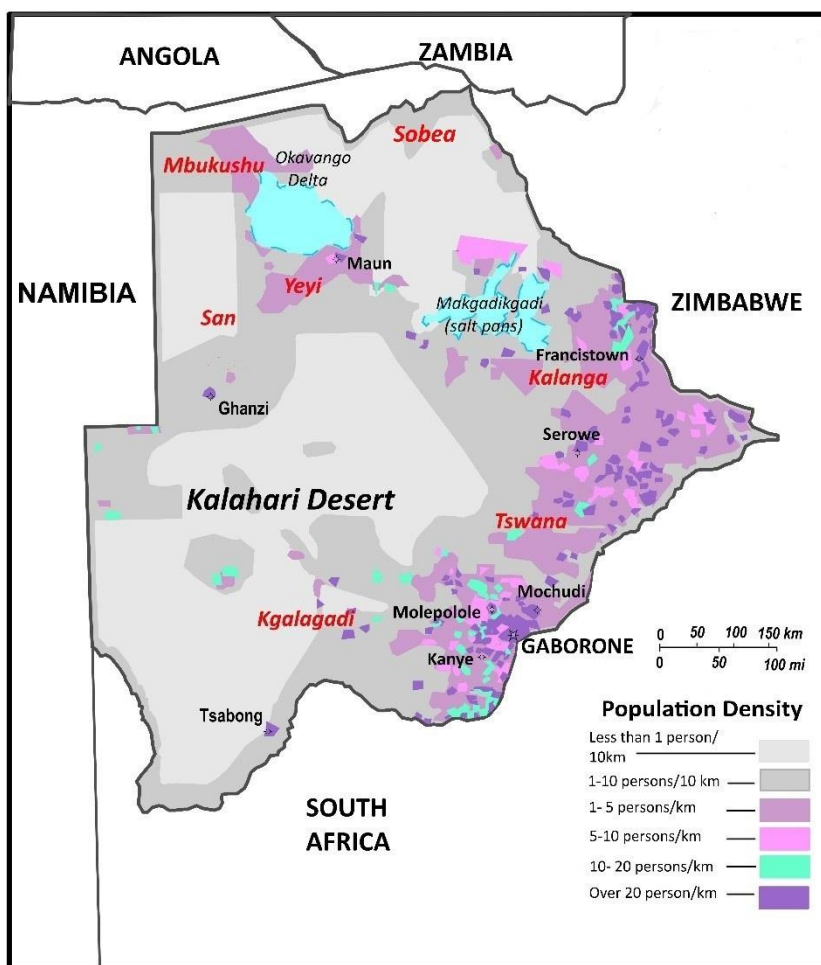


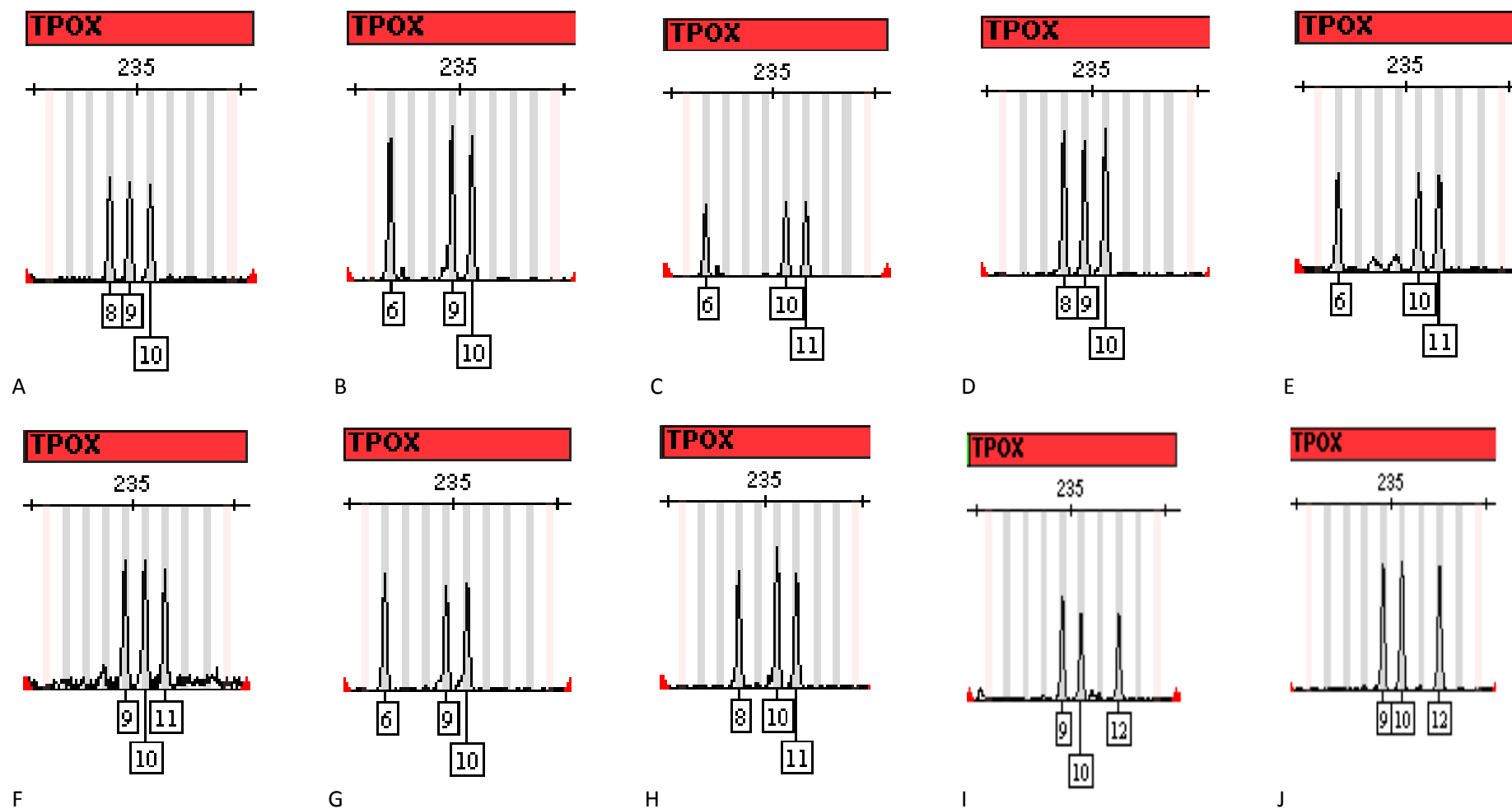
# Genetic variation and population structure of Botswana populations as identified with AmpFLSTR Identifiler short tandem repeat (STR) loci

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## Supplementary Information



**Supplementary Figure S1** Representation of the geographic locations of the different ethno-linguistic language groups used in the study. The Niger-Congo Bantoid branch (Bantu) being: Central S-Bantu-Kgalagadi, Tswana and Kalanga; Central K-Bantu- Yeyi; and Central R-Bantu- Mbukushu. The Khoisan (San) are also seen in the picture. Map adapted from [http://www.lib.utexas.edu/maps/africa/botswana-population\\_density-2005.png](http://www.lib.utexas.edu/maps/africa/botswana-population_density-2005.png) (Courtesy of the University of Texas Libraries, The University of Texas at Austin) using Surfer 9 (<http://www.ssg-surfer.com>).



Supplementary Figure S2 Electropherograms for samples exhibiting tri-allelic patterns for TPOX. Male samples are D and E.

**Supplementary Table S1** Rare “variant” and off-ladder variants observed in Botswana with the Identifiler kit. Off-ladder variants marked with an \*. Allele counts in this report are shown along with NIST number of records and publications for African populations along with their reported frequencies in brackets. See Supplementary Table S2 for allele frequencies reported in the study.

Allele	Current Study							NIST STR variant database ( <a href="http://www.cstl.nist.gov/strbase/var_tab.htm">http://www.cstl.nist.gov/strbase/var_tab.htm</a> )	African Published data		
	Bantu Central K	Bantu Central R	Bantu Central S	Khoisan Khoe-Kwadi	Khoisan Kx'a	Khoisan Tuu	Undeclared Khoisan		South African	Namibia	African
<b>D21S11 - tetranucleotide repeat ([TCTA] [TCTG])</b>											
24.3*			3	1				36 reports			
33.1*			10					86 reports	[2]Zulu (0.0102), [6] Bantu (0.0022 – 0.0120), [7] (Bantu 0.0039)		[4] Gabon (0.014), [5] Mozambique (0.004)
34.1*			6	3				21 reports	[1] Coloured (0.0100), [2]Xhosa (0.0042)		
35.1*	1	1	1	3	4			19 reports	[1] Coloured (0.0150), Bantu (0.0100), [2]Zulu (0.0051), Coloured (0.0090)		
36.1*			1					6 reports	[1] Coloured (0.0210)		
37.1*			1					1 report			
39			3		1			1 report	[1] Coloured (0.0050)		
<b>D7S820 - Tetranucleotide repeat (GATA)</b>											
5					3			1 report			
<b>FGA - complex Tetranucleotide repeat ([TTTC]3TTTTTCT[CTTT]nCTCC[TTCC]2)</b>											

16			2					8 reports			
18.2		2	8	1				10 reports	[2] Xhosa (0.0042)		[4] Gabon (0.005), [5] Mozambique (0.0292), [8] Mozambique (0.010), [9] Angolan (0.0146)
19.2			9	1				7 reports	[1] Bantu (0.0100), [2] Xhosa (0.0168), Zulu (0.0051)		[3] African (0.0050), [4] Gabon (0.005), [5] Mozambique (0.010), [8] Mozambique (0.0292), [9] Angolan (0.0010)
21.2			8	1				8 reports	[1] Coloured (0.0210), [2] Coloured (0.0045), Xhosa (0.0042), Zulu (0.0051)		
22.2	1		1					19 reports	[1] Coloured (0.0150) [2] Coloured (0.0045), Xhosa (0.0042), Zulu (0.0051)		[3] African (0.0100)
25.2				1				9 reports	[2] San (0.007)		
29.2			1					3 reports	[2] Xhosa (0.0042)		[5] Mozambique (0.003)
<b>CSF1PO- Tetranucleotide repeat (AGAT)</b>											
16						1		6 reports			
<b>D16S539- Tetranucleotide repeat (GATA)</b>											
17*					1			1 report			
<b>D19S433- Tetranucleotide repeat (AAGG)(AAAG)(AAGG)(TAGG)[AAGG]n</b>											
7		4	30	10	5		1	5 reports	[1] Coloured (0.0520), Bantu (0.0500), [2] Coloured (0.0225)	[1] Khoe (0.0190)	
<b>D18S51- Tetranucleotide repeat (GAAA)</b>											
15.2			7	10	2	1		22 reports	[1] Coloured (0.0310), Bantu (0.0100), [2] Xhosa (0.0126), Zulu (0.0051)	[1] Khoe (0.0380)	[3] African (0.0030), [8] Mozambique (0.0033), [9] Angolan (0.0021)
18.2			1					5 reports			[3] African (0.0050)

19.2	1		3					3 reports	[2] Zulu (0.0051)		[3] African (0.0030)
21.1			1					3 reports			
21.2			1					13 reports	[2] Zulu (0.0153)		[9] Angolan (0.0010)

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**Supplementary Table S2** Allele frequencies and Forensic summary statistics for each of the 15 Identifier STR loci found in the Botswana samples (n=990).

Allele	D8S1179	D21S11	D7S820	CSF1PO	D3S1358	TH01	D13S317	D16S539	D2S1338	D19S433	vWA	TPOX	D18S51	D5S818	FGA
5			0.002					0.005							
6				0.066		0.077						0.051		0.001	
7			0.007	0.053		0.309				0.025		0.026		0.001	
8			0.210	0.048		0.386	0.024	0.028				0.311		0.069	
9			0.136	0.048		0.169	0.009	0.186		0.002		0.274	0.002	0.035	
9,3						0.037									
10			0.308	0.233		0.020	0.050	0.146		0.018		0.107	0.011	0.045	
10,2													0.006		
11	0.027		0.218	0.197		0.002	0.283	0.356		0.057	0.005	0.226	0.001	0.193	
12	0.120		0.102	0.291	0.002		0.408	0.170		0.122		0.004	0.037	0.370	
12,2										0.034					
13	0.192		0.019	0.054			0.157	0.101		0.299	0.010		0.042	0.270	
13,2										0.053			0.003		
14	0.343			0.008	0.102		0.066	0.007		0.206	0.075		0.040	0.015	
14,2										0.067			0.004		
15	0.218			0.001	0.335		0.003	0.002	0.018	0.053	0.191		0.122	0.002	
15,2										0.031			0.011		
16	0.090			0.001	0.330				0.063	0.021	0.264		0.176		0.001
16,2										0.012					
17	0.009				0.190			0.001	0.068	0.001	0.203		0.164		0.001
18	0.001				0.041				0.072	0.001	0.147		0.134		0.010
18,2													0.001		0.006
19					0.001				0.160		0.077		0.121		0.054
19,2													0.003		0.005
20									0.088		0.020		0.068		0.057

21									0.175		0.005		0.026		0.091
21,1													0.001		
21,2													0.001		0.005
22									0.158		0.002		0.017		0.169
22,2															0.002
23									0.076		0.001		0.005		0.181
24									0.045				0.001		0.185
24,2		0.002							0.001						
24,3		0.002													
25		0.001							0.052				0.004		0.102
25,2															0.001
26		0.003							0.020				0.002		0.068
27		0.064							0.005				0.001		0.019
28		0.231							0.001						0.016
29		0.132													0.009
29,2		0.001													0.001
30		0.138													0.002
30,2		0.008													0.004
31		0.077													
31,2		0.072													0.005
32		0.020													
32,2		0.082													0.002
33		0.017													
33,1		0.006													
33,2		0.034													
33,3		0.001													
34		0.024													
34,1		0.005													
34,2		0.008													

35		0.034														
35,1		0.006														
35,2		0.002														
36		0.011														
36,1		0.001														
37		0.012														
37,1		0.001														
38		0.008														
39		0.002														
42																0.001
42,2																0.007
43,2																0.002



**Supplementary Table S3** Genetic diversity estimators for the Botswana populations: overall (A), according to the Bantu (B-D) and Khoisan (E-G) ethno-linguistic groups, and according to districts (H-P), The Observed heterozygosity (Hobs), expected heterozygosity (Hexp) and probability of Hardy-Weinberg equilibrium (HWE *P*). Hobs= observed heterozygosity. Significant *p* values ( $P < 0,0033$ ) are in bold. Values out of HWE that show Heterozygote deficiency indicated by an asterisk (\*).

	A			B			C			D		
	Overall population (n=990)			Bantu Central K (n=43)			Bantu Central R (n=58)			Bantu Central S (n=646)		
Locus	Hobs	Hexp	HWE <i>P</i>	Hobs	Hexp	HWE <i>P</i>	Hobs	Hexp	HWE <i>P</i>	Hobs	Hexp	HWE <i>P</i>
<b>D8S1179</b>	0.7727	0.7738	0.0967	0.7441	0.7447	0.2290	0.75862	0.7658	0.9885	0.7879	0.7673	0.0688
<b>D21S11</b>	0.8549	0.8854	0.1890	0.8837	0.8738	0.1684	0.81034	0.8574	0.6888	0.8529	0.8717	0.6748
<b>D7S820</b>	0.7779	0.7870	0.1144	0.7441	0.7647	0.7829	0.7069	0.7643	0.7171	0.7832	0.7768	0.3912
<b>CSF1PO</b>	0.7804	0.8076	<b>0.0007*</b>	0.7209	0.7865	0.5515	0.7931	0.8115	0.4016	0.7724	0.7959	0.6330
<b>D3S1358</b>	0.7201	0.7293	0.8766	0.6976	0.7398	0.2769	0.7069	0.7117	0.7010	0.7275	0.7319	0.4994
<b>TH01</b>	0.7111	0.7188	0.2138	0.6976	0.7275	0.2921	0.6724	0.7163	0.1131	0.7182	0.7119	0.1035
<b>D13S317</b>	0.7175	0.7264	0.0973	0.5116	0.5641	0.5553	0.5000	0.6699	<b>0.0014</b>	0.7414	0.7289	0.6245
<b>D16S539</b>	0.7522	0.7718	0.3057	0.7209	0.7326	0.2737	0.77586	0.7823	0.6910	0.7631	0.7616	0.9347
<b>D2S1338</b>	0.8575	0.8859	0.1093	0.8604	0.8640	0.6568	0.91379	0.9039	0.7171	0.8498	0.8769	0.2828
<b>D19S433</b>	0.8023	0.8399	<b>0.0006*</b>	0.7907	0.829	0.7670	0.91379	0.8523	0.0062	0.8235	0.8311	0.2338
<b>vWA</b>	0.8048	0.8209	0.7287	0.8837	0.8459	0.9779	0.77586	0.8231	0.4979	0.8003	0.8178	0.2902
<b>TPOX</b>	0.7509	0.7591	0.1434	0.8139	0.7734	0.3943	0.77586	0.7733	0.1884	0.7631	0.7654	0.9037
<b>D18S51</b>	0.8716	0.8831	0.7349	0.8837	0.8730	0.1972	0.7931	0.8729	0.2608	0.8823	0.8761	0.9091
<b>D5S818</b>	0.7471	0.7479	0.0076	0.7209	0.7507	0.9813	0.58621	0.6735	0.0253	0.7445	0.7399	0.0800
<b>FGA</b>	0.8446	0.8752	0.1581	0.8604	0.8741	0.6342	0.8965	0.8694	0.6888	0.8513	0.8731	0.0695

E

F

G

	Khoisan Khoe-kwadi (n=133)			Khoisan Kx'a (n=58)			Khoisan Tuu (n=32)		
Locus	Hobs	Hexp	HWE <i>P</i>	Hobs	Hexp	HWE <i>P</i>	Hobs	Hexp	HWE <i>P</i>
<b>D8S1179</b>	0.7519	0.7976	0.7088	0.7586	0.8093	0.6632	0.8438	0.7813	0.8805
<b>D21S11</b>	0.9023	0.9071	0.4263	0.9483	0.9041	0.2102	0.8438	0.8909	0.0652
<b>D7S820</b>	0.7820	0.8135	0.0427	0.8103	0.7939	0.1783	0.7188	0.7589	0.0570
<b>CSF1PO</b>	0.8045	0.8075	0.7385	0.7414	0.8000	0.1201	0.7813	0.8036	0.1402
<b>D3S1358</b>	0.7143	0.7212	0.7289	0.7241	0.7199	0.2188	0.6563	0.5947	0.8124
<b>TH01</b>	0.7368	0.7313	0.6719	0.6897	0.7003	0.2799	0.6875	0.6319	0.5724
<b>D13S317</b>	0.6993	0.7271	0.0562	0.7414	0.6891	0.4463	0.6250	0.7574	0.1643
<b>D16S539</b>	0.7293	0.8059	0.3992	0.7414	0.7924	0.5114	0.6250	0.8155	0.0584
<b>D2S1338</b>	0.8496	0.8861	0.3649	0.9655	0.9039	0.4063	0.9063	0.8958	0.0517
<b>D19S433</b>	0.8120	0.8593	0.1875	0.6379	0.7892	0.0609	0.5000	0.8115	<b>0.0013*</b>
<b>vWA</b>	0.8120	0.8206	0.9994	0.6724	0.7712	0.0885	0.6875	0.7753	0.7238
<b>TPOX</b>	0.6842	0.7053	0.2157	0.8103	0.7490	0.0490	0.7500	0.6701	<b>0.0018</b>
<b>D18S51</b>	0.8421	0.8946	0.5419	0.9138	0.8903	0.9802	0.9063	0.9023	0.9808
<b>D5S818</b>	0.7444	0.7755	0.0810	0.7241	0.7555	0.0406	0.7813	0.7173	0.6173
<b>FGA</b>	0.8196	0.8762	0.1149	0.8966	0.8906	0.0275	0.8750	0.8487	0.1045

	H			I			J			K			L		
	Central District (n=264)			Ghanzi District (n=168)			Kgalagadi District (n=20)			Kgatleng District (n=21)			Kweneng District (n=106)		
Locus	Hobs	Hexp	HWE <i>P</i>	Hobs	Hexp	HWE <i>P</i>	Hobs	Hexp	HWE <i>P</i>	Hobs	Hexp	HWE <i>P</i>	Hobs	Hexp	HWE <i>P</i>
<b>D8S1179</b>	0.7917	0.7563	0.1558	0.7738	0.7791	0.8184	0.8500	0.7923	0.9916	0.8095	0.8060	0.4429	0.8208	0.7786	0.2719
<b>D21S11</b>	0.8296	0.8677	0.1074	0.8869	0.8984	0.0375	0.8500	0.8885	0.7854	0.8571	0.8966	0.3985	0.8113	0.8683	0.6147
<b>D7S820</b>	0.7576	0.7711	0.2422	0.7321	0.8109	0.0318	0.9000	0.8064	0.1632	0.9048	0.7979	0.1585	0.8113	0.7752	0.7307
<b>CSF1PO</b>	0.7765	0.7898	0.7569	0.8214	0.8252	0.0023	0.6500	0.8013	0.0076	0.8571	0.8211	0.3982	0.8491	0.8068	0.4976
<b>D3S1358</b>	0.7689	0.7403	0.6279	0.6667	0.6784	0.5516	0.7500	0.7397	0.6453	0.7619	0.7549	0.6121	0.7359	0.7089	0.5400
<b>TH01</b>	0.7349	0.7014	0.1629	0.7024	0.7183	0.5698	0.6000	0.6180	0.8197	0.7143	0.6771	0.9085	0.6415	0.7206	0.2787
<b>D13S317</b>	0.7424	0.7415	0.4299	0.6786	0.6992	0.3339	0.7000	0.7769	0.2006	0.6191	0.6481	0.3524	0.7925	0.7314	0.4064
<b>D16S539</b>	0.7538	0.7641	0.4543	0.7202	0.8055	0.0129	0.7500	0.7756	0.7256	0.7143	0.7456	0.2767	0.7642	0.7359	0.3399
<b>D2S1338</b>	0.8182	0.8784	0.3588	0.8810	0.8917	0.2036	1.0000	0.8808	0.8813	0.8571	0.8862	0.3985	0.8868	0.8813	0.0073
<b>D19S433</b>	0.8220	0.8314	0.6908	0.7262	0.8402	<b>0.0007*</b>	0.7500	0.8397	0.3050	0.8571	0.9001	0.1621	0.7642	0.8154	0.5287
<b>vWA</b>	0.8447	0.8279	0.8909	0.8095	0.8010	0.9790	0.8500	0.8295	0.1221	0.8571	0.8420	0.5947	0.7642	0.8170	0.0466
<b>TPOX</b>	0.7424	0.7720	0.7687	0.7202	0.6952	0.0410	0.8000	0.7295	0.3789	0.6191	0.7096	0.1475	0.7736	0.7473	0.1849
<b>D18S51</b>	0.8864	0.8748	0.5201	0.8571	0.9000	0.4641	0.8500	0.9051	0.2641	0.9524	0.8432	0.7488	0.8868	0.8774	0.6548
<b>D5S818</b>	0.7500	0.7294	0.7622	0.7500	0.7824	0.0486	0.6000	0.7013	0.0730	0.7619	0.7689	0.9127	0.7453	0.7290	0.5017
<b>FGA</b>	0.8485	0.8741	0.0710	0.8155	0.8690	0.0018	0.8500	0.8782	0.4449	0.9048	0.9013	0.7433	0.8679	0.8789	0.1010

	M			N			O			P		
	Northeast District (n=45)			Northwest District (n=260)			Southeast District (n=32)			Southern District (n=74)		
Locus	Hobs	Hexp	HWE <i>P</i>	Hobs	Hexp	HWE <i>P</i>	Hobs	Hexp	HWE <i>P</i>	Hobs	Hexp	HWE <i>P</i>
<b>D8S1179</b>	0.7111	0.7738	0.5358	0.7462	0.7845	0.2919	0.8784	0.7550	0.2963	0.7973	0.7793	0.2805
<b>D21S11</b>	0.8444	0.8652	0.6275	0.9039	0.8958	0.2975	0.9063	0.8720	0.6853	0.8784	0.8590	0.6166
<b>D7S820</b>	0.8444	0.7758	0.0172	0.7846	0.7758	0.5823	0.7500	0.7674	0.9807	0.7838	0.7917	0.6176
<b>CSF1PO</b>	0.7556	0.7800	0.9794	0.7269	0.7942	0.1754	0.7500	0.8199	0.5721	0.7297	0.8121	0.1355
<b>D3S1358</b>	0.6222	0.7239	0.5922	0.7231	0.7358	0.5184	0.6875	0.7282	0.0856	0.6757	0.7411	0.1860
<b>TH01</b>	0.7333	0.7086	0.9956	0.7077	0.7258	0.0801	0.6563	0.7406	0.5800	0.8514	0.7289	0.0773
<b>D13S317</b>	0.6444	0.7316	0.0586	0.6615	0.6989	0.0142	0.6875	0.7064	0.4648	0.7568	0.7199	0.7431
<b>D16S539</b>	0.7111	0.7328	0.9665	0.7731	0.7731	0.1642	0.8125	0.7818	0.4018	0.7162	0.7911	0.2414
<b>D2S1338</b>	0.8444	0.8674	0.1625	0.8846	0.8904	0.8162	0.8438	0.8557	0.7053	0.8514	0.8689	0.4514
<b>D19S433</b>	0.8444	0.8315	0.5433	0.8346	0.8444	0.5295	0.8750	0.8581	0.5565	0.8108	0.8038	0.2050
<b>vWA</b>	0.8667	0.8162	0.9352	0.7269	0.8173	0.0619	0.8438	0.7986	0.9537	0.7162	0.8014	0.1353
<b>TPOX</b>	0.7333	0.7718	0.7762	0.7885	0.7768	0.0456	0.6563	0.7634	0.6332	0.7973	0.7570	0.7061
<b>D18S51</b>	0.8000	0.8582	0.2877	0.8654	0.8841	0.7004	0.8438	0.8765	0.0024	0.9324	0.8865	0.4182
<b>D5S818</b>	0.7556	0.7636	0.3835	0.7077	0.7258	0.3855	0.7813	0.7361	0.4285	0.6622	0.7605	0.1607
<b>FGA</b>	0.7556	0.8527	0.1670	0.8692	0.8751	0.4001	0.7813	0.8447	0.6009	0.9324	0.8800	0.4232

**Supplementary Table S4** Maximum Likelihood (ML) Silent allele frequencies for the Botswana populations: overall (A). according to the Bantu (B-D) and Khoisan (E-G) ethno-linguistic groups. and according to districts (H-P).

	A	B	C	D	E	F	G
	Overall population (n=990)	Bantu Central K (n=43)	Bantu Central R (n=58)	Bantu Central S (n=646)	Khoisan Khoe-kwadi (n=133)	Khoisan Kx'a (n=58)	Khoisan Tuu (n=32)
<b>Locus</b>							
<b>D8S1179</b>	0.006	0	0	0.002	0.027	0.017	0
<b>D21S11</b>	0.012	0	0.031	0.012	0	0	0.025
<b>D7S820</b>	0.006	0	0.023	0	0.009	0	0.044
<b>CSF1PO</b>	0.014	0.022	0	0.005	0	0.024	0.031
<b>D3S1358</b>	0.002	0.023	0	0	0	0.015	0
<b>TH01</b>	0	0.02	0.013	0	0	0	0
<b>D13S317</b>	0.01	0.008	0.083	0	0.022	0	0.083
<b>D16S539</b>	0.009	0	0	0	0.032	0.012	0.09
<b>D2S1338</b>	0.013	0	0	0.017	0.017	0	0
<b>D19S433</b>	0.024	0.027	0	0.006	0.034	0.078	0.17
<b>vWA</b>	0.01	0	0.019	0.001	0	0.052	0.038
<b>TPOX</b>	0.005	0	0	0	0.015	0	0.009
<b>D18S51</b>	0.009	0.013	0.045	0	0.018	0	0
<b>D5S818</b>	0.013	0	0.031	0.004	0.018	0.023	0
<b>FGA</b>	0.008	0	0	0.01	0.019	0	0.004

H            I            J            K            L            M            N            O            P

Locus	Central District (n=264)	Ghanzi District (n=168)	Kgalagadi District (n=20)	Kgatleng District (n=21)	Kweneng District (n=106)	Northeast District (n=45)	Northwest District (n=260)	Southeast District (n=32)	Southern District (74)
D8S1179	0	0.001	0	0	0	0.041	0.021	0.02	0
D21S11	0.023	0.004	0	0	0.037	0	0.001	0	0
D7S820	0.004	0.036	0	0	0	0.012	0	0	0
CSF1PO	0.007	0.011	0.051	0	0	0	0.019	0.01	0.009
D3S1358	0	0	0	0	0	0.042	0.009	0.026	0.032
TH01	0	0.013	0	0	0.02	0	0	0.014	0
D13S317	0	0.016	0.018	0	0	0.039	0.024	0	0
D16S539	0	0.041	0.017	0.02	0	0.009	0	0	0.044
D2S1338	0.037	0.003	0	0.021	0.007	0.004	0	0	0
D19S433	0.012	0.07	0.032	0	0.017	0	0.005	0	0
vWA	0	0	0	0.006	0.024	0	0.04	0	0.045
TPOX	0.012	0	0	0.063	0	0.014	0	0.038	0
D18S51	0	0.013	0.036	0	0.003	0.022	0.015	0.004	0
D5S818	0	0.024	0.023	0	0	0	0.014	0	0.048
FGA	0.01	0.018	0.02	0	0.003	0.037	0	0	0

**Supplementary Table S5** Pairwise genetic distance ( $F_{ST}$ ) (below diagonal) and P-values (above diagonal) of the different Bantu and Khoisan ethno-linguistic groups in Botswana. Significant values ( $P < 0.003$ ) are indicated by an asterisk \*.

	<b>Central K</b>	<b>Central R</b>	<b>Central S</b>	<b>Khoe</b>	<b>Kx'a</b>	<b>Tuu</b>
<b>Central K</b>	-	0.21226	0.00158	0.00000	0.00000	0.00000
<b>Central R</b>	0.00174	-	0.00020	0.00000	0.00000	0.00000
<b>Central S</b>	0.00402*	0.00377*	-	0.00000	0.00000	0.00000
<b>Khoe</b>	0.01363*	0.00921*	0.01024*	-	0.00000	0.00000
<b>Kx'a</b>	0.02312*	0.01905*	0.02026*	0.00656*	-	0.00000
<b>Tuu</b>	0.03069*	0.02819*	0.03009*	0.01622*	0.02183*	-

**Supplementary Table S6** Pairwise genetic distance ( $F_{ST}$ ) (below diagonal) and P-values (above diagonal) of the nine administrative districts in Botswana. Significant values ( $P < 0.0003$ ) are indicated by an asterisk \*.

	Central	Ghanzi	Kgalagadi	Kgatleng	Kweneng	Northeast	Northwest	Southeast	Southern
Central	-	0.00000	0.19444	0.78071	0.11118	0.88457	0.00000	0.18384	0.09395
Ghanzi	0.01456*	-	0.02792	0.00050	0.00000	0.00000	0.00000	0.00000	0.00000
Kgalagadi	0.00208	0.00571	-	0.75775	0.12771	0.23681	0.35393	0.44897	0.30344
Kgatleng	-0.00171	0.01017	-0.00298	-	0.65855	0.78052	0.50045	0.43996	0.57519
Kweneng	0.00071	0.01792*	0.00288	-0.00128	-	0.23235	0.00000	0.10593	0.06881
Northeast	-0.00125	0.01623*	0.00276	-0.00217	0.00101	-	0.50718	0.68290	0.07663
Northwest	0.00269*	0.01238*	0.00107	0.00011	0.00589*	0.00005	-	0.02505	0.00000
Southeast	0.00141	0.01190*	0.00058	0.00044	0.00231	-0.00091	0.00349	-	0.50431
Southern	0.00104	0.01226*	0.00144	-0.00061	0.00161	0.00254	0.00526*	-0.00004	-



**Supplementary Table S7** Forensic statistics metric for each locus for the Botswana populations: the overall population (A), according to the Bantu (B-D) and Khoisan (E-G) ethno-linguistic groups, and according to districts (H-P). Discrimination capacity (DC), match probability (MP), combined random match probability, polymorphic information capacity (PIC), typical paternity index (TPI), combined paternity index, power of exclusion (PE), and combined power of exclusion (CPE).

A

	<b>Overall population (n=990)</b>				
<b>Locus</b>	PE	MP	PD	PIC	TPI
<b>D8S1179</b>	0.558	0.086	0.914	0.742	2.2
<b>D21S11</b>	0.585	0.053	0.947	0.805	2.4
<b>D7S820</b>	0.558	0.079	0.921	0.752	2.2
<b>CSF1PO</b>	0.549	0.061	0.939	0.782	2.2
<b>D3S1358</b>	0.459	0.119	0.881	0.684	1.7
<b>TH01</b>	0.452	0.124	0.876	0.673	1.7
<b>D13S317</b>	0.439	0.119	0.881	0.678	1.7
<b>D16S539</b>	0.511	0.079	0.921	0.747	2.0
<b>D2S1338</b>	0.72	0.024	0.976	0.876	3.6
<b>D19S433</b>	0.492	0.112	0.888	0.70	1.9
<b>vWA</b>	0.582	0.056	0.944	0.794	2.3
<b>TPOX</b>	0.516	0.097	0.903	0.723	2.0
<b>D18S51</b>	0.744	0.025	0.975	0.873	3.9
<b>D5S818</b>	0.477	0.107	0.893	0.705	1.8
<b>FGA</b>	0.698	0.029	0.971	0.862	3.3
<b>Combined</b>	0.999996541	$6.28 \times 10^{-19}$			

	<b>B</b>					<b>C</b>					<b>D</b>				
	<b>Bantu Central K (n=43)</b>					<b>Bantu Central R (n=58)</b>					<b>Bantu Central S (n=646)</b>				
<b>Locus</b>	<b>PE</b>	<b>MP</b>	<b>PD</b>	<b>PIC</b>	<b>TPI</b>	<b>PE</b>	<b>MP</b>	<b>PD</b>	<b>PIC</b>	<b>TPI</b>	<b>PE</b>	<b>MP</b>	<b>PD</b>	<b>PIC</b>	<b>TPI</b>
<b>D8S1179</b>	0.499	0.131	0.868	0.700	1.9	0.524	0.091	0.909	0.724	2.0	0.585	0.0934	0.906	0.732	2.4
<b>D21S11</b>	0.662	0.104	0.895	0.849	3.0	0.520	0.074	0.925	0.834	2.0	0.607	0.0568	0.943	0.7925	2.5
<b>D7S820</b>	0.163	0.287	0.712	0.718	0.9	0.240	0.226	0.773	0.718	1.1	0.387	0.2179	0.782	0.742	1.5
<b>CSF1PO</b>	0.336	0.142	0.857	0.745	1.3	0.391	0.136	0.863	0.779	1.5	0.382	0.1405	0.859	0.767	1.5
<b>D3S1358</b>	0.424	0.127	0.872	0.685	1.6	0.439	0.143	0.856	0.651	1.7	0.477	0.1215	0.878	0.685	1.8
<b>TH01</b>	0.424	0.137	0.862	0.671	1.6	0.421	0.143	0.856	0.656	2.2	0.457	0.1335	0.866	0.661	1.7
<b>D13S317</b>	0.197	0.254	0.745	0.493	1.0	0.179	0.177	0.822	0.608	0.9	0.468	0.1295	0.870	0.685	1.8
<b>D16S539</b>	0.336	0.214	0.785	0.683	1.3	0.450	0.172	0.827	0.740	1.7	0.404	0.1525	0.847	0.729	1.5
<b>D2S1338</b>	0.715	0.051	0.948	0.839	3.5	0.823	0.033	0.966	0.887	5.8	0.696	0.0283	0.971	0.864	3.3
<b>D19S433</b>	0.496	0.094	0.905	0.799	1.9	0.651	0.210	0.789	0.830	2.9	0.485	0.1116	0.888	0.697	1.8
<b>vWA</b>	0.7623	0.056	0.943	0.815	4.3	0.555	0.066	0.933	0.791	2.2	0.607	0.0584	0.941	0.793	2.5
<b>TPOX</b>	0.599	0.680	0.320	0.730	2.5	0.240	0.308	0.691	0.728	1.1	0.126	0.3990	0.601	0.726	0.8
<b>D18S51</b>	0.795	0.071	0.928	0.849	5.0	0.585	0.045	0.954	0.852	2.4	0.747	0.0312	0.968	0.863	4.0
<b>D5S818</b>	0.363	0.162	0.837	0.704	1.4	0.196	0.225	0.774	0.610	1.0	0.399	0.1568	0.843	0.697	1.5
<b>FGA</b>	0.702	0.050	0.949	0.849	3.4	0.772	0.053	0.946	0.848	4.5	0.680	0.0346	0.965	0.860	3.1
<b>Combined</b>	0.999991	5.91 x10 <sup>-14</sup>				0.999976	1.05x10 <sup>-14</sup>				0.999986	6.28 x10 <sup>-16</sup>			

E

F

G

Locus	Khoisan Khoe-kwadi (n=133)					Khoisan Kx'a (n=58)					Khoisan Tuu (n=32)				
	PE	MP	PD	PIC	TPI	PE	MP	PD	PIC	TPI	PE	MP	PD	PIC	TPI
D8S1179	0.5130	0.074	0.925	0.764	2.0	0.524	0.071	0.928	0.774	2.0	0.682	0.103	0.896	0.733	3.2
D21S11	0.6531	0.041	0.925	0.896	2.9	0.898	0.08	0.920	0.887	10	0.487	0.113	0.886	0.866	1.9
D7S820	0.4557	0.200	0.799	0.783	1.7	0.543	0.242	0.757	0.756	2.1	0.378	0.283	0.716	0.708	1.5
CSF1PO	0.3294	0.182	0.817	0.782	1.3	0.209	0.160	0.839	0.763	1.05	0.330	0.157	0.842	0.764	1.3
D3S1358	0.2409	0.308	0.691	0.675	1.1	0.466	0.152	0.847	0.667	1.8	0.363	0.212	0.787	0.552	1.4
TH01	0.4875	0.118	0.881	0.687	1.9	0.412	0.145	0.854	0.655	1.6	0.409	0.197	0.802	0.581	1.6
D13S317	0.4174	0.133	0.866	0.686	1.6	0.418	0.197	0.802	0.651	1.6	0.226	0.173	0.826	0.703	1.0
D16S539	0.3662	0.124	0.875	0.775	1.4	0.409	0.156	0.843	0.754	1.6	0.165	0.163	0.836	0.775	0.9
D2S1338	0.6941	0.030	0.969	0.871	3.3	0.930	0.037	0.962	0.887	14.5	0.808	0.064	0.935	0.870	5.3
D19S433	0.5214	0.103	0.896	0.843	2.0	0.232	0.142	0.857	0.763	1.1	0.202	0.094	0.905	0.769	1.0
vWA	0.6216	0.057	0.942	0.793	2.6	0.386	0.101	0.898	0.728	1.5	0.409	0.099	0.900	0.725	1.6
TPOX	0.4507	0.591	0.408	0.653	1.7	0.187	0.500	0.500	0.707	1	0	0.555	0.444	0.611	0.5
D18S51	0.6490	0.027	0.972	0.882	2.8	0.817	0.036	0.963	0.871	5.6	0.795	0.04	0.960	0.880	5
D5S818	0.4202	0.156	0.843	0.742	1.6	0.407	0.158	0.841	0.708	1.5	0.477	0.210	0.789	0.658	1.8
FGA	0.6051	0.038	0.961	0.861	2.5	0.754	0.062	0.937	0.872	4.2	0.736	0.0884	0.911	0.815	3.8
Combined	0.999978	$1.21 \times 10^{-15}$				0.999999	$1.52 \times 10^{-14}$				0.999958	$6.28 \times 10^{-19}$			

H

I

J

	Central District (n=264)					Ghanzi District (n=168)					Kgalagadi District (n=20)				
Locus	PE	MP	PD	PIC	TPI	PE	MP	PD	PIC	TPI	PE	MP	PD	PIC	TPI
<b>D8S1179</b>	0.584	0.103	0.897	0.719	2.4	0.551	0.086	0.914	0.742	2.2	0.695	0.105	0.895	0.737	3.3
<b>D21S11</b>	0.609	0.066	0.934	0.784	2.5	0.769	0.028	0.972	0.887	4.4	0.695	0.060	0.940	0.853	3.3
<b>D7S820</b>	0.523	0.088	0.912	0.735	2.1	0.480	0.066	0.934	0.781	1.9	0.795	0.140	0.860	0.752	5.0
<b>CSF1PO</b>	0.556	0.076	0.924	0.758	2.2	0.639	0.062	0.938	0.800	2.8	0.355	0.135	0.865	0.751	1.4
<b>D3S1358</b>	0.543	0.120	0.880	0.695	2.2	0.379	0.156	0.844	0.632	1.5	0.510	0.140	0.860	0.673	2.0
<b>TH01</b>	0.484	0.151	0.849	0.646	1.9	0.432	0.120	0.880	0.677	1.7	0.291	0.179	0.821	0.557	1.3
<b>D13S317</b>	0.497	0.108	0.892	0.699	1.9	0.396	0.138	0.862	0.656	1.6	0.428	0.130	0.870	0.720	1.7
<b>D16S539</b>	0.906	0.090	0.910	0.730	1.3	0.460	0.069	0.931	0.774	1.8	0.510	0.110	0.890	0.725	2.0
<b>D2S1338</b>	0.633	0.028	0.972	0.865	2.8	0.757	0.028	0.972	0.879	4.2	1.000	0.075	0.925	0.845	0
<b>D19S433</b>	0.640	0.050	0.950	0.811	2.8	0.470	0.049	0.951	0.820	1.8	0.510	0.085	0.915	0.798	2.0
<b>vWA</b>	0.684	0.056	0.944	0.803	3.2	0.617	0.071	0.929	0.770	2.6	0.695	0.125	0.875	0.782	3.3
<b>TPOX</b>	0.497	0.089	0.911	0.733	1.9	0.460	0.166	0.834	0.643	1.8	0.599	0.170	0.830	0.671	2.5
<b>D18S51</b>	0.768	0.033	0.967	0.860	4.4	0.709	0.022	0.978	0.889	3.5	0.695	0.065	0.935	0.872	3.3
<b>D5S818</b>	0.510	0.124	0.876	0.684	2.	0.510	0.088	0.912	0.748	2.	0.291	0.185	0.815	0.625	1.3
<b>FGA</b>	0.692	0.032	0.968	0.860	3.3	0.628	0.039	0.961	0.853	2.7	0.695	0.075	0.925	0.840	3.3
<b>Combined</b>	0.999999563	7.27x10 <sup>-18</sup>				0.99999684	2.14x10 <sup>-18</sup>				1.000000	5.21x10 <sup>-15</sup>			

K

L

M

	Kgatleng District (n=21)					Kweneng District (n=106)					Northeast District (n=45)				
Locus	PE	MP	PD	PIC	TPI	PE	MP	PD	PIC	TPI	PE	MP	PD	PIC	TPI
<b>D8S1179</b>	0.617	0.116	0.884	0.754	2.6	0.638	0.098	0.902	0.740	2.8	0.446	0.094	0.906	0.730	1.7
<b>D21S11</b>	0.709	0.061	0.939	0.864	3.5	0.620	0.035	0.965	0.851	2.7	0.684	0.049	0.951	0.842	3.2
<b>D7S820</b>	0.805	0.152	0.848	0.746	5.3	0.620	0.094	0.906	0.737	2.7	0.684	0.136	0.864	0.730	3.2
<b>CSF1PO</b>	0.777	0.135	0.865	0.775	3.3	0.693	0.081	0.919	0.776	3.3	0.519	0.083	0.917	0.738	2.0
<b>D3S1358</b>	0.530	0.143	0.857	0.691	2.1	0.486	0.147	0.853	0.654	1.9	0.318	0.123	0.877	0.665	1.3
<b>TH01</b>	0.451	0.179	0.821	0.601	1.8	0.344	0.121	0.879	0.669	1.4	0.482	0.144	0.856	0.647	1.9
<b>D13S317</b>	0.314	0.202	0.798	0.580	1.3	0.585	0.126	0.874	0.684	2.4	0.348	0.128	0.872	0.681	1.4
<b>D16S539</b>	0.451	0.138	0.862	0.697	1.8	0.534	0.107	0.893	0.701	2.1	0.446	0.119	0.881	0.688	1.7
<b>D2S1338</b>	0.709	0.075	0.925	0.851	3.5	0.769	0.040	0.960	0.865	4.4	0.684	0.056	0.944	0.842	3.2
<b>D19S433</b>	0.709	0.070	0.930	0.868	3.5	0.534	0.059	0.941	0.790	2.1	0.684	0.064	0.936	0.802	3.2
<b>vWA</b>	0.709	0.088	0.912	0.798	3.5	0.534	0.073	0.927	0.789	2.1	0.728	0.075	0.925	0.780	3.8
<b>TPOX</b>	0.314	0.170	0.830	0.645	1.3	0.551	0.124	0.876	0.700	2.2	0.096	0.095	0.905	0.725	4.4
<b>D18S51</b>	0.903	0.093	0.907	0.800	10.5	0.769	0.037	0.963	0.860	4.4	0.599	0.053	0.947	0.832	2.5
<b>D5S818</b>	0.530	0.111	0.889	0.714	2.1	0.502	0.131	0.869	0.680	2.0	0.519	0.105	0.895	0.716	2.0
<b>FGA</b>	0.805	0.061	0.939	0.868	5.3	0.730	0.037	0.963	0.862	3.8	0.519	0.057	0.943	0.826	2.0
<b>Combined</b>	0.99999929	5.12x10 <sup>-15</sup>				0.99999927	5.12x10 <sup>-15</sup>				0.99999227	1.14x10 <sup>-16</sup>			

N

O

P

Locus	Northwest District (n=260)					Southeast District (n=32)					Southern District (74)				
	PE	MP	PD	PIC	TPI	PE	MP	PD	PIC	TPI	PE	MP	PD	PIC	TPI
<b>D8S1179</b>	0.503	0.079	0.921	0.752	2.0	0.565	0.129	0.871	0.705	2.3	0.594	0.098	0.902	0.740	2.5
<b>D21S11</b>	0.803	0.024	0.976	0.885	5.2	0.808	0.057	0.943	0.844	5.3	0.751	0.042	0.958	0.839	4.1
<b>D7S820</b>	0.571	0.086	0.914	0.739	2.3	0.510	0.098	0.902	0.716	2.	0.569	0.084	0.916	0.753	2.3
<b>CSF1PO</b>	0.471	0.068	0.932	0.763	1.8	0.510	0.074	0.926	0.782	2.0	0.476	0.068	0.932	0.782	1.9
<b>D3S1358</b>	0.465	0.119	0.881	0.688	1.8	0.409	0.162	0.838	0.669	1.6	0.392	0.123	0.877	0.690	1.5
<b>TH01</b>	0.440	0.122	0.878	0.678	1.7	0.364	0.117	0.883	0.688	1.5	0.698	0.165	0.835	0.677	3.4
<b>D13S317</b>	0.371	0.136	0.864	0.653	1.5	0.409	0.154	0.846	0.654	1.6	0.521	0.140	0.860	0.664	2.1
<b>D16S539</b>	0.550	0.088	0.912	0.739	2.2	0.622	0.119	0.881	0.734	2.7	0.454	0.088	0.912	0.753	1.8
<b>D2S1338</b>	0.764	0.024	0.976	0.879	4.3	0.683	0.059	0.941	0.825	3.2	0.698	0.044	0.956	0.849	3.4
<b>D19S433</b>	0.665	0.043	0.957	0.827	3.0	0.745	0.066	0.934	0.827	4.0	0.510	0.112	0.888	0.707	2.0
<b>vWA</b>	0.471	0.057	0.943	0.791	1.8	0.683	0.096	0.904	0.756	3.2	0.454	0.078	0.922	0.767	1.8
<b>TPOX</b>	0.578	0.091	0.909	0.740	2.4	0.364	0.102	0.898	0.709	1.5	0.594	0.112	0.888	0.711	2.5
<b>D18S51</b>	0.725	0.026	0.974	0.871	3.7	0.683	0.086	0.914	0.848	3.2	0.862	0.040	0.960	0.869	7.4
<b>D5S818</b>	0.440	0.119	0.881	0.683	1.7	0.565	0.150	0.850	0.675	2.3	0.372	0.098	0.902	0.716	1.5
<b>FGA</b>	0.733	0.032	0.968	0.861	3.8	0.565	0.061	0.939	0.811	2.3	0.862	0.043	0.957	0.861	7.4
<b>Combined</b>	0.99999863	1.02x10 <sup>-18</sup>				0.99999835	5.56x10 <sup>-16</sup>				0.999999666	4.25x10 <sup>-17</sup>			

**Supplementary Table S8** Population samples according to district and self-declared ethno-linguistic population group used in the study (n=990).

<b>District</b>	<b>n</b>	<b>Khoisan</b>	<b>Bantu</b>
<b>Central</b>	264	9	255
<b>Ghanzi</b>	168	156	12
<b>Kgalagadi</b>	20	6	14
<b>Kgatleng</b>	21	-	21
<b>Kweneng</b>	106	1	105
<b>Northeast</b>	45	-	45
<b>Northwest</b>	260	66	194
<b>Southeast</b>	32	-	32
<b>Southern</b>	74	-	74
<b>total</b>	990	238	752

**Supplementary Table S9** Population samples according to their self-declared Bantu and Khoisan ethno-linguistic population groups (n=990).

<b>Language Family</b>	<b>Branch</b>	<b>Sub-group</b>	<b>n</b>
<b>Niger-Congo</b>	Narrow Bantu (Bantu)	Central K <sup>1</sup>	43
		Central R <sup>2</sup>	58
		Central S <sup>3</sup>	646
		Undeclared Bantu	5
<b>Khoisan</b>	Southern African Khoisan (SAK)	Khoe-Kwadi	133
		Kx'a	58
		Tuu	32
		Undeclared Khoisan	15

<sup>1</sup>Central K- Subiya, Mbukushu

<sup>2</sup>Central R- Yeyi

<sup>3</sup>Central S- Kgalagadi, Kalanga, Tswana (and its dialects-Barolong, Bakwena, Bangwaketse, Balete, Bakgatla, Barolong, Bangwato, and Batawana)