



# National Institute of Standards & Technology

## Certificate of Analysis

### Standard Reference Material<sup>®</sup> 2392

#### Mitochondrial DNA Sequencing (Human)

Standard Reference Material (SRM) 2392 is intended to provide quality control when performing the polymerase chain reaction (PCR) and sequencing of human mitochondrial DNA (mtDNA) for forensic identifications, medical diagnosis, or mutation detection. It may also be used as a control when amplifying (PCR) and sequencing any DNA. This SRM can also be used for quality assurance when assigning values to in-house control materials. It is certified for the sequences of the entire human mtDNA (16 569 base pairs) from two lymphoblastoid cell culture lines (CHR and GM09947A) from apparently normal individuals, plus the cloned HV1 region of CHR containing a C-stretch which is difficult to sequence. The SRM is packaged in a single box containing three components: (1) extracted DNA from cell culture line CHR (tube contains 60  $\mu\text{L}$  of DNA at a concentration of 1  $\text{ng}/\mu\text{L}$ ); (2) extracted DNA from cell culture line GM09947A (tube contains 60  $\mu\text{L}$  of DNA at a concentration of 1  $\text{ng}/\mu\text{L}$ ); and (3) cloned DNA from the CHR HV1 region containing the C-stretch (tube contains 10  $\mu\text{L}$  of DNA at a concentration of 100  $\text{ng}/\mu\text{L}$ ).

This SRM is composed of well-characterized extracted human DNA from CHR and GM09947A and cloned DNA from the HV1 region of CHR. Table 1 contains the certified sequence information of two entire mtDNA templates (CHR and GM09947A). Table 2 contains the reference sequences of 58 unique primer sets which were designed to amplify any portion or the entire human mtDNA. The sequence information of a third DNA template (GM03798) that was amplified and sequenced in its entirety three to four times at NIST is provided in the attached paper [1]. Although the extracted DNA from GM03798 is not provided, the cell culture line can be obtained from NIGMS Human Genetic Mutant Cell Repository, Coriell Institute for Medical Research, Camden, NJ.

**Expiration of Certification:** The certification of this SRM is valid until **31 May 2003** provided the SRM is handled and stored in accordance with the instructions given in this certificate. This certification is nullified if the SRM is damaged, contaminated, or modified.

**Maintenance of SRM Certification:** NIST will monitor this SRM over the period of its certification. If substantive technical changes occur that affect the certification before the expiration of this certificate, NIST will notify the purchaser. Return of the attached registration card will facilitate notification.

The analytical determination and technical measurements for the certification of this SRM were performed by B.C. Levin, H. Cheng, L.A. Tully, M.P. Jones, and D.J. Reeder of the NIST DNA Technologies Group, Biotechnology Division.

The overall direction and coordination of the technical measurements leading to the certification were performed by B.C. Levin and D.J. Reeder of the NIST DNA Technologies Group, Biotechnology Division.

The support aspects involved in the preparation, certification, and issuance of this SRM were coordinated through the NIST Standard Reference Materials Program by J.C. Colbert.

Gary L. Gilliland, Chief  
Biotechnology Division

Gaithersburg, MD 20899  
Certificate Issue Date: 29 December 1999

Thomas E. Gills, Director  
Office of Measurement Services

## NOTICE AND WARNINGS TO USER

**Warning:** SRM 2392 IS A HUMAN SOURCE MATERIAL. SINCE THERE IS NO CONSENSUS ON THE INFECTIOUS STATUS OF EXTRACTED DNA, HANDLE PRODUCT AS A BIOHAZARDOUS MATERIAL CAPABLE OF TRANSMITTING INFECTIOUS DISEASE.

**Storage:** Store frozen at a temperature of -20 °C. Do not use a self-defrosting freezer because periodic cycling of temperatures may cause shortened shelf life of this SRM.

**Use:** It is recommended that once thawed, each SRM component should be used in its entirety. Repeated freezing and thawing is **NOT** recommended as this might shorten the shelf life of the SRM. If it is necessary to perform repeated analyses, thaw the SRM and divide the tube contents into aliquots that will be kept frozen until use. Thawing can be conducted at refrigerator temperatures, room temperature, or at 37 °C. Once thawed, the sample should be processed without delay.

## SOURCE AND ANALYSIS

**Source of Material:** CHR DNA, both extracted and cloned, was prepared in the NIST DNA Technologies Group, Biotechnology Division. DNA for GM09947A was prepared by Life Technologies, Inc.<sup>1</sup>, Gaithersburg, MD.

**NIST Analysis:** NIST extracted DNA from the CHR cell culture, PCR was used to amplify both the CHR DNA and GM09947A DNA with all 58 primer sets multiple times, and sequenced the PCR products with a Perkin-Elmer Applied Biosystems, Inc. (ABI) 373 automated sequencer or an ABI 310 sequencer. The cloned DNA was prepared at NIST as described in the attached paper. The sequence of the CHR clone and of representative PCR products of the final CHR and GM09947A DNA included in SRM 2392 was reanalyzed to ensure sequence accuracy.

**Interlaboratory Analysis:** An interlaboratory evaluation of the amplification, sequencing, and data analysis of the CHR template was conducted by four laboratories, including NIST. These laboratories were: The Bode Technology Group, Inc., Sterling, VA; IIT Research Institute, Virginia Technology Center, Newington, VA; and Lark Technologies, Inc., Houston, TX. Description of the interlaboratory analyses is described in the attached paper.

**Description of Components:** Three components are included in each unit; all components must be stored at -20 °C.

- # 1 Extracted DNA from cell culture line CHR (tube contains 60 µL of DNA at a concentration of 1 ng/µL)
- # 2 Extracted DNA from cell culture line GM09947A (tube contains 60 µL of DNA at a concentration of 1 ng/µL)
- # 3 Cloned DNA from the CHR HV1 region containing the C-stretch (tube contains 10 µL of DNA at a concentration of 100 ng/µL)

**NOTE:** DNA concentrations given are nominal values and are not intended for use as concentration standards.

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<sup>1</sup> Certain commercial equipment, instruments, materials, or companies are identified in this paper to specify the experimental procedure. Such identification does not imply recommendation or endorsement by NIST, nor does it imply that the materials or equipment identified are the best available for this purpose.

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Table 1. Certified Human mtDNA Sequence Differences from the Anderson [2] Sequence Found in Two Templates at NIST

Primer Set	Amplified Region <sup>a</sup>	Length of Amplified Region	Comparison with Anderson			Amino Acid Change	
			Anderson #	bp	Template CHR		
1 (HV2)	15 - 484	470	73	A	Start 39 G	Start 39	
			93	A	-	G	
			195	T	C	C	
			204	T	C	C	
			207	G	A	-	
			214	A	-	G	
			263	A	G	G	
			309.1		C(ins)	C(ins)	
			309.2		-	C(ins)	
			315.1		C(ins)	C(ins)	End 473
2	361 - 921	561	709	G	Start 429 A	Start 421	
			750	A	G	G	End 846
3	756 - 1425	670	NONE		Start 778 End 1197		
4	873 - 1425	553	NONE		Start 928 End 1377		
5	1234 - 1769	536	1438 1719	A G	Start 1279 G A End 1738	Start 1275 G - End 1741	

Table 1. Continued

6	1587 - 2216	630	1719 <sup>d</sup>	G	Start 1632 A End 2106	Start 1632 - End 2106	
7	1657 - 2216	560	1719 <sup>d</sup>	G	Start 1691 A End 2170	Start 1686 - End 2173	
8	1993 - 2216	224	NONE		Start 2036 End 2213	Start 2018 End 2217	
9	2105 - 2660	556	NONE		Start 2157 End 2636	Start 2150 End 2586	
10	2417 - 3006	590	2706	A	Start 2465 G End 2920	Start 2458 - End 2956	
11	2834 - 3557	724	3106/3107	C	Start 2861 Del End 3350	Start 2869 Del End 3373	
12	2972 - 3557	586	3106/3107 <sup>d</sup> 3423	C G	Start 2999 Del E End 3422	Start 2999 Del T End 3460	Silent
13	3234 - 3557	324	3423 <sup>d</sup>	G	Start 3265 T End 3548	Start 3258 T End 3545	Silent <sup>d</sup>
14	3441 - 3940	500	NONE		Start 3487 End 3916	Start 3491 End 3920	
15	3635 - 4162	528	NONE		Start 3667 End 4126	Start 3662 End 4061	

Table 1. Continued

6	1587 - 2216	630	1719 <sup>d</sup>	G	Start 1632 A End 2106	Start 1632 - End 2106
7	1657 - 2216	560	1719 <sup>d</sup>	G	Start 1691 A End 2170	Start 1686 - End 2173
8	1993 - 2216	224	NONE		Start 2036 End 2213	Start 2018 End 2217
9	2105 - 2660	556	NONE		Start 2157 End 2636	Start 2150 End 2586
10	2417 - 3006	590	2706	A	Start 2465 G End 2920	Start 2458 - End 2956
11	2834 - 3557	724	3106/3107	C	Start 2861 Del End 3350	Start 2869 Del End 3373
12	2972 - 3557	586	3106/3107 <sup>d</sup> 3423	C G	Start 2999 Del E End 3422	Start 2999 Del T End 3460
13	3234 - 3557	324	3423 <sup>d</sup>	G	Start 3265 T End 3548	Start 3258 T End 3545
14	3441 - 3940	500	NONE		Start 3487 End 3916	Start 3491 End 3920
15	3635 - 4162	528	NONE		Start 3667 End 4126	Start 3662 End 4061

Table 1. Continued

16	3931 - 4728	798	4135	T	Start 3964 - End 4399	Start 3968 C End 4427	Try→His
17	4183 - 4728	546	NONE		Start 4208 End 4657	Start 4249 End 4657	
18	4392 - 4982	591	4769	A	Start 4449 G End 4860	Start 4453 G End 4935	Silent
19	4447 - 4982	536	4769 <sup>d</sup>	A	Start 4492 G End 4958	Start 4492 G End 4921	Silent <sup>d</sup>
20	4797 - 5553	757	4985 5186	G A	Start 4838 A G End 5327	Start 4845 A - End 5324	Silent Silent
21	4976 - 5553	578	5186 <sup>d</sup>	A	Start 5000 G End 5516	Start 5007 - End 5521	Silent <sup>d</sup>
22	5318 - 5882	565	NONE		Start 5361 End 5754	Start 5360 End 5758	
23	5700 - 6262	563	NONE		Start 5741 End 6149	Start 5744 End 6163	
24	5999 - 6526	528	6221 6371	T C	Start 6043 C T End 6442	Start 6058 - - End 6503	Silent Silent
25	6242 - 6526	285	6371 <sup>d</sup>	C	Start 6271 T End 6520	Start 6302 - End 6520	Silent <sup>d</sup>

Table 1. Continued

26	6426 - 7030	605	6791 6849*	A A	Start 6451 G G(0.3A) <sup>h*</sup> End 6916	Start 6474 - - End 6930	Silent Thr→Ala*
27	6744 - 7255	512	6849 <sup>d*</sup> 7028	A C	Start 6775 G(0.3A) <sup>h*</sup> T End 7215	Start 6782 - - End 7221	Thr→Ala <sup>d*</sup> Silent
28	7075 - 7792	718	NONE		Start 7123 End 7602	Start 7123 End 7601	
29	7215 - 7792	578	7645	T	Start 7263 - End 7722	Start 7280 C End 7769	Silent
30	7645 - 8215	571	7861	T	Start 7671 - End 8149	Start 7666 C End 8155	Silent
31	7901 - 8311	411	NONE		Start 7960 End 8289	Start 7959 End 8288	
32	8164 - 8669	506	8448 8503	T T	Start 8211 - C End 8646	Start 8212 C - End 8641	Met→Thr Silent
33	8539 - 9059	521	8860	A	Start 8581 G End 9019	Start 8582 G End 8999	Thr→Ala
34	8903 - 9403	501	9315	T	Start 8947 - End 9380	Start 8944 C End 9381	Phe→Leu



Table 1. Continued

26	6426 - 7030	605	6791 6849*	A A	Start 6451 G G(0.3A) <sup>h</sup> * End 6916	Start 6474 - - End 6930	Silent Thr→Ala*
27	6744 - 7255	512	6849 <sup>d</sup> * 7028	A C	Start 6775 G(0.3A) <sup>h</sup> * T End 7215	Start 6782 - - End 7221	Thr→Ala <sup>d</sup> * Silent
28	7075 - 7792	718	NONE		Start 7123 End 7602	Start 7123 End 7601	
29	7215 - 7792	578	7645	T	Start 7263 - End 7722	Start 7280 C End 7769	Silent
30	7645 - 8215	571	7861	T	Start 7671 - End 8149	Start 7666 C End 8155	Silent
31	7901 - 8311	411	NONE		Start 7960 End 8289	Start 7959 End 8288	
32	8164 - 8669	506	8448 8503	T T	Start 8211 - C End 8646	Start 8212 C - End 8641	Met→Thr Silent
33	8539 - 9059	521	8860	A	Start 8581 G End 9019	Start 8582 G End 8999	Thr→Ala
34	8903 - 9403	501	9315	T	Start 8947 - End 9380	Start 8944 C End 9381	Phe→Leu

Table 1. Continued

35	9309 - 9848	540	9559	G	Start 9334 C End 9823	Start 9333 C End 9827	Arg→Pro
36	9449 - 9995	547	9559 <sup>d</sup>	G	Start 9476 C End 9964	Start 9485 C End 9940	Arg→Pro <sup>d</sup>
37	9754 - 10275	522	NONE		Start 9777 End 10225	Start 9781 End 10251	
38	10127 - 10556	430	NONE		Start 10168 End 10534	Start 10166 End 10536	
39	10386 - 11166	781	NONE		Start 10410 End 10899	Start 10416 End 10916	
40	10704 - 11267	564	NONE		Start 10734 End 11223	Start 10742 End 11197	
41	11001 - 11600	600	11335	T	Start 11026 C End 11461	Start 11040 C End 11517	Silent
42	11403 - 11927	525	11719	G	Start 11428 A End 11795	Start 11432 - End 11853	Silent
43	11760 - 12189	430	11878	T	Start 11784 C End 12159	Start 11802 - End 12164	Silent
44	11901 - 12876	976	NONE		Start 11926 End 12404	Start 11926 End 12443	
45	12357 - 12876	520	12612 12705	A C	Start 12404 G T End 12769	Start 12391 - - End 12849	Silent Silent

Table 1. Continued

46	12601 - 13123	523	12705 <sup>d</sup>	C	Start 12627 T End 13102	Start 12645 - End 13045	Silent <sup>d</sup>
47	12793 - 13343	551	NONE		Start 12817 End 13295	Start 12807 End 13307	
48	13188 - 13611	424	13572	T	Start 13238 - End 13587	Start 13238 C End 13593	Silent
49	13518 - 13935	418	13572 <sup>d</sup> 13702 13708 13759	T G G G	Start 13541 - C C A - End 13910	Start 13541 C C - A End 13921	Silent <sup>d</sup> Gly→Arg Ala→Thr Ala→Thr
50	13715 - 14118	404	13966	A	Start 13775 G End 14094	Start 13760 - End 14110	Thr→Ala
51	13899 - 14388	490	13966 <sup>d</sup> 14199 14272 14365	A G G G	Start 13926 G T C C End 14369	Start 13927 - T C C End 14374	Thr→Ala <sup>d</sup> Pro→Thr Phe→Leu Silent
52	14189 - 14926	738	14272 <sup>d</sup> 14365 <sup>d</sup> 14368 14470 14766	G G G T T	Start 14216 C C C C E End 14699	Start 14216 C C C C - C End 14806	Phe→Leu <sup>d</sup> Silent <sup>d</sup> Phe→Leu Silent Ile→Thr

Table 1. Continued

46	12601 - 13123	523	12705 <sup>d</sup>	C	Start 12627 T End 13102	Start 12645 - End 13045	Silent <sup>d</sup>
47	12793 - 13343	551	NONE		Start 12817 End 13295	Start 12807 End 13307	
48	13188 - 13611	424	13572	T	Start 13238 - End 13587	Start 13238 C End 13593	Silent
49	13518 - 13935	418	13572 <sup>d</sup> 13702 13708 13759	T G G G	Start 13541 - C A - End 13910	Start 13541 C C - A End 13921	Silent <sup>d</sup> Gly→Arg Ala→Thr Ala→Thr
50	13715 - 14118	404	13966	A	Start 13775 G End 14094	Start 13760 - End 14110	Thr→Ala
51	13899 - 14388	490	13966 <sup>d</sup> 14199 14272 14365	A G G G	Start 13926 G T C C End 14369	Start 13927 - T C C C End 14374	Thr→Ala <sup>d</sup> Pro→Thr Phe→Leu Silent
52	14189 - 14926	738	14272 <sup>d</sup> 14365 <sup>d</sup> 14368 14470 14766	G G G T T	Start 14216 C C C C E End 14699	Start 14216 C C C - C End 14806	Phe→Leu <sup>d</sup> Silent <sup>d</sup> Phe→Leu Silent Ile→Thr

Table 1. Continued

53	14470 - 14996	527	14766 <sup>d</sup>	T	Start 14502 - End 14957	Start 14513 C End 14972	Ile→Ihr <sup>d</sup>
54	14909 - 15396	488	15326	A	Start 14941 G End 15380	Start 14933 G End 15373	Thr→Ala
55	15260 - 15774	515	15326 <sup>d</sup>	A	Start 15305 G End 15754	Start 15293 G End 15950	Thr→Ala <sup>d</sup>
56	15574 - 16084	511	NONE		Start 15637 End 16056	Start 15599 End 16058	
57 (HV1)	15971 - 16451	481	16183 16189 16311	A T T	Start 16014 C C E End 16193	Start 16011 - - C End 16430	
58	16097 - 336	809	16183 <sup>d</sup> 16189 <sup>d</sup> 16311 <sup>d</sup> 16519	A T T T	Start 16125 C C E E End 16193	Start 16130 - - C C End 59	
-21M13 <sup>c</sup> cloned DNA	16133 - 40	477	16183 <sup>d</sup> 16189 <sup>d</sup> 16193.1 16223 16278 16519 <sup>d</sup>	A T C C T	Start 16131 C C C(ins) T T C End 40	ND	

Table 1. Continued

<sup>a</sup>	Numbers correspond to Anderson sequence [2].
B	Base pair change came before the readable sequence.
E	Base pair change came after the readable sequence.
-	Base pair same as in Anderson sequence.
h*	Possible heteroplasmic site.
*	This heteroplasmy seen in the first CHR cell culture line was not seen with the second CHR cell culture line. It is the second CHR cell culture line that is supplied in NIST SRM 2392.
c	This primer is used for sequencing the cloned DNA of the HIV region.
d	Change also seen in previous primer set.
Start	Start of readable sequence.
End	End of readable sequence.
CHR cells	Sequence based on two amplifications and cycle sequencing procedures in first cell culture line and at least one amplification and cycle sequencing procedure with the second cell culture line.
GM09947A cells	Sequence based on two amplifications and cycle sequencing procedures.
Ins	Insertion
Del	Deletion
ND	Not done

Table 1. Continued

a	Numbers correspond to Anderson sequence [2].
B	Base pair change came before the readable sequence.
E	Base pair change came after the readable sequence.
-	Base pair same as in Anderson sequence.
h*	Possible heteroplasmic site.
*	This heteroplasmy seen in the first CHR cell culture line was not seen with the second CHR cell culture line that is supplied in NIST SRM 2392.
c	This primer is used for sequencing the cloned DNA of the HV1 region.
d	Change also seen in previous primer set.
Start	Start of readable sequence.
End	End of readable sequence.
CHR cells	Sequence based on two amplifications and cycle sequencing procedures in first cell culture line and at least one amplification and cycle sequencing procedure with the second cell culture line.
GM09947A cells	Sequence based on two amplifications and cycle sequencing procedures.
Ins	Insertion
Del	Deletion
ND	Not done

Table 2. Reference Sequences for Primer Sets Used for PCR Amplification of Human mtDNA

PRIMER SET NUMBER		PRIMER SEQUENCE
1 (HV2)	F15 R484	CACCCTATTAACCACTCACG TGAGATTAGTAGTATGGGAG
2	F361 R921	ACAAAGAACCCTAACACCAGC ACTTGGGTTAATCGTGTGACC
3	F756 R1425	CATCAAGCACGCAGCAATG AATCCACCTTCGACCCTTAAG
4	F873 R1425	GGTTGGTCAATTCGTGCCAG AATCCACCTTCGACCCTTAAG
5	F1234 R1769	CTCACCACTCTTGCTCAGC GCCAGGTTTCAATTTCTATCG
6	F1587 R2216	TGCACTTGGACGAACCAGAG TGTTGAGCTTGAACGCTTTC
7	F1657 R2216	CTTGACCGCTCTGAGCTAAAC TGTTGAGCTTGAACGCTTTC
8	F1993 R2216	AAACCTACCGAGCCTGGTG TGTTGAGCTTGAACGCTTTC
9	F2105 R2660	GAGGAACAGCTCTTTGGACAC AGAGACAGCTGAACCCTCGTG
10	F2417 R3006	CACTGTCAACCCAACACAGG ATGTCCTGATCCAACATCGAG
11	F2834 R3557	CCCAACCTCCGAGCAGTACATG AGAAGAGCGATGGTGAGAGC
12	F2972 R3557	ATAGGGTTTACGACCTCGATG AGAAGAGCGATGGTGAGAGC
13	F3234 R3557	AGATGGCAGAGCCCGGTAATC AGAAGAGCGATGGTGAGAGC
14	F3441 R3940	ACTACAACCCTTCGCTGACG TGAAGCCTGAGACTAGTTCGG
15	F3635 R4162	GCCTAGCCGTTTACTCAATCC TGAGTTGGTCGTAGCGGAATC
16	F3931 R4728	TCAGGCTTCAACATCGAATACG TTATGGTTCATTGTCCGGAGAG



Table 2. Continued

17	F4183 R4728	TTTCTACCACTCACCCCTAGCATTAC TTATGGTTCATTGTCCGGAGAG
18	F4392 R4983	CCCATCCTAAAGTAAGGTCAGC GGTTTAATCCACCTCAACTGCC
19	F4447 R4982	TTGGTTATAACCCTTCCCCTAC GTTTAATCCACCTCAACTGCC
20	F4797 R5553	CCCTTTCACTTCTGAGTCCCAG AGGGCTTTGAAGGCTCTTG
21	F4976 R5553	ATTAAACCAGACCCAGCTACG AGGGCTTTGAAGGCTCTTG
22	F5318 R5882	CACCATCACCCCTCCTTAACC GCTGAGTGAAGCATTGGACTG
23	F5700 R6262	TAAGCACCCCTAATCAACTGGC GCCTCCACTATAGCAGATGCG
24	F5999 R6526	TCTAAGCCTCCTTATTCGAGC ATAGTGATGCCAGCAGCTAGG
25	F6242 R6526	CGCATCTGCTATAGTGGAGG ATAGTGATGCCAGCAGCTAGG
26	F6426 R7030	GCCATAACCCAATACCAAACG TGGGCTACAACGTAGTACGTG
27	F6744 R7255	GGCTTCCTAGGGTTTATCGTG TTTCATGTGGTGTATGCATCG
28	F7075 R7792	GAGGCTTCATTCAGTATTTC GGGCAGGATAGTTCAGACGG
29	F7215 R7792	CGACGTTACTCGGACTACCC GGGCAGGATAGTTCAGACGG
30	F7645 R8215	TATCACCTTTCATGATCACGC GACGATGGGCATGAAACTG
31	F7901 R8311	TGAACCTACGAGTACACCGACTAC AAGTTAGCTTTACAGTGGGCTCTAG
32	F8164 R8669	CGGTCAATGCTCTGAAATCTGTG CATTGTTGGGTGGTGATTAGTCG
33	F8539 R9059	CTGTTGCTTCATTCATTGCC GTGGCGCTTCCAATTAGGTG
34	F8903 R9403	CCCACTTCTTACCACAAGGC GTGCTTTCTCGTGTTACATCG

Table 2. Continued

17	F4183 R4728	TTTCTACCACTCACCTAGCATTAC TTATGGTTCATTGTCCGGAGAG
18	F4392 R4983	CCCATCCTAAAGTAAGGTCAGC GGTTTAATCCACCTCAACTGCC
19	F4447 R4982	TTGGTTATACCCTTCCCGTAC GTTTAATCCACCTCAACTGCC
20	F4797 R5553	CCCTTTCACTTCTGAGTCCCAG AGGGCTTTGAAGGCTCTTG
21	F4976 R5553	ATTAAACCAGACCCAGCTACG AGGGCTTTGAAGGCTCTTG
22	F5318 R5882	CACCATCACCTCCTTAACC GCTGAGTGAAGCATTGGACTG
23	F5700 R6262	TAAGCACCTAATCAACTGGC GCCTCCACTATAGCAGATGCG
24	F5999 R6526	TCTAAGCCTCCTTATTCGAGC ATAGTGATGCCAGCAGCTAGG
25	F6242 R6526	CGCATCTGCTATAGTGGAGG ATAGTGATGCCAGCAGCTAGG
26	F6426 R7030	GCCATAACCCAATACCAAACG TGGGCTACAACGTAGTACGTG
27	F6744 R7255	GGCTTCCTAGGGTTTATCGTG TTTCATGTGGTGTATGCATCG
28	F7075 R7792	GAGGCTTCATTCAGTATTCC GGCAGGATAGTTCAGACGG
29	F7215 R7792	CGACGTTACTCGGACTACCC GGCAGGATAGTTCAGACGG
30	F7645 R8215	TATCACCTTTCATGATCACGC GACGATGGGCATGAAACTG
31	F7901 R8311	TGAACCTACGAGTACACCGACTAC AAGTTAGCTTTACAGTGGGCTCTAG
32	F8164 R8669	CGGTCAATGCTCTGAAATCTGTG CATTGTTGGGTGGTGATTAGTCG
33	F8539 R9059	CTGTTGCTTCATTCATTGCC GTGGCGCTTCCAATTAGGTG
34	F8903 R9403	CCCACTTCTTACCACAAGGC GTGCTTCTCGTGTTACATCG

Table 2. Continued

35	F9309 R9848	TTTCACTTCCACTCCATAACGC GAAAGTTGAGCCAATAATGACG
36	F9449 R9995	CGGGATAATCCTATTTATTACCTCAG AGAGTAAGACCCTCATCAATAGATGG
37	F9754 R10275	AGTCTCCCTTCACCATTTCGG AAAGGAGGGCAATTTCTAGATC
38	F10127 R10556	ACTACCACAACCTCAACGGCTAC GGAGGATATGAGGTGTGAGCG
39	F10386 R11166	GGATTAGACTGAACCGAATTGG CATCGGGTGATGATAGCCAAG
40	F10704 R11267	GTCTCAATCTCCAACACATATGG TGTTGTGAGTGTAATTAGTGCG
41	F11001 R11600	AACGCCACTTATCCAGTGAACC CTGTTTGTCTGATAGGCAGATGG
42	F11403 R11927	GACTCCCTAAAGCCCATGTGC TTGATCAGGAGAACGTGGTTAC
43	F11760 R12189	ACGAACGCACTCACAGTCG AAGCCTCTGTTGTCAGATTCAC
44	F11901 R12876	TGCTAGTAACCACGTTCTGGTG GATATCGCCGATACGGTTG
45	F12357 R12876	AACCACCCTAACCCTGACTTCC GATATCGCCGATACGGTTG
46	F12601 R13123	TTCATCCCTGTAGCATTGTTCCG AGCGGATGAGTAAGAAGATTCC
47	F12793 R13343	TTGCTCATCAGTTGATGATACG TTGAAGAAGGCGTGGGTACAG
48	F13188 R13611	CACTCTGTTTCGACAGTATG TCGAGTGCTATAGGCGCTTGTC
49	F13518 R13935	CATCATCGAAACCGCAAAC TGTGATGCTAGGGTAGAATCCG
50	F13715 R14118	GAAGCCTATTCGCAGGATTTCC TGGGAAGAAGAAAGAGAGGAAG
51	F13899 R14388	TTTCTCCAACATACTCGGATTC TTAGCGATGGAGGTAGGATTCG
52	F14189 R14926	ACAAACAATGGTCAACCAGTAAC TGAGGCGTCTGGTGAGTAGTGC

Table 2. Continued

53	F14470 R14996	TCCAAAGACAACCATCATTCC CGTGAAGGTAGCGGATGATTC
54	F14909 R15396	TACTCACCAGACGCCTCAACCG TTATCGGAATGGGAGGTGATTC
55	F15260 R15774	AGTCCCACCCTCACACGATTC ACTGGTTGTCCTCCGATTCAGG
56	F15574 R16084	CGCCTACACAATTCTCCGATC CGGTTGTTGATGGGTGAGTC
57 (HV1)	F15971 R16451	TTAACTCCACCATTAGCACC GCGAGGAGAGTAGCACTCTTG
58	F16097 R336	TACATTACTGCCAGCCACCATG TTAAGTGCTGTGGCCAGAAG
-21M13	F	TGTAAAACGACGGCCAGT

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- [1] Levin, B.C., Cheng, H., and Reeder, D.J., "Human Mitochondrial DNA Standard Reference Material for Quality Control in Forensic Identification, Medical Diagnosis, and Mutation Detection," *Genomics* **55**, pp. 135-146, (1999).
- [2] Anderson, S., Bankier, A.T., Barrell, B.G., deBruijn, M.H.L., Coulson, A.R., Drouin, J., Eperon, I.C., Nierlich, D.P., Roe, B.A., Sanger, F., Schreier, P.H., Smith, A.J.H., Staden, R., and Young, I.G., "Sequence and Organization of the Human Mitochondrial Genome," *Nature* **290**, pp. 457-465, (1981).

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Table 2. Continued

53	F14470 R14996	TCCAAAGACAACCATCATTCC CGTGAAGGTAGCGGATGATTC
54	F14909 R15396	TACTCACCAGACGCCTCAACCG TTATCGGAATGGGAGGTGATTC
55	F15260 R15774	AGTCCCACCCTCACACGATTC ACTGGTTGTCCTCCGATTCAGG
56	F15574 R16084	CGCCTACACAATTCTCCGATC CGGTTGTTGATGGGTGAGTC
57 (HV1)	F15971 R16451	TTAACTCCACCATTAGCACC GCGAGGAGAGTAGCACTCTTG
58	F16097 R336	TACATTACTGCCAGCCACCATG TTAAGTGCTGTGGCCAGAAG
-21M13	F	TGTA AAAACGACGGCCAGT

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- [1] Levin, B.C., Cheng, H., and Reeder, D.J., "Human Mitochondrial DNA Standard Reference Material for Quality Control in Forensic Identification, Medical Diagnosis, and Mutation Detection," *Genomics* **55**, pp. 135-146, (1999).
- [2] Anderson, S., Bankier, A.T., Barrell, B.G., deBruijn, M.H.L., Coulson, A.R., Drouin, J., Eperon, I.C., Nierlich, D.P., Roe, B.A., Sanger, F., Schreier, P.H., Smith, A.J.H., Staden, R., and Young, I.G., "Sequence and Organization of the Human Mitochondrial Genome," *Nature* **290**, pp. 457-465, (1981).

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