

PROTOCOLS FOR FORENSIC MITOCHONDRIAL DNA ANALYSIS

APPENDIX A OLIGONUCLEOTIDE PRIMER SEQUENCES		
DATE EFFECTIVE 06-20-2016	APPROVED BY MITOCHONDRIAL DNA TECHNICAL LEADER	PAGE 1 OF 2

Appendix A –Oglionucleotide Primer Sequences

Region	Primer	Nucleotide (base) Sequence	Size (no. of bases)
HVI	A1 ¹	5'- CAC CAT TAG CAC CCA AAG CT -3'	20
	A4	5'- CCC CAT GCT TAC AAG CAA GT -3'	20
	B1	5'- GAG GAT GGT GGT CAA GGG AC -3'	20
	B4	5'- TTT GAT GTG GAT TGG GTT T -3'	19
	HVIF ²	5'- CTC CAC CAT TAG CAC CCA A -3'	19
	HVIR	5'- ATT TCA CGG AGG ATG GTG -3'	18
HVII	C1	5'- CTC ACG GGA GCT CTC CAT GC -3'	20
	C2	5'- TTA TTT ATC GCA CCT ACG TTC AAT -3'	24
	D1	5'- CTG TTA AAA GTG CAT ACC GCC A -3'	22
	D2	5'- GGG GTT TGG TGG AAA TTT TTT G -3'	22
	HVIIF	5'- CAC CCT ATT AAC CAC TCA CG -3'	20
	HVIIR	5'- CTG TTA AAA GTG CAT ACC GC -3'	20

1. Nucleotide sequences for primers A1, A4, B1, B4, C1, C2, D1, and D2 are from the FBI Laboratory DNA Analysis Unit II Mitochondrial DNA Analysis Protocol (mtDNA Protocol Manual, DNA Amplification - Rev. 8, Issue Date 02/01/05 for primers A1, B1, C1, C2, D1, and D2; mtDNA Protocol Manual, Cycle Sequencing - Rev. 8, Issue Date 09/10/04 for primers A4 and B4). The primer sequences in the FBI mtDNA Protocol Manual are based on those described in the following:

Wilson MR, DiZinno JA, Polanskey D, Replogle J, Budowle, B. Validation of mitochondrial DNA sequencing for forensic casework analysis. Int J of Leg Med 1995; 108:68-74.

Wilson MR, Polanskey D, Butler J, DiZinno JA, Replogle J, Budowle B. Extraction, PCR amplification, and sequencing of mitochondrial DNA from human hair shafts, BioTechniques 1995; 18(4):662-669.

2. Nucleotide sequences for oligonucleotide primers HVIF, HVIR, HVIIF, HVIIR are from the product insert for the LINEAR ARRAY Mitochondrial DNA HVI/HVII Region-Sequence Typing Kit that is

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available from Roche Applied Sciences (Cat. No. 03-527-867-001; product information is available at www.roche-applied-science.com). The primer sequences in the typing kit are based on those described in:

Gabriel MN, Calloway CD, Reynolds RL, Primorac D. Identification of human remains by immobilized sequence-specific oligonucleotide probe analysis of mtDNA hypervariable regions I and II. Croat Med J 2003; 44:293-298.

Kline MC, Vallone PM, Redman JW, Duewer DL, Calloway CD, Butler JM. Mitochondrial DNA typing screens with control region and coding region SNPs. J Forensic Sci 2005; 50:377-385.