

***Leptospira* pathogenic sps MLST protocol**

Gene	Primer Sequence (5'→3')	Tm (°C)	Size (bp)	Allele size (bp)	
				MLST_2	MLST_3
Adenylate Kinase (adk)	F: GGGCTGGAAGGTACACAA R: ACGCAAGCTCCTTTGAATC	58	531	430	429
	F2-ACATTATCTTCATGGGACCTCC R2- TTACACAAGCTCCCTTTGAAT	54	557		
Isocitrate dehydrogenase (icdA)	F: GGGACGAGATGACCAGGAT R:CTTTTTTGAGATCCGCAGCTTT	58	674	557	555
Lipoprotein L32 (LipL32)	F:ATCTCCGTTGCACTCTTTGC R:ACCATCATCATCATCGTCCA	58	474	450	450
Lipoprotein L41 (LipL41)	F:TAGGAAATTGCGCAGCTACA R:GCATCGAGAGGAATTAACATCA	58	520	493	492
UDP-N-acetylglucosamine pyrophosphorylase (glmU)	F:AGGATAAGGTCGCTGTGGTA R:AGTTTTTTTCCGGAGTTTCT	46	650	444	-
NAD(P) transhydrogenase subunit alpha (pntA)	F:TAGGAAARATGAAACCRGGAAC R:AAGAAGCAAGATCCACAAYTAC	46	621	525	-
Rod shape-determining protein rodA (mreA)	F:GGCTCGCTCTYGACGGAAA R:TCCRTAACATCATAAAMGACAAAGG	46	719	435	-
16S ribosomal RNA (rrs2)	F-CATGCAAGTCAAGCGGAGTA R-AGTTGAGCCCCGAGTTTTTC	58	541	-	450
Pre-protein Translocase SecY subunit (secY)	F-ATGCCGATCATTGCTTC R-CCGTCCCTTAATTTTAGACTTCTTC	58	549	-	501
	R2-CCTTCCTTTAATTTTAGACTTTTTTC	54			

* Same primers were used for sequencing

References:

1. Varni V, Ruybal P, Lauthier JJ, Tomasini N, Brihuega B, et al. (2014) Reassessment of MLST schemes for *Leptospira* spp. typing worldwide. *Infect Genet Evol* 22: 216–222.
2. Boonsilp S, Thaipadungpanit J, Amornchai P, Wuthiekanun V, Bailey MS, et al. (2013) A single multilocus sequence typing (MLST) scheme for seven pathogenic *Leptospira* species. *PLoS Negl Trop Dis* 7: e1954.
3. Ahmed A, Thaipadungpanit J, Boonsilp S, Wuthiekanun V, Nalam K, et al. (2011) Comparison of two multilocus sequence based genotyping schemes for *Leptospira* species. *PLoS Negl Trop Dis* 5: e1374.
4. Nalam K, Ahmed A, Devi SM, Francalacci P, Baig M, et al. (2010) Genetic affinities within a large global collection of pathogenic *Leptospira*: implications for strain identification and molecular epidemiology. *PLoS One* 5: e12637.
5. Ahmed N, Devi SM, Valverde M de los A, Vijayachari P, Machang'u RS, et al. (2006) Multilocus sequence typing method for identification and genotypic classification of pathogenic *Leptospira* species. *Ann Clin Microbiol Antimicrob* 5: 28.