

The *Bacteroides fragilis* MLST scheme uses internal fragments of seven single-copy core genes associated with housekeeping functions. The fragments are amplified using the following primers pairs and PCR protocol.

Primer	Sequence (5' to 3')	Amplicon size (bp)	MLST fragment size (bp)
<b>GroL - Heat shock protein</b>			
groL_fw	CGGTTATCGGTAAACTGATTGC	607	498
groL_rv	GATTAGTAGCAGCAATCTGAGC		
<b>RpoB - RNA polymerase b-subunit</b>			
rpoB_fw	GCCGATTATCCGGTTGTAG	614	498
rpoB_rv	CGAACTTCGAGTGAATACTCTTCTAC		
<b>DnaJ - Chaperone protein</b>			
dnaJ_fw	GGATAAACGTGCCGCTAC	582	480
dnaJ_rv	C(G/C)CCCATAGAGAGTTGC		
<b>RprX - Histidine kinase</b>			
rprX_fw	TACATCCGTGCGAAATGC	607	498
rprX_rv	CTTCACAATACTCATCTTCGCAG		
<b>PrfA - Release factor</b>			
prfA_fw	CTCAGGA(T/C)GGTAA(G/A)AATGCC	573	471
prfA_rv	CGTCGATATACTTCTGATGTTCC		
<b>FusA- Elongation factor G</b>			
fusA_fw	CTACAACCTCGTCAGGTAAG	588	486
fusA_rv	GGAATGTTACCACCCCTCAC		
<b>RecA - DNA repair recombinase</b>			
recA_fw	GCTGCCATGGACAAGATAG	569	468
recA_rv	ACACCGATTTCACCGC		

Final length of concatenated MLST fragments: 3399bp

#### PCR protocol

All the primers have been designed with a GC content of 40-60% and a melting temperature (tm) of 54-56°C. The following PCR programs are suggestions and should be adjusted according to the specifications of the polymerase used.

Phusion polymerase	Taq polymerase
Initial denaturation: 98°C 30s	Initial denaturation: 95°C 30s
Denaturation: 98°C 10s	Denaturation: 95°C 10s
Annealing: 60°C 20 s	Annealing: 55°C 20 s
Extension: 72°C 30s	Extension: 72°C 30s
Repeat previous 3 steps 30 times	Repeat previous 3 steps 30 times
Final extension: 72°C 5m	Final extension: 72°C 5m