

Department of Forensic Science

**FORENSIC BIOLOGY
PROCEDURES MANUAL**

**INTERPRETATION OF
POWERPLEX[®] 16 BIO
SYSTEM DATA**

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1 INTERPRETATION OF POWERPLEX® 16 BIO SYSTEM DATA

1.1 Technical Notes

- 1.1.1 STR alleles are small in size, generally less than 500 bp and contain repeat units ranging from 3 to 7 bases.
- 1.1.2 If an allele contains an incomplete repeat, the allele is considered a microvariant and is designated by the number of complete repeats present followed by a decimal point, followed by the number of bases of the incomplete repeat. For example, the FGA 22.2 allele contains 22 tetrameric repeats plus 2 bases. Because of a deletion of two bases the FGA 22.2 allele is two bases shorter than the FGA 23 allele.
- 1.1.3 The characteristics of the PowerPlex® 16 BIO System and the allelic ladders are given in the table below:

Locus	* Repeat Sequence 5' 3'	Chromosome Location	Size Range of Allelic Ladder (bp)	Alleles present in Allelic Ladder	Fluorescent Label
FGA	TTTC Complex	4q28	322-444	16-30, 31.2, 43.2, 44.2, 45.2, 46.2	Rhodamine Red™ -X
TPOX	AATG	2p23-2pter	262-290	6-13	Rhodamine Red™ -X
D8S1179	TCTA Complex	8q	203-247	7-18	Rhodamine Red™ -X
vWA	TCTA Complex	12p12-pter	123-171	10-22	Rhodamine Red™ -X
Amelogenin	NA	Xp22.1-22.3 and Y	106 - X 112 - Y	X, Y	Rhodamine Red™ -X
Penta E	AAAGA	15q	379-474	5-24	Fluorescein
D18S51	AGAA	18q21.3	290-366	8-10, 10.2, 11-13, 13.2, 14-27	Fluorescein
D21S11	TCTA Complex	21q11-21q21	203-259	24, 24.2, 25, 25.2, 26-28, 28.2, 29, 29.2, 30, 30.2, 31, 31.2, 32, 32.2, 33, 33.2, 34, 34.2, 35, 35.2, 36-38	Fluorescein
TH01	AATG	11p15.5	156-195	4-9, 9.3, 10-11, 13.3	Fluorescein
D3S1358	TCTA Complex	3p	115-147	12-20	Fluorescein
Penta D	AAAGA	21q	376-449	2.2, 3.2, 5, 7-17	JOE
CSF1PO	AGAT	5q33.3-34	321-357	6 - 15	JOE
D16S539	AGAT	16q24-qter	264 - 304	5, 8 -15	JOE
D7S820	AGAT	7q11.21-22	215 -247	6 - 14	JOE
D13S317	AGAT	13q22-q31	169 -201	7 - 15	JOE
D5S818	AGAT	5q23.3-32	119 -155	7 - 16	JOE

* All repeat sequences are defined using the recommendation of the DNA Commission of the International Society of Forensic Haemogenetics (ISFH): 1) for STR loci within coding genes, the

coding strand shall be used and the repeat sequence motif defined using the first possible 5' nucleotide of the repeat motif; and 2) for STR loci not associated with a coding gene, the first database entry or original literature description shall be used.

FMBIO II and FMBIO III Plus Fluorescent Image Analysis Systems:

Fluorescein is detected at a wavelength of 505 nm (FMBIO II) or 520 nm (FMBIO III Plus) - Green

Rhodamine Red™ -X is detected at a wavelength of 598 nm - Red

JOE = 6-carboxy-4',5'-dichloro 2',7' - dimethoxyfluorescein is detected at a wavelength of 577 nm - Yellow

- 1.1.4 The Fluorescent Internal Lane Standard 600 BIO (Texas Red®-X) consists of 21 DNA fragments (80,100, 120, 140, 160, 180, 200, 225, 250, 275, 300, 325, 350, 375, 400, 425, 450, 475, 500, 550, and 600 bp) and can be detected at a wavelength of 665 nm (FMBIO II) or 650 nm (FMBIO III Plus) – Blue.
- 1.1.5 The “Known” Genotypes for the Control DNA (GM9947A Cell Line) using the PowerPlex® 16 BIO System are given in the table below:

Locus	Genotype GM9947A
Penta E	12,13
D18S51	15,19
D21S11	30,30
TH01	8,9.3
D3S1358	14,15
FGA	23,24
TPOX	8,8
D8S1179	13,13
VWA	17,18
Amelogenin	X,X
Penta D	12,12
CSF1PO	10,12
D16S539	11,12
D7S820	10,11
D13S317	11,11
D5S818	11,11

1.2 Procedure

- 1.2.1 The gel image is visually inspected to determine if the number, position, and intensity of the alleles for the allelic ladder, controls and samples are suitable for interpretation.

- 1.2.1.1 If the overall quality of the gel image is unsuitable for interpretation, no further comparisons are conducted.
- 1.2.1.2 If the overall quality of the gel image is suitable for interpretation, a visual comparison is performed.
- 1.2.2 The source of a DNA sample may be from a single person or more than one person. This can be determined by examination of the number of alleles at each locus, optical densities and/or band intensities.
 - 1.2.2.1 A DNA profile may be considered to have originated from a single individual if the expected number of alleles (i.e., 1 or 2) is observed at each locus and the intensity of the alleles within a locus is approximately the same. All loci should be taken into account when making this determination.
 - 1.2.2.2 A sample may be considered to be a mixture of DNA from two or more individuals if the sample contains 3 or more bands at one or more loci and/or there is a distinct difference in signal intensity. All loci should be taken into account when making this determination.
- 1.2.3 To the extent possible the DNA profiles of the evidence samples are determined first and then compared to the reference samples.
 - 1.2.3.1 If the banding patterns of samples under comparison are distinctly different in position, it is concluded that the samples originated from different sources than the individual of interest and the individual is excluded.
 - 1.2.3.2 If the banding patterns of samples under comparison appear visually consistent in position, the possibility that both samples may have originated from the same source cannot be eliminated. Therefore the individual of interest is included as a possible source.
 - 1.2.3.3 If evidence samples under comparison contain a partial profile (i.e., allele dropout) or an incomplete profile (i.e., locus dropout) due to degradation, inhibition or limited DNA, the DNA profile may or may not be interpretable.
 - 1.2.3.3.1 All loci will be taken into account when making this determination using knowledge of the system and experience.
 - 1.2.3.3.2 If allele/locus dropout is observed at a majority of the loci, in order to include an individual, at least four callable loci from the evidence must match the known standard (this includes loci where masking has occurred). Otherwise the partial profile will be reported as inconclusive or may be used only for elimination purposes. However, additional information such as the presence of a rare allele observed only in a small portion of the population will be taken into consideration in consultation with the Biology Program Manager when reaching a conclusion.
 - 1.2.3.4 In criminal paternity/maternity and missing person cases, an individual must be eliminated at three or more loci to account for the possibility of mutations before the individual is eliminated as a parent/offspring.
 - 1.2.3.4.1 When a couple is evaluated as possible biological parents of a missing person, each possible parent's DNA profile will be evaluated separately to determine if the individual is included or eliminated as a biological parent. Subsequently the profiles from both individuals will be evaluated together to determine if as a couple they could have conceived the missing person.

1.2.4 All controls must work appropriately.

1.2.4.1 Reagent Blanks

1.2.4.1.1 If a weak signal is detected in a reagent blank at a single locus and the signal is demonstrated to be part of the control, the test results associated with the reagent blank will be considered inconclusive at that locus. If a weak signal is detected in a reagent blank at multiple loci, the test results for all loci will be considered inconclusive.

1.2.4.1.2 If a strong signal is detected in a reagent blank at a single locus or at multiple loci, the test results for all loci will be considered inconclusive.

1.2.4.2 The Control DNA (GM9947A Cell Line) must elicit the "Known" genotype for each locus. If an allele is detected in the Control DNA at a specific locus that is not consistent with the known genotype, the test will be considered inconclusive at that locus.

1.2.4.3 Negative Amplification Control

1.2.4.3.1 If a weak signal is detected in the negative amplification control at a single locus and the signal is demonstrated to be part of the control, the test results associated with the negative amplification control will be considered inconclusive at that locus.

1.2.4.3.2 If a weak signal is detected in the negative amplification control at multiple loci, the test results for all loci will be considered inconclusive.

1.2.4.3.3 If a strong signal is detected in the negative amplification control at a single locus or at multiple loci, the test results for all loci will be considered inconclusive.

1.2.5 When a band stronger in intensity is accompanied by a band weaker in intensity that has migrated one allele position (n-4) farther than the more intense band, this may be a stutter band.

NOTE: Stutter may also be seen at an n+4 position to that of the more intense band. In addition, if the sample is overloaded, a high concentration of DNA was amplified, or the sample is degraded, artifactual bands may also be seen at n-1, n-2, n-3 and n-8 positions to the intense band or the stutter band may have an elevated optical density values.

1.2.5.1 All loci must be taken into account when determining if a sample is a mixture of biological material from more than one source. In order to determine if a weak band in an n-4 position is the result of normal stutter within a locus or a mixture of two or more sources of biological material, the analyst will use experience and/or the percent stutter values to make an informed decision. If the ratio of the OD (optical density) for the strong band to the weak band is less than the established values (listed below) the band may be considered to be stutter. The following percent stutter values will serve as a guide:

Locus	% Stutter (FMBIO II)	% Stutter (FMBIO III Plus)	Locus	% Stutter (FMBIO II)	% Stutter (FMBIO III Plus)
FGA	11.0	13.0	D3S1358	12.0	16.0
TPOX	8.0	11.0	Penta D	2.0**	2.0**
D8S1179	10.0	12.0	CSF1PO	11.0	13.0
VWA	16.0	16.0	D16S539	12.0	13.0
Penta E	2.0**	2.0**	D7S820	11.0	12.0
D18S51	13.0	13.0	D13S317	10.0	11.0

NOTE: table continued on next page

Locus	% Stutter (FMBIO II)	% Stutter (FMBIO III Plus)	Locus	% Stutter (FMBIO II)	% Stutter (FMBIO III Plus)
D21S11	15.0	15.0	D5S818	13.0	13.0
TH01	5.0	5.0			

** No stutter was observed during the validation studies. Therefore, the percent stutter specified is based upon recommendations of the manufacturer reported in the PowerPlex® 2.1 System Technical Manual.

1.2.5.2 If the ratio of the OD (optical density) for the strong band to the weak band is less than the established stutter value (listed above), the allele will be considered to be stutter and will not be called even if it is believed that the band is a true allele.

1.2.5.3 If the ratio of the OD (optical density) for the strong band to the weak band is above the established stutter value (this event is generally observed in samples containing a high concentration of DNA or the DNA is partially degraded), the allele may still be called stutter once all loci have been taken into account.

1.2.6 When a strong band in intensity and a weak band in intensity are observed within a single locus and the bands are separated by greater than one repeat unit, the difference in intensity could be the result of a null allele or the inability of the primer to bind to the template fully in the flanking region of one of the alleles. In order to determine if a DNA profile containing bands with a difference in intensity is a result of a null allele, primer mis-pairing or a mixture of biological material from more than one source, the analyst must take into account all of the loci, use experience and/or the heterozygous percent intensity values to make an informed decision. If the percentage values obtained from the ratio of the OD (optical density) for the stronger band to the weaker band is equal to or greater than the established values (listed below), both bands within the locus may be considered to have originated from a single donor. The following heterozygous percent intensity values will serve as a guide:

Locus	Lower Limit Difference Between Two Heterozygous Alleles (3 STD below the mean)	Locus	Lower Limit Difference Between Two Heterozygous Alleles (3 STD below the mean)
Penta E	45%	VWA	65%
D18S51	54%	Amelogenin	N/A
D21S11	63%	Penta D	No data available at this time
TH01	70%	CSF1PO	62%
D3S1358	67%	D16S539	64%
FGA	57%	D7S820	63%
TPOX	63%	D13S317	66%
D8S1179	63%	D5S818	68%

NOTE: Data generated from internal validation conducted by the Virginia Department of Forensic Science

1.3 Interpretation of PowerPlex® 16 BIO System Alleles

Amplified PowerPlex® 16 BIO System alleles are typed by noting which allelic ladder band(s) lines up with the test sample band(s), as demonstrated in the following examples:

	C	L	S	
46.2		—		
45.2		—		
44.2		—	—	
43.2		—		
				FGA
31.2		—		
30		—		
29		—		
28		—		
27		—	—	
26		—		
25		—		
24		—		
23	—	—		
22	—	—		
21		—		
20		—		
19		—		
18		—		
17		—		
13		—		
12		—	—	
11		—	—	
10		—	—	
9		—	—	TPOX
8	—	—		
7	—	—		
6		—		
18		—		
17		—		
16		—	—	
15		—		
14		—		D8S1179
13	—	—		
12		—		
11		—		
10		—		
9		—		
8		—		
7		—		
22		—		
21		—		
20		—	—	
19		—		
18	—	—		vWA
17	—	—		
16		—		
15		—	—	
14		—		
13		—		
12		—		
11		—		
10		—		
Y		—	—	
X	—	—	—	Amelogenin

In the example on the previous page, the FGA alleles for the sample lane (S) line up with alleles 28 and 45.2 of the allelic ladder (L), the TPOX alleles line up with alleles 10 and 12 of the allelic ladder, the D8S1179 allele lines up with allele 16 of the allelic ladder, the vWA alleles line up with alleles 15 and 21 of the allelic ladder, and the Amelogenin alleles line up with the X and Y alleles of the allelic ladder. Therefore, this sample has a genotype of FGA - 28, 45.2; TPOX - 10,12; D8S1179 – 16; vWA - 15,21; and Amelogenin X,Y. The Control DNA (9947A Cell Line designated as C) which is run on every gel has a genotype of FGA -23,24; TPOX – 8; D8S1179 – 13; vWA - 17,18; and Amelogenin X,X.

1.4 Interpretation of a Profile Different from an Assumed Contributor in a Mixture Searched in CODIS (Combined DNA Index System)

Example:

Channel 4 – JOE

	V	L	E	
17		—		
16		—		
15		—		
14		—		
	13	—		
12		—	—	—
11		—	
10		—		Penta D
9	—	—	—	
8		—	
7		—		
5		—		
3.2				
2.2				
15		—		
14	—	—	—	
13		—	
12		—		
11		—		CSF1PO
10	—	—	—	
9		—	—	
8		—	
7		—		
6		—		
15		—		
14		—	
13		—		
12	—	—	—	
11	—	—	—	D16S539
10		—		
9		—		
8		—		
5		—		
14		—		
13		—		
12		—	
11	—	—	—	
10	—	—	—	D7S820
9	—	—	—	
8		—		
7		—		
	6	—	—	
15		—		
14		—		
13		—		
12		—	
11		—		
10		—		
9		—		
8	—	—	—	
7		—		
15		—		
14		—		
13		—	
12	—	—	—	
11	—	—	—	D5S818
10	—	—	—	
9		—		
8		—		
7		—		

Note: Indicates an allele of weaker intensity

— Indicates an allele of stronger intensity

In the example on the previous page, the victim (V) has the following profile: Penta D 9, 13; CSF1PO 10, 14; D16S539 11,12; D7S820 9,11; D13S317 8; and D5S818 11,12. Therefore, the minimum DNA profile different from the victim that would be searched in CODIS would be:

Penta D	8, 12	(weaker intensity alleles)
CSF1PO	8, 13	(weaker intensity alleles)
D16S539	11, 14	The 11 allele is twice as intense as the 12 allele which is provided by the victim. Therefore, a conclusion can be reached that the contribution of the 11 allele based on the intensity is not solely from the victim. The 14 allele is different from the victim.
D7S820	10, 12	(weaker intensity alleles)
D13S317	12	(weaker intensity allele)
D5S818	11, 13	The 11 allele is twice as intense as the 12 allele which is provided by the victim. Therefore, a conclusion can be reached that the contribution of the 11 allele based on the intensity is not solely from the victim. The 13 allele is different from the victim.

APPENDIX A - REFERENCES

1. PowerPlex® 16 BIO System Technical Manual
2. Bär W. *et al.* (1997) DNA recommendations: further report of the DNA Commission of the ISFH regarding the use of short tandem repeat systems, *Int. J. Legal Med.* **110**, 175.
3. Gill, P. *et al.* (1997) Considerations from the European DNA profiling group (EDNAP) concerning STR nomenclature. *Forensic Science International* **87**, 185-192.
4. Gill, P. *et al.* (2000) An investigation of the rigor of interpretation rules for STRs derived from less than 100 pg of DNA, *Forensic Science International* **112**, 17-40.
5. Curran, J.M., *et al.* (2005) Interpretation of repeat measurement DNA evidence allowing for multiple contributors and population substructure, *Forensic Science International* **148**, 47-53.

APPENDIX B – STR POPULATION FREQUENCIES

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
BLACK POPULATION DATA
CSF1P0, TPOX, TH01 AND vWA ALLELE FREQUENCY TABLE**

DATE: JUNE 1, 1998 N = 194

ALLELE CSF1P0	OBSERVATION	FREQUENCY		ALLELE TH01	OBSERVATION	FREQUENCY
15	0*	0.01289		11	0*	0.01289
14	2*	0.01289		10	4*	0.01289
13	24	0.06186		9.3	34	0.08762
12	107	0.27577		9	50	0.12886
11	90	0.23196		8	96	0.24742
10	92	0.23711		7	154	0.39690
9	14	0.03608		6	47	0.12113
8	29	0.07474		5	3*	0.01289
7	30	0.07732				
6	0*	0.01289				
n = 388				n = 388		
ALLELE TPOX	OBSERVATION	FREQUENCY		ALLELE VWA	OBSERVATION	FREQUENCY
13	1*	0.01289		21	1*	0.01289
12	8	0.02062		20	11	0.02835
11	88	0.22680		19	28	0.07216
10	32	0.08247		18	44	0.11340
9	86	0.22165		17	61	0.15722
8	138	0.35567		16	110	0.28351
7	6	0.01546		15	96	0.24742
6	29	0.07474		14	29	0.07474
				13	6	0.01546
				12	0*	0.01289
				11	2*	0.01289
n = 388				n = 388		

Note: * = Alleles with fewer than 5 observations are defaulted to a frequency of 5 per 388, or 0.01289. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed above a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
BLACK POPULATION DATA
D16S539, D7S820, D13S317 AND D5S818 ALLELE FREQUENCY TABLE**

DATE: JUNE 1, 1998 N = 194

ALLELE D16S539	OBSERVATION	FREQUENCY		ALLELE D13S317	OBSERVATION	FREQUENCY
15	1*	0.01289		15	1*	0.01289
14	12	0.03093		14	24	0.06190
13	56	0.14433		13	54	0.13918
12	73	0.18814		12	158	0.40722
11	113	0.29124		11	127	0.32732
10	37	0.09536		10	7	0.01804
9	81	0.20876		9	10	0.02577
8	15	0.03866		8	7	0.01804
5	0*	0.01289		7	0*	0.01289
n = 388				n = 388		
ALLELE D7S820	OBSERVATION	FREQUENCY		ALLELE D5S818	OBSERVATION	FREQUENCY
14	1*	0.01289		15	0*	0.01289
13	7	0.01804		14	10	0.02577
12	41	0.10567		13	89	0.22938
11	90	0.23196		12	141	0.36340
10	132	0.34021		11	99	0.25515
9	41	0.10567		10	26	0.06701
8	73	0.18814		9	8	0.02062
7	3*	0.01289		8	15	0.03866
6	0*	0.01289		7	0*	0.01289
n = 388				n = 388		

Note: * = Alleles with fewer than 5 observations are defaulted to a frequency of 5 per 388, or 0.01289. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed in the above table a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
BLACK POPULATION DATA
D18S51 AND D21S11 ALLELE FREQUENCY TABLE**

DATE: MAY 19, 2000 N = 192

ALLELE D18S51	OBSERVATION	FREQUENCY		ALLELE D18S51 cont.	OBSERVATION	FREQUENCY
17	60	0.15625		27	0*	0.01302
16	68	0.17708		26	0*	0.01302
15	66	0.17188		25	0*	0.01302
14	25	0.06510		24	0*	0.01302
13.2	1*	0.01302		23	1*	0.01302
13	27	0.07031		22	4*	0.01302
12	28	0.07292		21	6	0.01563
11	0*	0.01302		20	18	0.04688
10.2	3*	0.01302		19	28	0.07292
10	3*	0.01302		18	46	0.11979
9	0*	0.01302				
8	0*	0.01302		n = 384		
ALLELE D21S11	OBSERVATION	FREQUENCY		ALLELE D21S11 cont.	OBSERVATION	FREQUENCY
31.2	19	0.04948		38	0*	0.01302
31	26	0.06771		37	0*	0.01302
30.2	7	0.01823		36	3*	0.01302
30	74	0.19271		35.2	0*	0.01302
29.2	0*	0.01302		35	11	0.02865
29	79	0.20573		34.2	0*	0.01302
28	97	0.25260		34	4*	0.01302
27	16	0.04167		33.2	11	0.02865
26	1*	0.01302		33	3*	0.01302
25.2	0*	0.01302		32.2	30	0.07813
25	0*	0.01302		32.1	0*	0.01302
24.2	0*	0.01302		32	3*	0.01302
24	0*	0.01302		n = 384		

Note: * = Alleles with fewer than 5 observations are defaulted to a frequency of 5 per 384, or 0.01302. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed above a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
BLACK POPULATION DATA
Penta E AND D3S1358 ALLELE FREQUENCY TABLE**

DATE: MAY 19, 2000 N = 168 (Penta E) and 192 (D3S1358)

ALLELE Penta E	OBSERVATION	FREQUENCY		ALLELE Penta E cont.	OBSERVATION	FREQUENCY
15	18	0.05357		25	0*	0.01488
14	20	0.05952		24	0*	0.01488
13	46	0.13691		23	0*	0.01488
12	33	0.09821		22	0*	0.01488
11	26	0.07738		21	1*	0.01488
10	16	0.04762		20.3	0*	0.01488
9	12	0.03571		20	3*	0.01488
8	64	0.19048		19	0*	0.01488
7	37	0.11012		18	4*	0.01488
6	0*	0.01488		17	12	0.03571
5	34	0.10119		16	10	0.02976
				n = 336		
ALLELE D3S1358	OBSERVATION	FREQUENCY		ALLELE D3S1358 cont.	OBSERVATION	FREQUENCY
16.2	1*	0.01302		21	0*	0.01302
16	103	0.26823		20	0*	0.01302
15	124	0.32292		19	1*	0.01302
14	45	0.11719		18	21	0.05469
13	4*	0.01302		17	81	0.21094
12	3*	0.01302				
11	1*	0.01302		n = 384		

Note: * = Alleles for Penta E with fewer than 5 observations are defaulted to a frequency of 5 per 336, or 0.01488. Alleles for D3S1358 with fewer than 5 observations are defaulted to a frequency of 5 per 384, or 0.01302. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed in the above table a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
BLACK POPULATION DATA
FGA AND D8S1179 ALLELE FREQUENCY TABLE**

DATE: MAY 19, 2000 N = 192

ALLELE FGA	OBSERVATION	FREQUENCY		ALLELE FGA cont	OBSERVATION	FREQUENCY
25	33	0.08594		46.2	1*	0.01302
24.2	0*	0.01302		45.2	0*	0.01302
24	54	0.14063		44.2	2*	0.01302
23.2	0*	0.01302		43.2	0*	0.01302
23	66	0.17188		42.2	1*	0.01302
22.2	0*	0.01302		31.2	0*	0.01302
22	77	0.20052		31	1*	0.01302
21.2	1*	0.01302		30.2	1*	0.01302
21	39	0.10156		30	0*	0.01302
20.2	1*	0.01302		29	2*	0.01302
20	30	0.07813		28	7	0.01823
19.2	1*	0.01302		27	11	0.02865
19	25	0.06510		26.1	0*	0.01302
18.2	9	0.02344		26	18	0.04688
18	4*	0.01302		25.2	0*	0.01302
17	0*	0.01302				
				n = 384		
ALLELE D8S1179	OBSERVATION	FREQUENCY		ALLELE D8S1179	OBSERVATION	FREQUENCY
12	42	0.10938		18	0*	0.01302
11	19	0.04948		17	7	0.01823
10	11	0.02865		16	23	0.05990
9	2*	0.01302		15	71	0.18490
8	2*	0.01302		14	114	0.29688
7	0*	0.01302		13	93	0.24219
				n = 384		

Note: * = Alleles with fewer than 5 observations are defaulted to a frequency of 5 per 384, or 0.01302. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed in the above table a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
BLACK POPULATION DATA
Penta D ALLELE FREQUENCY TABLE**

DATE: JANUARY 7, 2002 N = 100

ALLELE Penta D	OBSERVATION	FREQUENCY		ALLELE Penta D cont.	OBSERVATION	FREQUENCY
10	22	0.11000		17	0*	0.02500
9	34	0.17000		16	0*	0.02500
8	23	0.11500		15	2*	0.02500
7	9	0.04500		14	4*	0.02500
5	8	0.04000		13	15	0.07500
3.2	3*	0.02500		12	20	0.10000
2.2	27	0.13500		11	33	0.16500
				n = 200		

Note: * = Alleles for Penta D with fewer than 5 observations are defaulted to a frequency of 5 per 200, or 0.0250. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed in the above table a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
CAUCASIAN POPULATION DATA
CSF1P0, TPOX, THO1 AND vWA ALLELE FREQUENCY TABLE**

DATE: JUNE 1, 1998 N = 174

ALLELE CSF1P0	OBSERVATION	FREQUENCY		ALLELE THO1	OBSERVATION	FREQUENCY
15	0*	0.01437		11	0*	0.01437
14	3*	0.01437		10	4*	0.01437
13	27	0.07759		9.3	108	0.31034
12	125	0.35919		9	50	0.14368
11	97	0.27874		8	35	0.10057
10	83	0.23851		7	66	0.18966
9	11	0.03161		6	83	0.23851
8	1*	0.01437		5	2*	0.01437
7	1*	0.01437				
6	0*	0.01437				
n = 348				n = 348		
ALLELE TPOX	OBSERVATION	FREQUENCY		ALLELE Vwa	OBSERVATION	FREQUENCY
13	0*	0.01437		21	0*	0.01437
12	18	0.05172		20	2*	0.01437
11	86	0.24713		19	29	0.08333
10	23	0.06609		18	81	0.23276
9	31	0.08908		17	96	0.27586
8	190	0.54598		16	73	0.20977
7	0*	0.01437		15	33	0.09483
6	0*	0.01437		14	33	0.09483
				13	1*	0.01437
				12	0*	0.01437
				11	0*	0.01437
n = 348				n = 348		

Note: * = Alleles with fewer than 5 observations are defaulted to a frequency of 5 per 348, or 0.01437. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed above a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
CAUCASIAN POPULATION DATA
D16S539, D7S820, D13S317, AND D5S818 ALLELE FREQUENCY TABLE**

DATE: JUNE 1, 1998 N = 174

ALLELE D16S539	OBSERVATION	FREQUENCY		ALLELE D13S317	OBSERVATION	FREQUENCY
15	1*	0.01437		15	0*	0.01437
14	6	0.01724		14	15	0.04310
13	62	0.17816		13	40	0.11494
12	112	0.32184		12	100	0.28736
11	89	0.25575		11	102	0.29310
10	24	0.06897		10	20	0.05747
9	48	0.13793		9	23	0.06609
8	6	0.01724		8	48	0.13793
5	0*	0.01437		7	0*	0.01437
n = 348				n = 348		
ALLELE D7S820	OBSERVATION	FREQUENCY		ALLELE D5S818	OBSERVATION	FREQUENCY
14	3*	0.01437		15	0*	0.01437
13	8	0.02299		14	8	0.02299
12	54	0.15517		13	55	0.15805
11	66	0.18966		12	121	0.34770
10	108	0.31034		11	128	0.36782
9	47	0.13506		10	20	0.05747
8	52	0.14943		9	15	0.04310
7	10	0.02874		8	1*	0.01437
6	0*	0.01437		7	0*	0.01437
n = 348				n = 348		

Note: * = Alleles with fewer than 5 observations are defaulted to a frequency of 5 per 348, or 0.01437. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed above a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
CAUCASIAN POPULATION DATA
D18S51 AND D21S11 ALLELE FREQUENCY TABLE**

DATE: MAY 19, 2000 N = 173

ALLELE D18S51	OBSERVATION	FREQUENCY		ALLELE D18S51 cont.	OBSERVATION	FREQUENCY
17	51	0.14740		27	0*	0.01445
16	47	0.13584		26	0*	0.01445
15	55	0.15896		25	0*	0.01445
14	55	0.15896		24	0*	0.01445
13.2	0*	0.01445		23	0*	0.01445
13	49	0.14162		22	2*	0.01445
12	43	0.12428		21	1*	0.01445
11	6	0.01734		20	5	0.01445
10.2	0*	0.01445		19	6	0.01734
10	1*	0.01445		18	25	0.07225
9	0*	0.01445				
8	0*	0.01445		n = 346		
ALLELE D21S11	OBSERVATION	FREQUENCY		ALLELE D21S11 cont.	OBSERVATION	FREQUENCY
31.2	27	0.07804		38	0*	0.01445
31	24	0.06936		37	0*	0.01445
30.2	13	0.03757		36	0*	0.01445
30	91	0.26301		35.2	1*	0.01445
29.2	0*	0.01445		35	0*	0.01445
29	73	0.21098		34.2	3*	0.01445
28	40	0.11561		34	0*	0.01445
27	7	0.02023		33.2	13	0.03757
26	0*	0.01445		33	0*	0.01445
25.2	1*	0.01445		32.2	43	0.12428
25	1*	0.01445		32.1	1*	0.01445
24.2	0*	0.01445		32	8	0.02312
24	0*	0.01445		n = 346		

Note: * = Alleles with fewer than 5 observations are defaulted to a frequency of 5 per 346, or 0.01445. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed above a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
CAUCASIAN POPULATION DATA
Penta E AND D3S1358 ALLELE FREQUENCY TABLE**

DATE: MAY 19, 2000 N =120 (Penta E) and 173 (D3S1358)

ALLELE Penta E	OBSERVATION	FREQUENCY		ALLELE Penta E cont.	OBSERVATION	FREQUENCY
15	20	0.08333		25	0*	0.02083
14	11	0.04583		24	0*	0.02083
13	18	0.07500		23	0*	0.02083
12	41	0.17083		22	0*	0.02083
11	30	0.12500		21	0*	0.02083
10	19	0.07917		20.3	0*	0.02083
9	2*	0.02083		20	3*	0.02083
8	2*	0.02083		19	5	0.02083
7	50	0.20833		18	3*	0.02083
6	0*	0.02083		17	11	0.04583
5	16	0.06667		16	9	0.03750
				n = 240		
ALLELE D3S1358	OBSERVATION	FREQUENCY		ALLELE D3S1358 cont.	OBSERVATION	FREQUENCY
16.2	0*	0.01445		21	0*	0.01445
16	89	0.25723		20	2*	0.01445
15	82	0.23699		19	7	0.02023
14	48	0.13873		18	51	0.14734
13	0*	0.01445		17	66	0.19075
12	0*	0.01445				
11	1*	0.01445		n = 346		

Note: * = Alleles for Penta E with fewer than 5 observations are defaulted to a frequency of 5 per 240, or 0.02083. Alleles for D3S1358 with fewer than 5 observations are defaulted to a frequency of 5 per 346, or 0.01445. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed above a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
CAUCASIAN POPULATION DATA
FGA AND D8S1179 ALLELE FREQUENCY TABLE**

DATE: MAY 19, 2000 N = 173

ALLELE FGA	OBSERVATION	FREQUENCY		ALLELE FGA cont	OBSERVATION	FREQUENCY
25	31	0.08960		46.2	0*	0.01445
24.2	0*	0.01445		45.2	0*	0.01445
24	43	0.12428		44.2	0*	0.01445
23.2	0*	0.01445		43.2	0*	0.01445
23	45	0.13006		42.2	0*	0.01445
22.2	4*	0.01445		31.2	0*	0.01445
22	56	0.16185		31	0*	0.01445
21.2	0*	0.01445		30.2	0*	0.01445
21	66	0.19075		30	0*	0.01445
20.2	0*	0.01445		29	0*	0.01445
20	58	0.16763		28	0*	0.01445
19.2	1*	0.01445		27	4*	0.01445
19	20	0.05780		26.1	0*	0.01445
18.2	0*	0.01445		26	14	0.04046
18	3*	0.01445		25.2	0*	0.01445
17	1*	0.01445				
				n = 346		
ALLELE D8S1179	OBSERVATION	FREQUENCY		ALLELE D8S1179	OBSERVATION	FREQUENCY
12	49	0.14162		18	0*	0.01445
11	32	0.09249		17	3*	0.01445
10	37	0.10694		16	12	0.03468
9	2*	0.01445		15	31	0.08960
8	5	0.01445		14	68	0.19653
7	0*	0.01445		13	107	0.30925
				n = 346		

Note: * = Alleles with fewer than 5 observations are defaulted to a frequency of 5 per 346, or 0.01445
Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed above a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
CAUCASIAN POPULATION DATA
Penta D ALLELE FREQUENCY TABLE**

DATE: JANUARY 7, 2002 N = 101

ALLELE Penta D	OBSERVATION	FREQUENCY		ALLELE Penta D cont.	OBSERVATION	FREQUENCY
10	27	0.13366		17	0*	0.02475
9	46	0.22772		16	0*	0.02475
8	2*	0.02475		15	2*	0.02475
7	1*	0.02475		14	11	0.05446
5	0*	0.02475		13	43	0.21287
3.2	0*	0.02475		12	41	0.20297
2.2	0*	0.02475		11	29	0.14356
				n = 202		

Note: * = Alleles for Penta D with fewer than 5 observations are defaulted to a frequency of 5 per 202, or 0.02475. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed in the above table a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
HISPANIC POPULATION DATA
CSF1P0, TPOX, THO1 AND vWA ALLELE FREQUENCY TABLE**

DATE: JUNE 1, 1998 N = 181

ALLELE CSF1P0	OBSERVATION	FREQUENCY		ALLELE THO1	OBSERVATION	FREQUENCY
15	1*	0.01381		11	0*	0.01381
14	3*	0.01381		10	2*	0.01381
13	20	0.05525		9.3	73	0.20166
12	120	0.33149		9	49	0.13536
11	109	0.30111		8	34	0.09392
10	90	0.24862		7	100	0.27624
9	12	0.03315		6	104	0.28729
8	3*	0.01381		5	0*	0.01381
7	4*	0.01381				
6	0*	0.01381				
n = 362				n = 362		
ALLELE TPOX	OBSERVATION	FREQUENCY		ALLELE VWA	OBSERVATION	FREQUENCY
13	2*	0.01381		21	0*	0.01381
12	31	0.08564		20	5	0.01381
11	100	0.27624		19	24	0.06630
10	19	0.05249		18	52	0.14365
9	31	0.08564		17	89	0.24590
8	169	0.46685		16	106	0.29282
7	3*	0.01381		15	53	0.14641
6	7	0.01934		14	31	0.08564
				13	1*	0.01381
				12	0*	0.01381
				11	1*	0.01381
n = 362				n = 362		

Note: * = Alleles with fewer than 5 observations are defaulted to a frequency of 5 per 362, or 0.01381. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed above a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
HISPANIC POPULATION DATA
D16S539, D7S820, D13S317 AND D5S818 ALLELE FREQUENCY TABLE**

DATE: JUNE 1, 1998 N = 181

ALLELE D16S539	OBSERVATION	FREQUENCY		ALLELE D13S317	OBSERVATION	FREQUENCY
15	0*	0.01381		15	0*	0.01381
14	9	0.02486		14	27	0.07459
13	58	0.16022		13	56	0.15470
12	97	0.26796		12	80	0.22099
11	73	0.20166		11	79	0.21823
10	55	0.15193		10	20	0.05525
9	62	0.17127		9	61	0.16851
8	7	0.01934		8	39	0.10773
5	1*	0.01381		7	0*	0.01381
n = 362				n = 362		
ALLELE D7S820	OBSERVATION	FREQUENCY		ALLELE D5S818	OBSERVATION	FREQUENCY
14	4*	0.01381		15	0*	0.01381
13	6	0.01657		14	3*	0.01381
12	72	0.19890		13	58	0.16022
11	94	0.25967		12	104	0.28729
10	94	0.25967		11	126	0.34807
9	34	0.09392		10	21	0.05801
8	55	0.15193		9	27	0.07459
7	3*	0.01381		8	5	0.01381
6	0*	0.01381		7	18	0.04972
n = 362				n = 362		

Note: * = Alleles with fewer than 5 observations are defaulted to a frequency of 5 per 362, or 0.01381. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed above a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
HISPANIC POPULATION DATA
D18S51 AND D21S11 ALLELE FREQUENCY TABLE**

DATE: MAY 19, 2000 N = 183

ALLELE D18S51	OBSERVATION	FREQUENCY		ALLELE D18S51 cont.	OBSERVATION	FREQUENCY
17	55	0.15027		27	0*	0.01366
16	43	0.11749		26	0*	0.01366
15	51	0.13934		25	0*	0.01366
14	60	0.16393		24	0*	0.01366
13.2	0*	0.01366		23	2*	0.01366
13	51	0.13934		22	1*	0.01366
12	37	0.10109		21	1*	0.01366
11	4*	0.01366		20	10	0.02732
10.2	1*	0.01366		19	23	0.06284
10	1*	0.01366		18	26	0.07104
9	0*	0.01366				
8	0*	0.01366		n = 366		
ALLELE D21S11	OBSERVATIONS	FREQUENCY		ALLELE D21S11 cont.	OBSERVATION	FREQUENCY
31.2	42	0.11475		38	0*	0.01366
31	30	0.08197		37	0*	0.01366
30.2	4*	0.01366		36	1*	0.01366
30	88	0.24044		35.2	0*	0.01366
29.2	1*	0.01366		35	3*	0.01366
29	78	0.21312		34.2	2*	0.01366
28	42	0.11475		34	1*	0.01366
27	7	0.01913		33.2	16	0.04372
26	0*	0.01366		33	0*	0.01366
25.2	0*	0.01366		32.2	46	0.12568
25	0*	0.01366		32.1	0*	0.01366
24.2	0*	0.01366		32	5	0.01366
24	0*	0.01366		n = 366		

Note: * = Alleles with fewer than 5 observations are defaulted to a frequency of 5 per 366, or 0.01366. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed above a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
HISPANIC POPULATION DATA
Penta E AND D3S1358 ALLELE FREQUENCY TABLE**

DATE: MAY 19, 2000 N = 181 (Penta E) and 183(D3S1358)

ALLELE Penta E	OBSERVATION	FREQUENCY		ALLELE Penta E cont.	OBSERVATION	FREQUENCY
15	32	0.08840		25	1*	0.01381
14	28	0.07735		24	0*	0.01381
13	29	0.08011		23	1*	0.01381
12	58	0.16022		22	4*	0.01381
11	21	0.05801		21	8	0.02210
10	27	0.07459		20.3	0*	0.01381
9	6	0.01658		20	4*	0.01381
8	21	0.05801		19	12	0.03315
7	34	0.09392		18	14	0.03867
6	0*	0.01381		17	16	0.04420
5	19	0.05249		16	27	0.07459
				n = 362		
ALLELE D3S1358	OBSERVATION	FREQUENCY		ALLELE D3S1358 cont.	OBSERVATION	FREQUENCY
16	93	0.25410		21	1*	0.01366
15	143	0.39071		20	0*	0.01366
14	26	0.07104		19	3*	0.01366
13	1*	0.01366		18	30	0.08197
12	0*	0.01366		17	69	0.18853
11	0*	0.01366		16.2	0*	0.01366
				n = 366		

Note: * = Alleles for Penta E with fewer than 5 observations are defaulted to a frequency of 5 per 362, or 0.01381. Alleles for D3S1358 with fewer than 5 observations are defaulted to a frequency of 5 per 366, or 0.01366. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed in the above table a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
HISPANIC POPULATION DATA
FGA AND D8S1179 ALLELE FREQUENCY TABLE**

DATE: MAY 19, 2000 N = 183

ALLELE FGA	OBSERVATION	FREQUENCY		ALLELE FGA cont	OBSERVATION	FREQUENCY
25	47	0.12842		46.2	0*	0.01366
24.2	0*	0.01366		45.2	1*	0.01366
24	58	0.15847		44.2	0*	0.01366
23.2	0*	0.01366		43.2	0*	0.01366
23	46	0.12568		42.2	0*	0.01366
22.2	1*	0.01366		31.2	0*	0.01366
22	36	0.09836		31	0*	0.01366
21.2	1*	0.01366		30.2	0*	0.01366
21	65	0.17760		30	1*	0.01366
20.2	0*	0.01366		29	1*	0.01366
20	40	0.10929		28	2*	0.01366
19.2	0*	0.01366		27	9	0.02459
19	31	0.08470		26.1	1*	0.01366
18.2	0*	0.01366		26	25	0.06831
18	1*	0.01366		25.2	0*	0.01366
17	0*	0.01366				
				n = 366		
ALLELE D8S1179	OBSERVATION	FREQUENCY		ALLELE D8S1179	OBSERVATION	FREQUENCY
12	40	0.10929		18	0*	0.01366
11	17	0.04645		17	0*	0.01366
10	30	0.08197		16	17	0.04645
9	1*	0.01366		15	44	0.12022
8	3*	0.01366		14	96	0.26230
7	0*	0.01366		13	118	0.32240
				n = 366		

Note: * = Alleles with fewer than 5 observations are defaulted to a frequency of 5 per 366, or 0.01366. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed in the above table a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

**VIRGINIA DEPARTMENT OF FORENSIC SCIENCE
HISPANIC POPULATION DATA
Penta D ALLELE FREQUENCY TABLE**

DATE: NOVEMBER 13, 2001 N = 157

ALLELE Penta D	OBSERVATION	FREQUENCY		ALLELE Penta D.cont	OBSERVATION	FREQUENCY
10	49	0.15556		17	1*	0.01587
9	66	0.20952		16	0*	0.01587
8	9	0.02857		15	1*	0.01587
7	3*	0.01587		14	16	0.05079
5	3*	0.01587		13	56	0.17778
3.2	0	0.01587		12	59	0.18730
2.2	9	0.02857		11	43	0.13651
				n = 315**		

Note: * = Alleles for Penta D with fewer than 5 observations are defaulted to a frequency of 5 per 315, or 0.01587. Therefore, allele frequencies on this page will NOT add to 1. If an allele is observed that is not listed in the above table a frequency of 5/n will be used.

N = Total number of individuals
n = Total number of alleles from N individuals

** One of the samples analyzed for the creation of the Hispanic population database contained a 3 banded pattern at Penta D. Therefore, the number of alleles observed in the 157 individuals was 31