DiversiLab® Kit: Bacterial (cat.# 270633)

DiversiLab Library Name: DL Gram-positive Library

Application: Strain-typing for all Gram-positive organisms and characterization of Gram-positive organisms at the genus/species level\*

Reference Methods: Phenotyping, sequencing

Library Contents Overview				
Total Number of Samples represented:	529			
Number of genera represented:	42			
Number of species represented:	161			
Cutoff for species match:	Indistinguishable			

See complete sample list on pages 4-5.

Viewable Report Fields				
Sample ID	Sample ID			
Genus	Genus			
Species	species			
Subspecies	subspecies			
Strain	ATCC number			

## **General Information:**

The Gram-positive library is comprised of samples amplified using the DiversiLab® Bacterial fingerprinting kit. This kit must be used to process samples for comparison to the library and this should be the selected kit inside the DiversiLab software. The culture collection largely used to populate this library is derived from geographically-diverse clinical and industrial sources. On the clinical side, the collection houses isolates with various antimicrobial resistance mechanisms of interest. From the industrial community, isolates are included from agri-food sources as well as from the environment.

Because of the large number of samples contained in the Gram-positive library, the library should be created in a specific way for a quicker report processing speed and analysis. During report creation, the following options should be selected. Figure 1 illustrates how to select these settings in the DiversiLab Software.

- Type Classification, allows use of the Top Match feature
- Viewable Report Fields Sample ID, Genus, Species, fields listed above are optional
- Color-coding Genus (or Species if more specific information is required)
- Collapse (%) 85-90%, allows dendrogram to condense to manageable size

After you have created the report as shown above, report processing may take some time depending on the number of samples that you are comparing due to the large nature of the report. Please be patient. When first looking at the report, it is recommended to:

Note: The Library should be used as a guide.

<sup>\*</sup> For the highest level of discrimination of a particular organism, use of the genus-specific kit is recommended when available. The use of the Bacterial fingerprinting kit is not recommended for *Mycobacteria* or fungi.

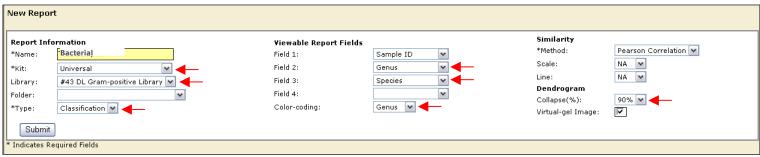


Figure 1. Recommended settings for report creation

1. Check the top match section of the report: if the query sample has a number of the same organism in its match list, there may be a match.

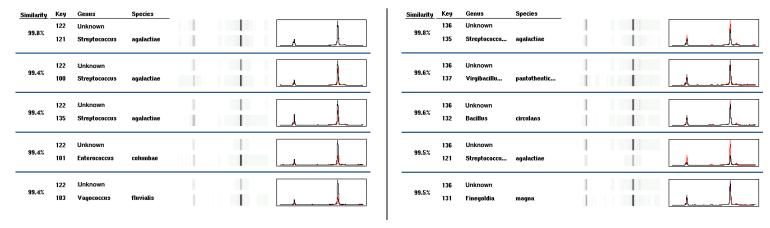


Figure 2. Top match report showing an unknown sample, key 122, with top matches to *S. agalactiae* (left). Top match report showing a different unknown sample, key 136, also with top match to *S. agalactiae*; however, other organisms are also a high match (right). Notice that the % similarity of all matches is high.

Examine graph overlays for indistinguishable fingerprints. It is especially important to view the sample graphs of Gram-positive fingerprints as
they often have one or two large peaks with many smaller peaks. Because of this, multiple genera may cluster together on the dendrogram;
however, smaller peaks will discriminate these samples from each other. Additionally, selection of the Kullback-Leibler calculation method
may aid the clustering of Gram-positive data.

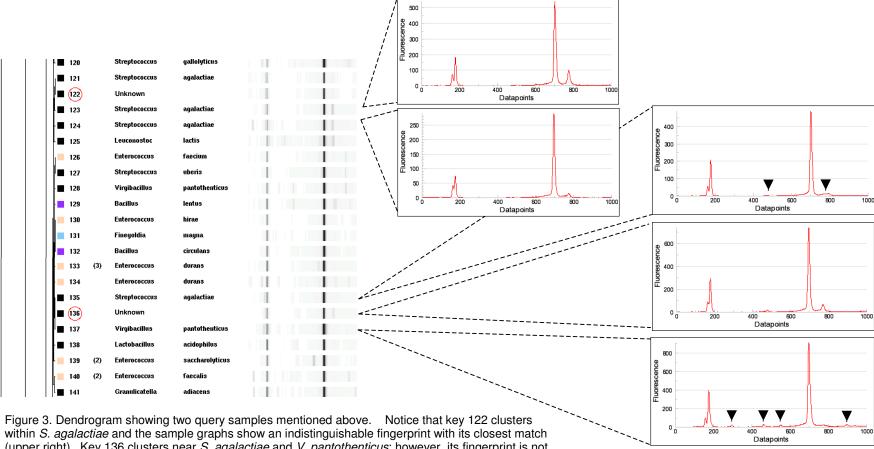


Figure 3. Dendrogram showing two query samples mentioned above. Notice that key 122 clusters within *S. agalactiae* and the sample graphs show an indistinguishable fingerprint with its closest match (upper right). Key 136 clusters near *S. agalactiae* and *V. pantothenticus*; however, its fingerprint is not indistinguishable to either of these top matches (lower right). Therefore, key 122 might be characterized as *S. agalactiae* and key 136 cannot be characterized.

In general, the library shows clustering at the genus/species level. Oftentimes, a specific genus will show a large cluster in which multiple species are contained. However, each species has a distinct fingerprint. It is important to note that dendrograms created in the DiversiLab software show the similarity between sample fingerprints; they do not indicate phylogenetic similarity between samples. Since rep-PCR can discriminate at the sub-species level<sup>1</sup>, there may be multiple fingerprint patterns (strains) for an individual species. Therefore the following are possible:

- 1. There may be multiple clusters for a single species represented in the library.
- 2. The query sample fingerprint may not have a match in the library, even though the particular species is represented.
- 1. Olive, D. M. and P. Bean. 1999. Principles and applications of methods for DNA-based typing of microbial organisms. J. Clin. Microbiol. 37(6): 1661-1669

Genus	species	#	Genus	species	#	Genus	species	#
Actinomyces	israelii	1	Erysipelothrix	rhusiopathiae	2		arlettae	2
	meyeri	2	Eubacterium	limosum	1		aureus	12
	naeslundii	1	Facklamia	hominis	1		auricularis	3
	odontolyticus	2	Finegoldia	magna	1		capitis	7
	urinae	4		bergeri	2		caprae	6
Aerococcus	viridans	1	Gemella	haemolysans	1		carnosus	2
Alloiococcus	otitis	1		sanguinis	1		chromogenes	2
	haemolyticum	5	0 1 "	stearothermophilus	5		cohnii	7
Arcanobacterium	pyogenes	4	Geobacillus	thermodentrificans	1		epidermidis	2
	amyloliquefaciens	1	Globicatella	sulfidifaciens	2		equorum	5
	cereus/thuringiensis	43	6 - 1 - 1 - 1	adiacens	2		gallinarum	4
	circulans	2	Granulicatella	elegans	2		haemolyticus	3
	clausii	2	Helcococcus	kunzii	3	Staphylococcus	hominis	6
	coagulans	2		kristinae	5		hyicus	2
Bacillus	firmus	6	Kocuria	rosea	3		intermedius	6
	lentus	1		varians	1		kloosii	4
	licheniformis	3	Kytococcus	sedentarius	1		lentus	3
	megaterium	4		acidophilus	1		lugdunensis	4
	mycoides	4	Lactobacillus	brevis	1		saccharolyticus	3
	pumilus	9		fermentum	1		saprophyticus	3
	subtilis	9		gasseri	3		schleiferi	5
Bifidobacterium -	adolescentis	1		plantarum	1		sciuri	2
	dentium	1		reuteri	2		simulans	5
Brevibacillus	agri	3		salivarius	1		vitulinus	1
	laterosporus	4		garvieae	1		warneri	3
	baratii	4	Lactococcus	lactis	7	Streptococcus	agalactiae	7
Clostridium	butyricum	1	Leuconostoc	pseudomesenteroides	4		alactolyticus	1
	cadaveris	3	Listeria	grayi	5		canis	5
	clostridioforme	5		innocua	5		cristatus	3
	difficile	2		ivanovii	8		dysgalactiae	6
Clostridium	histolyticum	3	Listeria	monocytogenes	5	Streptococcus	equi	10

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	perfringens	1		seeligeri	5		gallolyticus	3
	sordellii	1		welshimeri	2		hyointestinalis	1
	subterminale	5	Lysinibacillus	fusiformis	1		hyovaginalis	1
	tertium	2	Lysiiiibaciiius	sphaericus	4		infantarius	1
Collinsella	aerofaciens	2	Macrococcus	caseolyticus	1		intermedius	1
	amycolatum	3	Micrococcus	luteus	2		mitis	5
	jeikeium	4		lylae	2		oralis	2
	pseudodiphtheriticum	5	Nocardia	asteroides	2		parasanguinis	2
Corynebacterium	pseudotuberculosis	3		brasiliensis	2		'pasteuri'	1
	striatum	4		cyriacigeorgica	2		phocae	1
	ulcerans	5		otitidiscaviarum	1		pneumoniae	1
	urealyticum	3		pseudobrasiliensis	1		pseudoporcinus	1
Eggerthella	lenta	3		puris	1		pyogenes	2
	avium	6		transvalensis	3		sobrinus	3
	casseliflavus	3	- Paenibacillus	alvei	5		suis	3
	cecorum	3		lentimorbus	1		uberis	3
	columbae	4		polymyxa	6	Vagococcus	fluvialis	2
	durans	8		validus	4	Virgibacillus	pantothenticus	4
Enterococcus	faecalis	6	Parvimonas	micra	4	Viridibacillus	arvi	1
	faecium	2	Pediococcus	acidilactici	3			
	gallinarum	5		pentosaceus	1			
	hirae	3	Peptoniphilus	asaccharolyticus	1			

Note: The Library should be used as a guide.

raffinosus

saccharolyticus

2

4

Propionibacterium

Rhodococcus



granulosum

equi