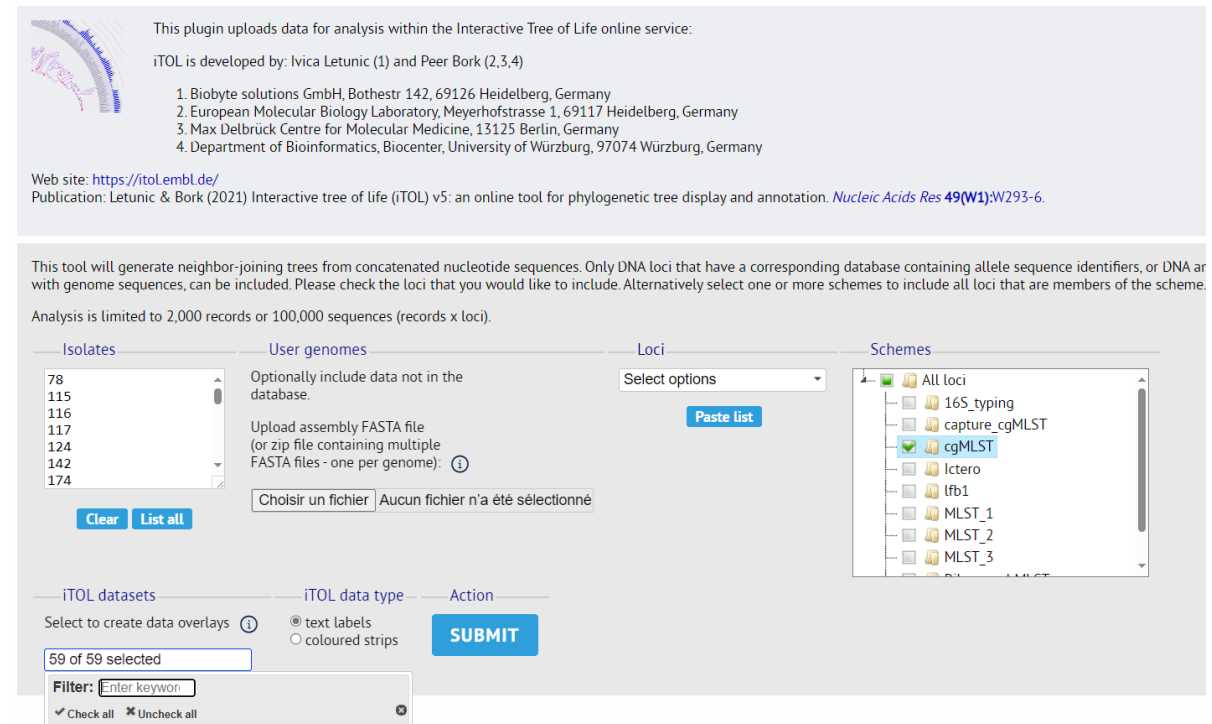
Breakdown: **Fields** **Two Field** **Combinations** **Polymorphic sites** **Publications** **Sequence bin**

Analysis: **BURST** **Codons** **Gene Presence** **Genome Comparator** **BLAST** **rMLST species id** **PCR**

Export: **Dataset** **Contigs** **Sequences**

Third party: **GrapeTree** **iTOL** **ReporTree**

iTOL - Interactive Tree of Life - *Leptospira* cgMLST



This plugin uploads data for analysis within the Interactive Tree of Life online service:
iTOL is developed by: Ivica Letunic (1) and Peer Bork (2,3,4)

1. Biobyte solutions GmbH, Bothestr. 142, 69126 Heidelberg, Germany
2. European Molecular Biology Laboratory, Meyerhofstrasse 1, 69117 Heidelberg, Germany
3. Max Delbrück Centre for Molecular Medicine, 13125 Berlin, Germany
4. Department of Bioinformatics, Biocenter, University of Würzburg, 97074 Würzburg, Germany

Web site: <https://itol.embl.de/>
Publication: Letunic & Bork (2021) Interactive tree of life (iTOL) v5: an online tool for phylogenetic tree display and annotation. *Nucleic Acids Res* 49(W1):W293-6.

This tool will generate neighbor-joining trees from concatenated nucleotide sequences. Only DNA loci that have a corresponding database containing allele sequence identifiers, or DNA ar with genome sequences, can be included. Please check the loci that you would like to include. Alternatively select one or more schemes to include all loci that are members of the scheme. Analysis is limited to 2,000 records or 100,000 sequences (records x loci).

Isolates: 78, 115, 116, 117, 124, 142, 174

User genomes: Optionally include data not in the database. Upload assembly FASTA file (or zip file containing multiple FASTA files - one per genome): Choisir un fichier. Aucun fichier n'a été sélectionné.

Loci: Select options. Paste list

Schemes: All loci, 16S_typing, capture_cgMLST, cgMLST, lctero, lfb1, MLST_1, MLST_2, MLST_3

iTOL datasets: Select to create data overlays (59 of 59 selected). Filter: Enter keyword. Check all, Uncheck all

iTOL data type: text labels (selected), coloured strips

Action: SUBMIT

BLAST tool to analyse lfb1 sequence

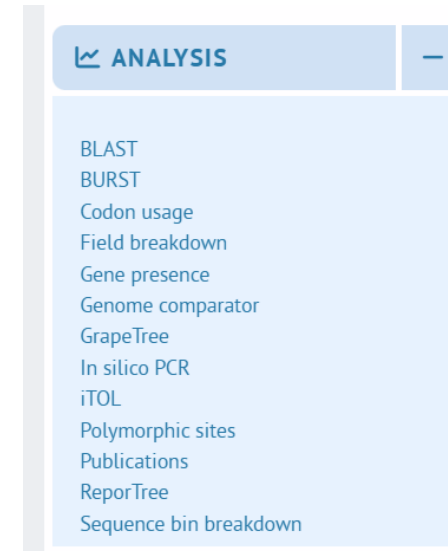
BLAST lfb1 sequence to find related sequence in database

Study case :

lfb1 sequence available in file
« GenEpiBioVirtualTraining8.txt »
Or displayed in chat

Sequence

```
« attcttcctcgcccttctagtagtattcgggtttatctcgaccgcttacgcacagatcgggtca  
aatcaatccttctccatcagcggtaaatacaagggttctggaaccaacccgaacgggt  
cctcttacggcggttagcggttacgatttctgaatctaacggagaataccttttacctgga  
cagtcgccgggtcaaactttcacaggaaccggaacccttgaagggtactactttgacagt  
agattggggagagggtgaaccgggtgatctatgaagtaaaaa »
```



Output BLAST lfb1 sequence

Output :

Table showing three sequence with 100% identity

Direct link to epidemiological data by click on isolate id

Job status viewer

Status

Job id: BIGSdb_3190924_4250059301_15065
 Title: blast-lfb1-kirschneri-grippotyphosa
 Submit time: 2024-07-11 17:26:58
 Status: finished
 Start time: 2024-07-11 17:27:02
 Progress: 100%
 Stop time: 2024-07-11 17:29:46
 Total time: 2 minutes and 43 seconds

Output

Isolate id	Isolate	% identity	Alignment length	Mismatches	Gaps	Seqbin id	Start	End	Orientation	E-value	Bit score
78	UT130	99.281	278	2	0	22217	1931	2208	extract	2.92e-140	493
115	200703020	99.640	278	1	0	30456	77465	77742	extract	2.32e-141	498
116	200703021	99.640	278	1	0	30532	18788	19065	extract	2.33e-141	498
117	201402975	99.640	278	1	0	30598	248097	248374	extract	2.33e-141	498
124	Brem 179	99.640	278	1	0	31826	10255	10532	extract	2.32e-141	498
142	20021189	99.640	278	1	0	37251	132174	132451	extract	2.32e-141	498
174	200801774	99.640	278	1	0	44521	10269	10546	extract	2.48e-141	498
176	201401993	99.640	278	1	0	44829	21782	22059	extract	2.47e-141	498
214	RM52	99.640	278	1	0	48752	34553	34630	extract	2.36e-141	498
305	Brem 127 Duyster	99.640	278	1	0	67447	4590	4867	extract	2.29e-141	498
315	dessain	99.640	278	1	0	68584	10137	10414	extract	2.17e-141	498
383	201600670	99.640	278	1	0	84571	19393	19670	extract	2.46e-141	498
403	201700045	96.763	278	9	0	90417	41293	41570	extract	1.68e-130	462
442	ratnapura1236	96.763	278	9	0	113117	44548	44825	extract	1.68e-130	462
444	ratnapura1439	96.763	278	9	0	354527	71154	71431	extract	1.68e-130	462
451	grippotyphosa1473	99.640	278	1	0	112446	33741	34018	extract	2.31e-141	498
484	kirsch ratnapura1142	96.763	278	9	0	116521	14732	15009	extract	1.70e-130	462
489	kirsch ratnapura1439	96.763	278	9	0	117699	44548	44825	extract	1.69e-130	462
499	LEP1226	99.640	278	1	0	119642	15303	15580	extract	2.32e-141	498
507	kirsch ratnapura1236	96.763	278	9	0	121926	6239	6516	extract	1.69e-130	462
628	11280	98.640	278	1	0	141854	132750	134027	extract	2.34e-141	498
700	201900331;Moskva V	100.000	278	0	0	190637	4580	4857	extract	5.52e-143	502
1038	Ag07_2020	100.000	278	0	0	238872	4580	4857	extract	5.54e-143	502
1094	200201189	99.640	278	1	0	254018	1214	1491	extract	2.30e-141	498
1176	Wumalasesna	96.763	278	9	0	276447	245675	245952	extract	1.70e-130	462
1348	202107008	98.640	278	1	0	280108	4634	4901	extract	2.34e-141	498
1618	Moskva	100.000	278	0	0	360801	121502	121779	extract	5.48e-143	502
1624	200702274	99.640	278	1	0	363407	98183	98460	extract	2.32e-141	498

Files

FASTA FASTA with flanking Table (tab-delimited text)

Table (Excel format) Tar file containing all output files

cgMLST *Leptospira kirschneri* output

Output

iTOL unrooted Phylogeny annotated with metadata, to reroot midpoint in advanced tree

Possibility to check metadata of interest to display close to the tree

Job status viewer

Status


Job id: BIGSdb_251876_2382014523_07518
 Title: kirgri_cgMLST_itol
 Submit time: 2024-07-12 13:29:54
 Status: finished
 Start time: 2024-07-12 13:30:01
 Progress: 100%
 Stop time: 2024-07-12 13:46:54
 Total time: 16 minutes and 52 seconds


Output


The following ids had no sequences for all the selected loci: 485.
 They have been removed from the analysis.


[Launch iTOL](#)

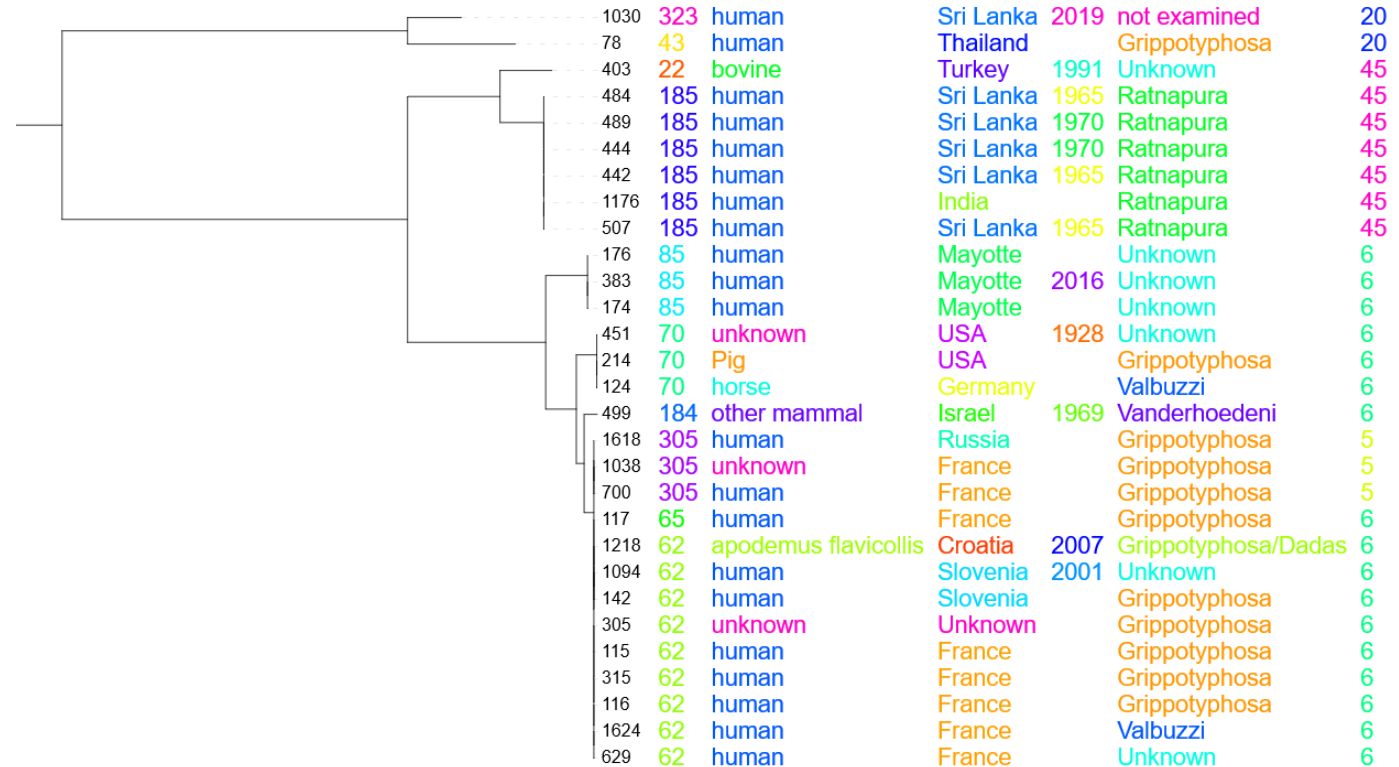
Files

 Concatenated FASTA (16.6 MB)

 NJ tree (Newick format)

 iTOL datasets (Zip format)

 Tar file containing all output files (only files <10MB included - download larger files separately)



bigDB search for species Interrogans

Search for isolate based on « species »

Study case : species « interrogans »

Output : List of 636 genomes

Home > Organism > Leptospira cgMLST > Search or browse database

Search or browse database

Enter search criteria or leave blank to browse all records. Modify form parameters to filter or enter a list of values.

Isolate provenance/primary metadata fields
 species = interrogans

Display/sort options
 Order by: id ascending
 Display: 25 records per page

RESET SEARCH

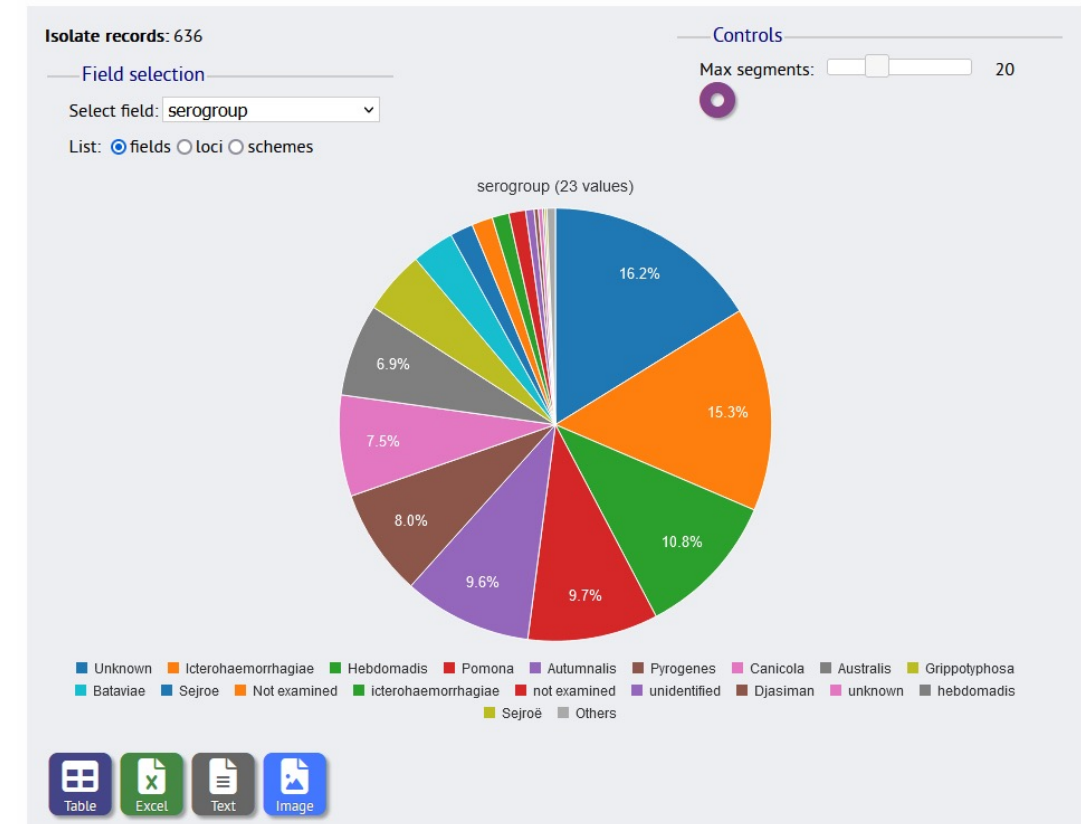
Isolate count: 636
 Genome count: 636
 Species: interrogans

636 records returned (1 - 25 displayed). Click the hyperlinks for detailed information.

Navigation: << < 1 2 3 4 5 6 > >>

Home > Organism > Leptospira cgMLST > Plugins > Field breakdown

Field breakdown of dataset



bigSDB search for species Interrogans

Then make a combined subselection serogroup « Icterohaemorrhagiae » + select host NOT unknown

Search or browse database

Enter search criteria or leave blank to browse all records. Modify form parameters to filter or enter a list of values.

Isolate provenance/primary metadata fields

Combine with: AND

species	starts with	interrogans	+
serogroup	starts with	ict	
host	NOT contain	unknown	

Display/sort options

Order by: id

Display: all records per page

Isolate count

98

▲ 0 [month]

Genome count

98

▲ 0 [month]

Then ANALYSIS : iTOL schemes cgMLST datasets « General »

iTOL - Interactive Tree of Life - Leptospira cgMLST

This plugin uploads data for analysis within the Interactive Tree of Life online service.
iTOL is developed by: Ivica Letunic (1) and Peer Bork (2,3,4)

1. Biobyte solutions GmbH, Bothestr. 142, 69126 Heidelberg, Germany
2. European Molecular Biology Laboratory, Meyerhofstrasse 1, 69117 Heidelberg, Germany
3. Max Delbrück Centre for Molecular Medicine, 13125 Berlin, Germany
4. Department of Bioinformatics, BioCenter, University of Würzburg, 97074 Würzburg, Germany

Web site: <https://itol.embl.de/>
Publication: Letunic & Bork (2021) Interactive tree of life (iTOL) v5: an online tool for phylogenetic tree display and annotation. *Nucleic Acids Res* 49(W1):W293-6.

This tool will generate neighbor-joining trees from concatenated nucleotide sequences. Only DNA loci that have a corresponding database containing allele sequence identifiers, or DNA and peptide loci with genome sequences, can be included. Please check the loci that you would like to include. Alternatively select one or more schemes to include all loci that are members of the scheme.

Analysis is limited to 2,000 records or 100,000 sequences (records x loci).

Isolates

10
26
27
87
97
105
106

Clear List all

User genomes

Optionally include data not in the database.

Upload assembly FASTA file (or zip file containing multiple FASTA files - one per genome):

Parcourir... Aucun fichier sélectionné.

Loci

Select options

Paste List

Schemes

- All loci
- 16S_typing
- capture_cgMLST
- cgMLST
- ictero
- ifb1
- MLST_1
- MLST_2
- MLST_3

iTOL datasets

Select to create data overlays

34 of 59 selected

Filter: Enter keywords

Check all Uncheck all

General

- isolate
- biosample
- accession number
- clade
- species
- serogroup

iTOL data type: Action

text labels coloured strips

SUBMIT

Job status viewer

Home > Organism > Leptospira cgMLST > Job status viewer

Status

Job id: BIGSdb_3130439_0757378360_90855

Title: int_ictero_host

Submit time: 2024-07-15 11:13:01

Status: finished

Start time: 2024-07-15 11:13:02

Progress: 100%

Stop time: 2024-07-15 11:29:04

Total time: 16 minutes and 2 seconds

Output

Launch iTOL

Files

Concatenated FASTA (56.2 MB)

NJ tree (Newick format)

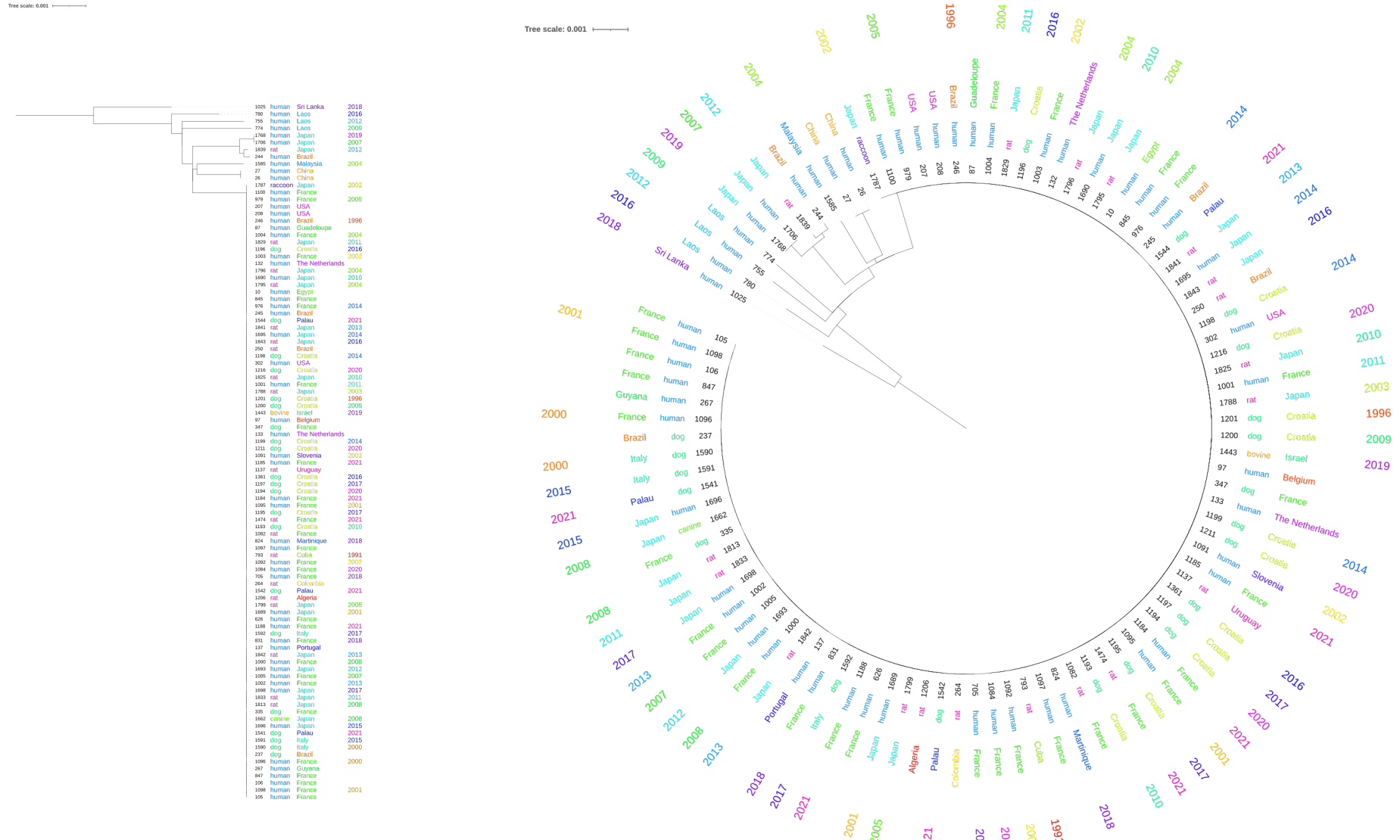
iTOL datasets (Zip format)

Tar file containing all output files (only files <10MB included - download larger files separately)

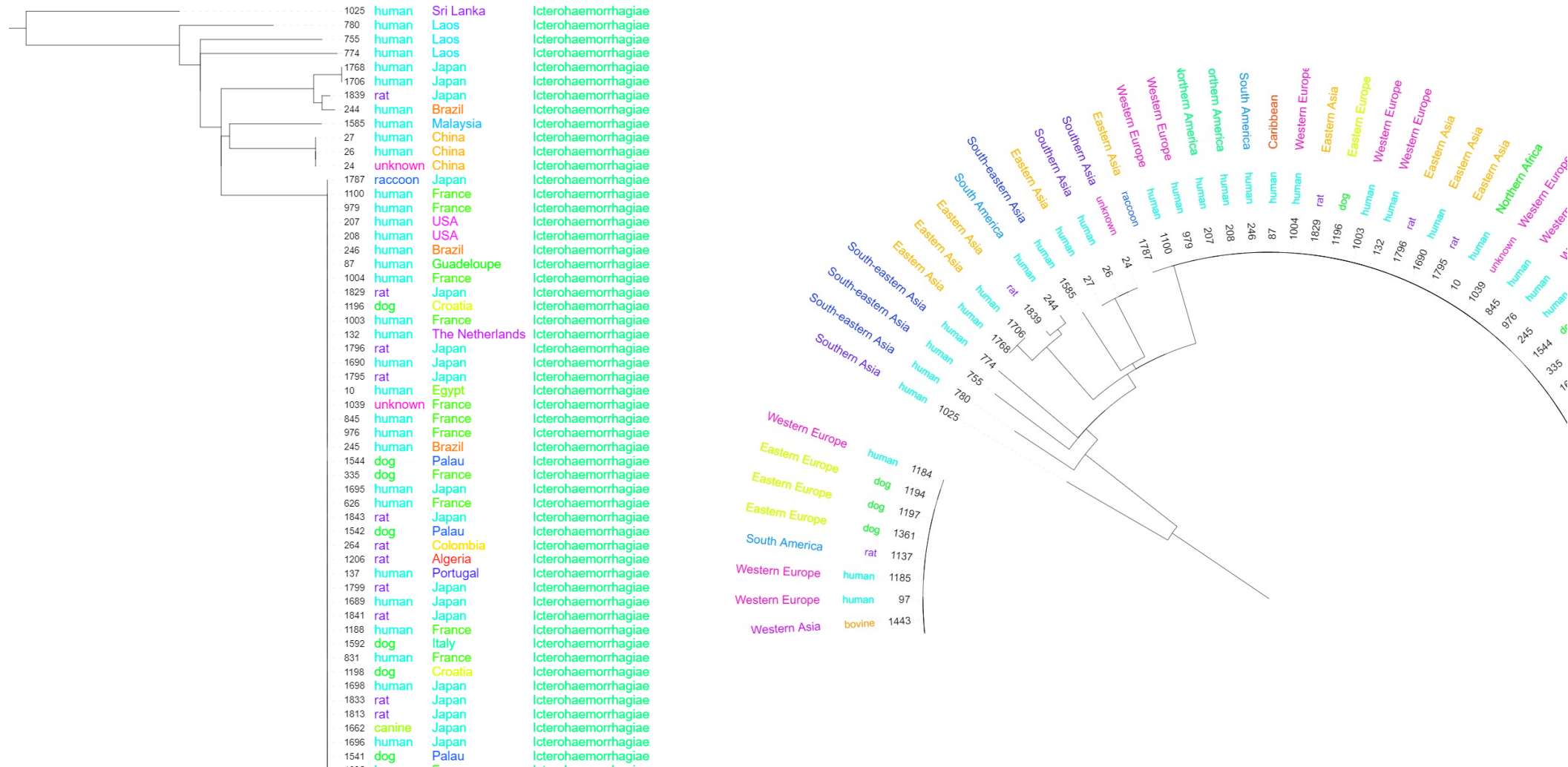
Please note that job results will remain on the server for 2 days.

bigDB search for species Interrogans

iTOL output



bigSDB search for species Interrogans



In summary

Discovery of power of bigsDB interface :

- Looking for existing isolates based on epidemiological data
- From known sequences how to retrieve closes isolates available
example: lfb1 in subselection of Lesptospira kirschneri serogroup Grippotyphosa
- By combining epidemiological criteria search and sequence data build sequence relationship with public data

References



BIGSdb

Jolley, Keith & Maiden, Martin. (2010). BIGSdb: Scalable analysis of bacterial genome variation at the population level. *BMC bioinformatics*. 11. 595. 10.1186/1471-2105-11-595.

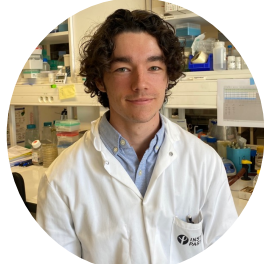
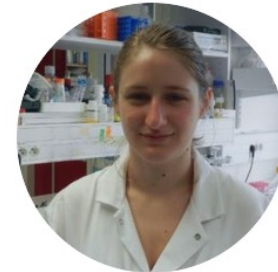
Jolley KA, Bray JE and Maiden MCJ. Open-access bacterial population genomics: BIGSdb software, the PubMLST.org website and their applications [version 1; peer review: 2 approved]. *Wellcome Open Res* 2018, 3:124 (<https://doi.org/10.12688/wellcomeopenres.14826.1>)

lfb1

Garcia-Lopez M, Lorigou C, Soares A, Trombert-Paolantoni S, Harran E, Ayrat F, Picardeau M, Djelouadji Z, Bourhy P. Genetic diversity of *Leptospira* strains circulating in humans and dogs in France in 2019-2021. *Front. Cell. Infect. Microbiol.* Volume 13 – 2023 DOI : 10.3389/fcimb.2023.1236866

cgMLST

Guglielmini J, Bourhy P, Schiettekate O, Zinini F, Brisse S, Picardeau M. Genus-wide *Leptospira* core genome multilocus sequence typing for strain taxonomy and global surveillance. *PLoS Negl Trop Dis.* **2019**; 13(4):e0007374. [PMID 31026256](#)



**Biology of Spirochetes unit
National Reference Center for Leptospirosis
WHO Collaborating Center for Research and Reference on Leptospirosis**

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