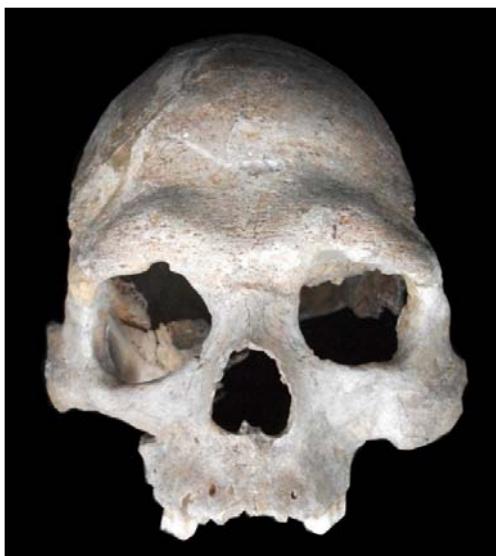


Extended Data

Extended Data Figure 1: Photographs of analyzed ancient samples. (A) Loschbour skull; (B) Stuttgart skull, missing the lower right M2 we sampled; (C) excavation at Kanaljorden in Motala, Sweden; (D) Motala 1 in situ.

A



B



C



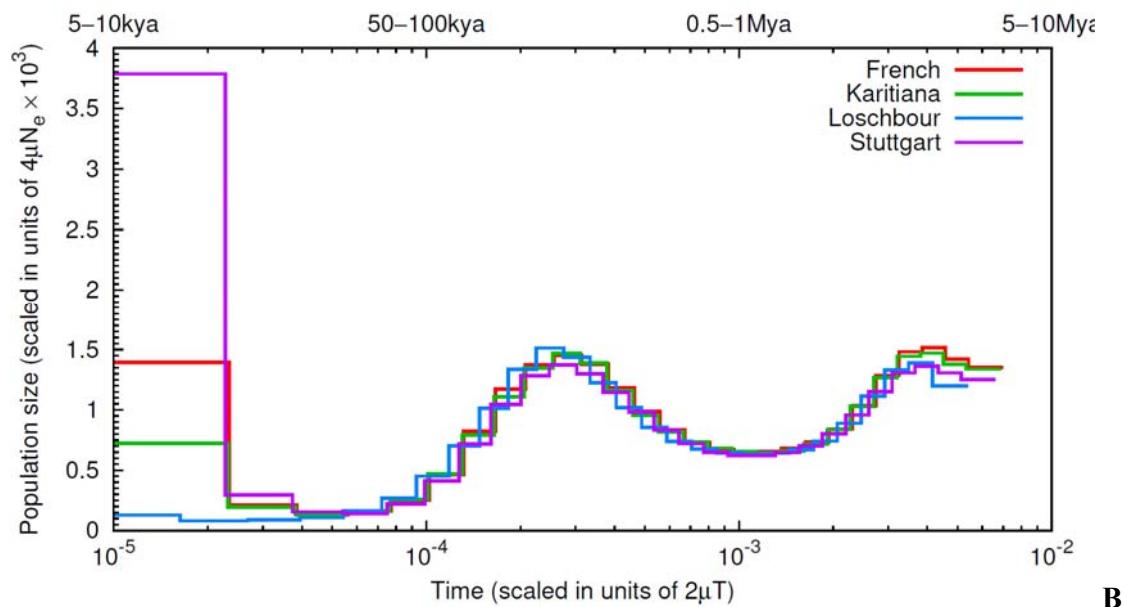
D



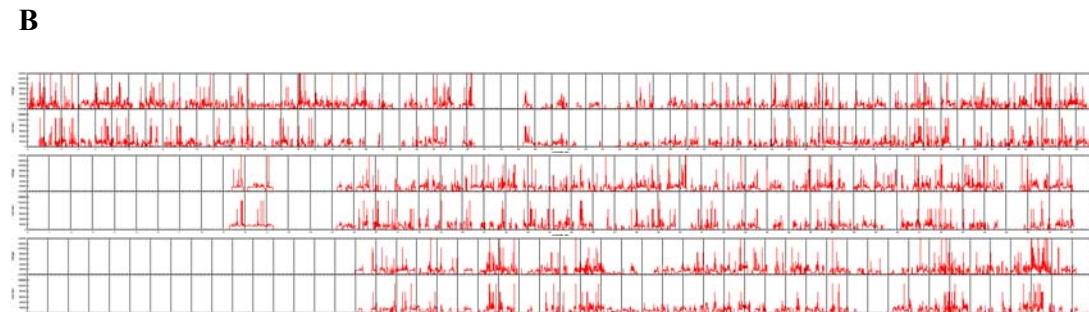
Extended Data Figure 2: Pairwise Sequential Markovian Coalescent (PSMC) analysis.

(A) Inference of population size as a function of time, showing a very small recent population size over the most recent period in the ancestry of Loschbour (at least the last 5-10 thousand years). (B) Inferred time since the most recent common ancestor from the PSMC for chromosomes 20, 21, 22 (top to bottom); Stuttgart is plotted on top and Loschbour at bottom.

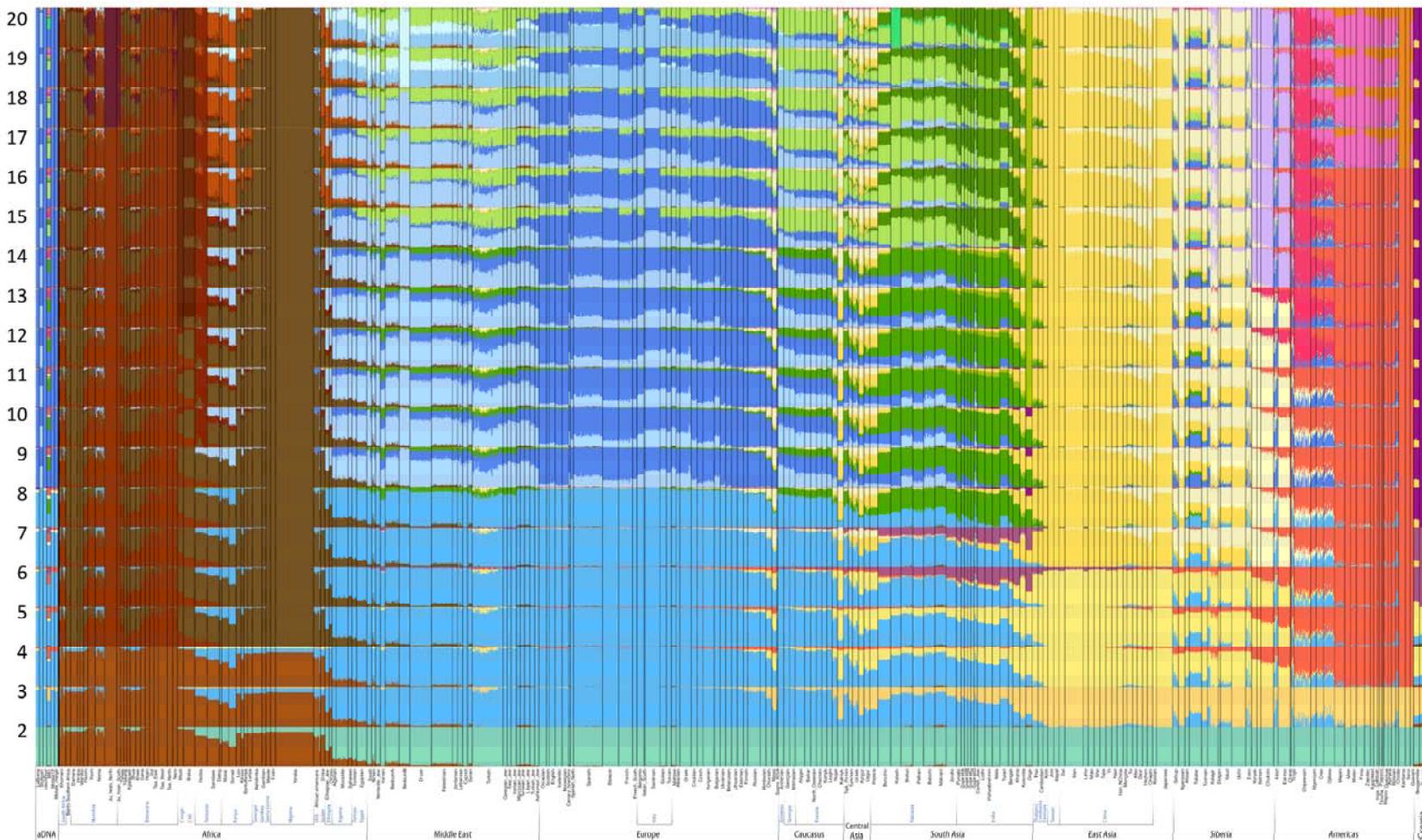
A



B

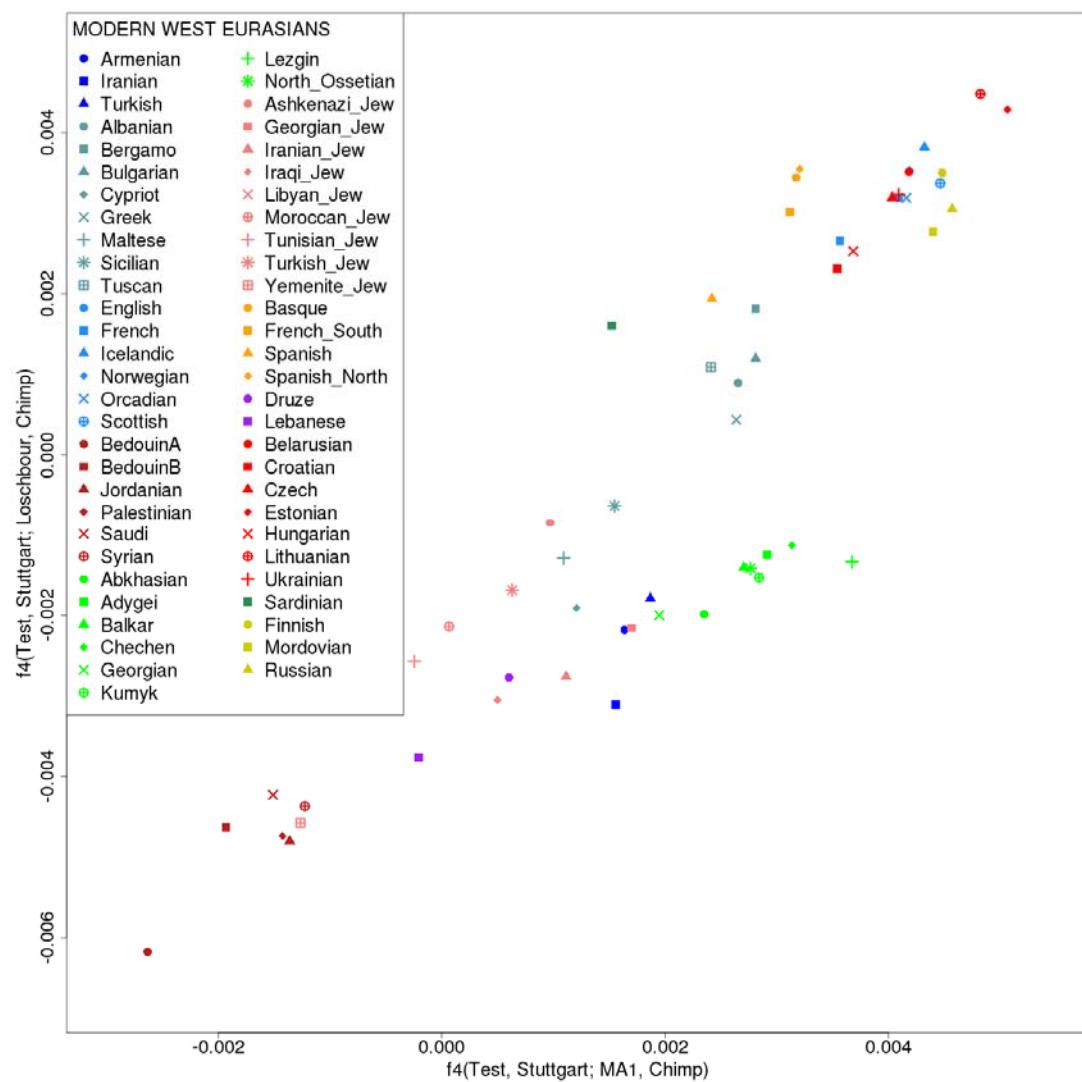


Extended Data Figure 3: ADMIXTURE analysis (K=2 to K=20). Ancient samples (Loschbour, Stuttgart, Motala_merge, Motala12, MA1, and LaBrana) are at left.



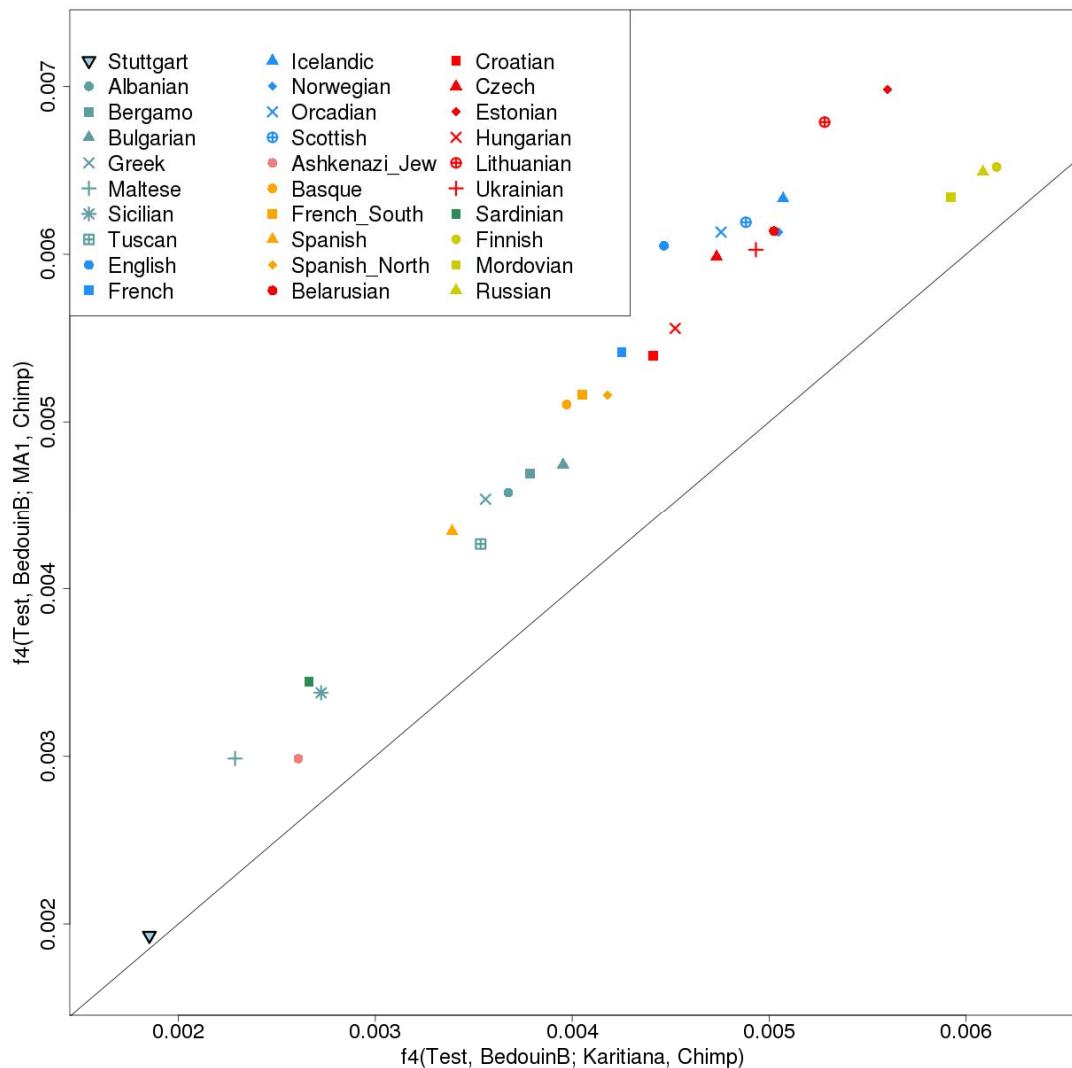
Extended Data Figure 4: ANE ancestry is present in both Europe and the Near East but WHG ancestry is restricted to Europe, which cannot be due to a single admixture event.

(x-axis) We computed the statistic $f_4(\text{Test}, \text{Stuttgart}; \text{MA1}, \text{Chimp})$, which measures where MA1 shares more alleles with a test population than with Stuttgart. It is positive for most European and Near Eastern populations, consistent with ANE (MA1-related) gene flow into both regions. (y-axis) We computed the statistic $f_4(\text{Test}, \text{Stuttgart}; \text{Loschbour}, \text{Chimp})$, which measures whether Loschbour shares more alleles with a test sample than with Stuttgart. Only European populations show positive values of this statistic, providing evidence of WHG (Loschbour-related) admixture only in Europeans.



Extended Data Figure 5: MA1 is the best surrogate for ANE for which we have data.

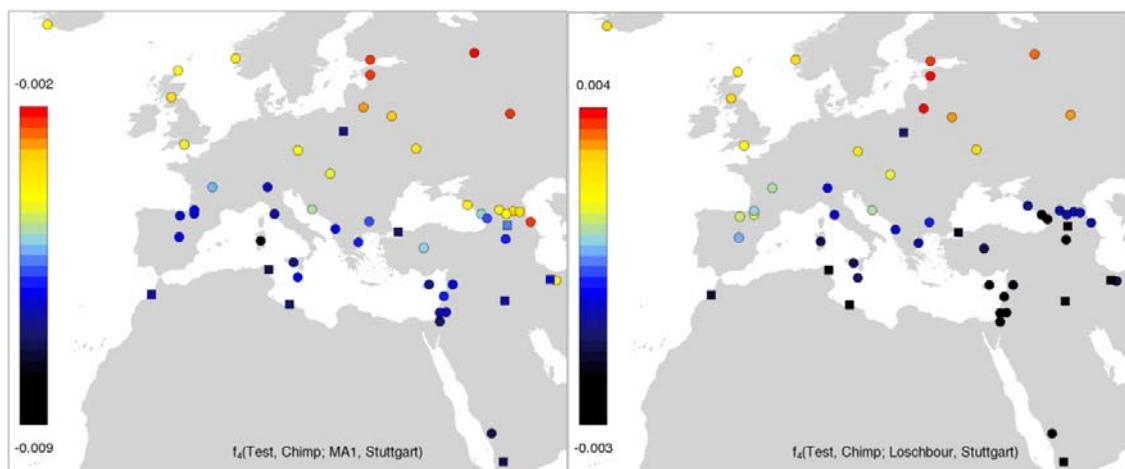
Europeans share more alleles with MA1 than with Karitiana, as we see from the fact that in a plot of $f_4(\text{Test}, \text{BedouinB}; \text{MA1}, \text{Chimp})$ and $f_4(\text{Test}, \text{BedouinB}; \text{Karitiana}, \text{Chimp})$, the European cline deviates in the direction of MA1, rather than Karitiana (the slope is >1 and European populations are above the line indicating equality of these two statistics).



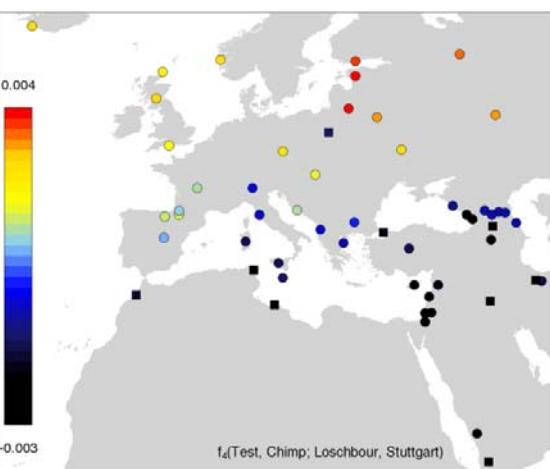
Extended Data Figure 6: The differential relatedness of West Eurasians to Stuttgart (EEF), Loschbour (WHG), and MA1 (ANE) cannot be explained by two-way mixture.

We plot on a West Eurasian map the statistic $f_4(\text{Test}, \text{Chimp}; A_1, A_2)$, where A_1 and A_2 are a pair of the three ancient samples representing the three ancestral populations of Europe. (A) In both Europe and the Near East/Caucasus, populations from the south have more relatedness to Stuttgart than those from the north where ANE influence is also important. (B) Northern European populations share more alleles with Loschbour than with Stuttgart, as they have additional WHG ancestry beyond what was already present in EEF. (C) We observe a striking contrast between Europe west of the Caucasus and the Near East in degree of relatedness to WHG. In Europe, there is a much higher degree of allele sharing with Loschbour than with MA1, which we ascribe to the 60-80% WHG/(WHG+ANE) ratio in most Europeans that we report in SI14. In contrast, the Near East has no appreciable WHG ancestry but some ANE ancestry, especially in the northern Caucasus. (Jewish populations are marked with a square in this figure to assist in interpretation as their ancestry is often anomalous for their geographic regions.)

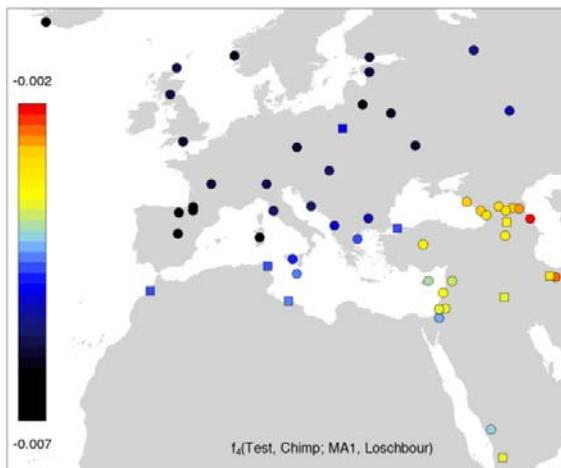
A



B

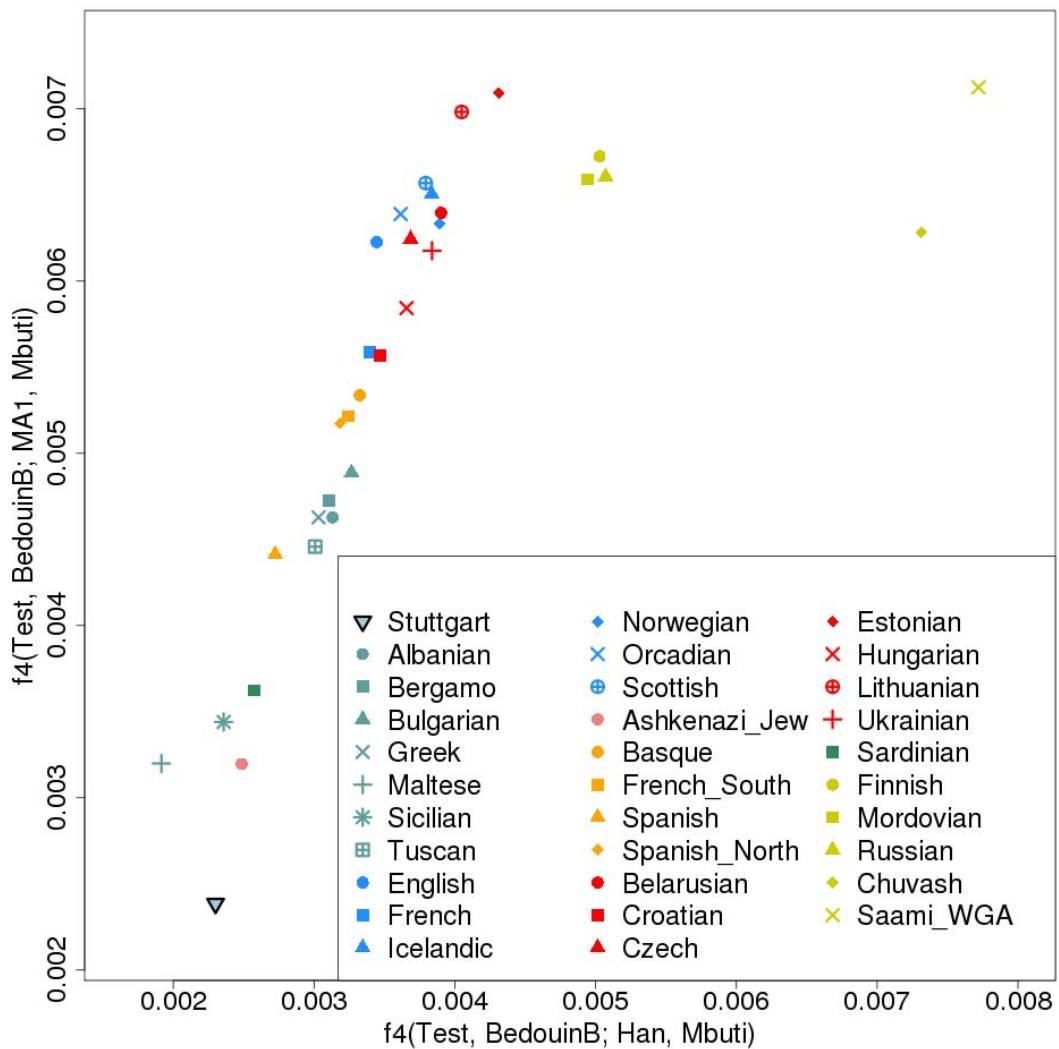


C



Extended Data Figure 7: Evidence for Siberian gene flow into far northeastern Europe.

Some northeastern European populations (Chuvash, Finnish, Russian, Mordovian, Saami) share more alleles with Han Chinese than with other Europeans who are arrayed in a cline from Stuttgart to Lithuanians/Estonians in a plot of $f_4(\text{Test}, \text{BedouinB}; \text{Han}, \text{Mbuti})$ against $f_4(\text{Test}, \text{BedouinB}; \text{MA1}, \text{Mbuti})$.



Extended Data Table 2: Confirmation of key findings on transversions and on whole genome sequence data.

Interpretation	D(A, B; C, D) on Human Origins genotype data								D(A, B; C, D) on whole genome sequence data transversions					
					594,924 SNPs		110,817 transversions						statistic	
	A	B	C	D	statistic	Z	statistic	Z	A	B	C	D	statistic	Z
Stuttgart has Near Eastern ancestry	Stuttgart	Armenian	Loschbour	Chimp	0.0219	4.5	0.0189	2.9						
Europeans have more WHG-related ancestry than Stuttgart	Stuttgart	French	Loschbour	Chimp	-0.0266	-5.7	-0.031	-5.0	Stuttgart	French2	Loschbour	Chimp	-0.03	-4.7
	Lithuanian	Stuttgart	Loschbour	Chimp	0.0446	9.1	0.0477	7.2						
West Eurasians have more ANE-related ancestry than Stuttgart	French	Stuttgart	MA1	Chimp	0.0367	7.7	0.0386	5.5	French2	Stuttgart	MA1	Chimp	0.037	6.4
	Lezgin	Stuttgart	MA1	Chimp	0.0372	7.6	0.0409	5.6						
MA1 is a better surrogate of ANE ancestry than Karitiana	French	Chimp	MA1	Karitiana	0.0207	4.5	0.0214	2.8	French2	Chimp	MA1	Karitiana2	0.026	3.8
Eastern non-Africans closer to WHG/ANE/SHG than to EEF	Loschbour	Stuttgart	Onge	Chimp	0.0196	3.5	0.0202	2.5	Loschbour	Stuttgart	Papuan2	Chimp	0.017	2.7
	Loschbour	Stuttgart	Papuan	Chimp	0.0142	2.6	0.0127	1.5	Loschbour	Stuttgart	Dai2	Chimp	0.018	2.9
	Loschbour	Stuttgart	Dai	Chimp	0.0164	3.2	0.021	2.8	MA1	Stuttgart	Papuan2	Chimp	0.018	2.8
	MA1	Stuttgart	Papuan	Chimp	0.0139	2.2	0.0103	1.0	MA1	Stuttgart	Dai2	Chimp	0.028	4.3
	MA1	Stuttgart	Dai	Chimp	0.0174	3.0	0.016	1.7	Motala12	Stuttgart	Papuan2	Chimp	0.023	3.7
	Motala12	Stuttgart	Papuan	Chimp	0.0182	3.2	0.011	1.1	Motala12	Stuttgart	Dai2	Chimp	0.02	3.2
	Motala12	Stuttgart	Dai	Chimp	0.0156	2.8	0.0149	1.6	LaBrana	Stuttgart	Papuan2	Chimp	0.02	3.2
	LaBrana	Stuttgart	Papuan	Chimp	0.0123	2.3	0.0101	1.1	LaBrana	Stuttgart	Dai2	Chimp	0.024	3.7
	LaBrana	Stuttgart	Dai	Chimp	0.0149	2.9	0.0228	2.5						
Native Americans closer to ANE than to WHG	Karitiana	Chimp	MA1	Loschbour	0.0467	7.1	0.0467	4.4	Karitiana2	Chimp	MA1	Loschbour	0.052	7.1
West Eurasians closer to Native Americans than to other Eastern non-Africans	Stuttgart	Chimp	Karitiana	Papuan	0.0559	10.9	0.0474	6.6	Stuttgart	Chimp	Karitiana2	Papuan2	0.052	7.6
	Stuttgart	Chimp	Karitiana	Onge	0.0237	5.1	0.0179	2.6						
Ancient Eurasian hunter-gatherers equally related to Eastern non-Africans other than Native Americans	Loschbour	MA1	Dai	Chimp	-0.0015	-0.2	0.0016	0.2	Loschbour	MA1	Dai2	Chimp	-0.013	-1.9
	Loschbour	MA1	Papuan	Chimp	0.0002	0.0	0.0012	0.1	Loschbour	MA1	Papuan2	Chimp	-0.003	-0.4
	Loschbour	Motala12	Dai	Chimp	0.0024	0.4	0.009	0.9	Loschbour	Motala12	Dai2	Chimp	-0.002	-0.3
	Loschbour	Motala12	Papuan	Chimp	-0.0028	-0.4	0.0046	0.5	Loschbour	Motala12	Papuan2	Chimp	-0.004	-0.6
	MA1	Motala12	Dai	Chimp	0.0026	0.4	0.0047	0.4	MA1	Motala12	Dai2	Chimp	0.01	1.5
	MA1	Motala12	Papuan	Chimp	-0.0047	-0.7	-0.001	-0.1	MA1	Motala12	Papuan2	Chimp	-0.004	-0.5
LaBrana and Loschbour are a clade	LaBrana	Loschbour	Dai	Chimp	-0.0028	-0.5	0.0024	0.3	LaBrana	Loschbour	Dai2	Chimp	0.007	1.1
	LaBrana	Loschbour	Papuan	Chimp	-0.0031	-0.5	-0.0012	-0.1	LaBrana	Loschbour	Papuan2	Chimp	0.002	0.3
	LaBrana	Loschbour	MA1	Chimp	-0.006	-0.8	0.0101	0.7	LaBrana	Loschbour	MA1	Chimp	0.005	0.7
SHG closer to ANE than to WHG	Motala12	Loschbour	MA1	Chimp	0.0425	5.3	0.0353	2.6	Motala12	Loschbour	MA1	Chimp	0.042	5.9
	Motala12	LaBrana	MA1	Chimp	0.0465	5.8	0.0347	2.4	Motala12	LaBrana	MA1	Chimp	0.038	5.4
LaBrana and Loschbour equally related to Stuttgart	LaBrana	Loschbour	Stuttgart	Chimp	-0.0176	-2.6	-0.0106	-1.0	LaBrana	Loschbour	Stuttgart	Chimp	-0.012	-1.8

Extended Data Table 3: Admixture proportions for European populations. The estimates from the model with minimal assumptions are from SI17. The estimates from the full modeling are from SI14 either by single population analysis or co-fitting population pairs and averaging over fits (these averages are the results plotted in Fig. 2B). Populations that do not fit the models are not reported.

	Full modeling of population relationships (individual fits)			Full modeling of population relationships (averaged fits)						Modeling of population relationships with minimal assumptions			Model-based (averaged) - Model with minimal assumptions (Z-score)		
	EEF	WHG	ANE	EEF		WHG		ANE		EEF	WHG	ANE	EEF	WHG	ANE
				Mean	Range	Mean	Range	Mean	Range						
Albanian	0.781	0.092	0.127	0.781	0.772-0.819	0.082	0.032-0.098	0.137	0.129-0.158	0.595 ± 0.112	0.353 ± 0.150	0.052 ± 0.049	1.658	-1.807	1.741
Ashkenazi_Jew	0.931	0	0.069							0.938 ± 0.146	-0.021 ± 0.185	0.083 ± 0.049			
Basque	0.593	0.293	0.114	0.569	0.527-0.616	0.335	0.255-0.392	0.096	0.076-0.129	0.569 ± 0.091	0.315 ± 0.124	0.115 ± 0.041	-0.001	0.165	-0.472
Belarusian	0.418	0.431	0.151	0.426	0.397-0.464	0.408	0.338-0.443	0.167	0.150-0.199	0.272 ± 0.094	0.554 ± 0.131	0.174 ± 0.047	1.637	-1.118	-0.158
Bergamo	0.715	0.177	0.108	0.721	0.704-0.793	0.163	0.061-0.189	0.117	0.104-0.147	0.644 ± 0.125	0.248 ± 0.170	0.108 ± 0.053	0.615	-0.503	0.162
Bulgarian	0.712	0.147	0.141	0.718	0.707-0.778	0.132	0.047-0.151	0.151	0.138-0.175	0.556 ± 0.110	0.328 ± 0.143	0.116 ± 0.043	1.469	-1.372	0.804
Croatian	0.561	0.293	0.145	0.564	0.548-0.586	0.285	0.242-0.310	0.151	0.137-0.172	0.453 ± 0.122	0.407 ± 0.159	0.140 ± 0.046	0.911	-0.768	0.238
Czech	0.495	0.338	0.167	0.489	0.460-0.531	0.348	0.273-0.382	0.163	0.145-0.196	0.402 ± 0.117	0.400 ± 0.162	0.198 ± 0.050	0.744	-0.322	-0.698
English	0.495	0.364	0.141	0.503	0.476-0.536	0.353	0.296-0.382	0.144	0.130-0.169	0.475 ± 0.091	0.357 ± 0.125	0.168 ± 0.043	0.304	-0.028	-0.561
Estonian	0.322	0.495	0.183	0.323	0.293-0.345	0.49	0.451-0.520	0.187	0.172-0.205	0.072 ± 0.121	0.778 ± 0.176	0.150 ± 0.064	2.070	-1.636	0.584
French	0.554	0.311	0.135	0.563	0.537-0.601	0.297	0.230-0.328	0.14	0.126-0.169	0.498 ± 0.097	0.359 ± 0.127	0.142 ± 0.039	0.672	-0.487	-0.060
French_South	0.675	0.195	0.13	0.636	0.589-0.738	0.256	0.111-0.323	0.108	0.088-0.151	0.636 ± 0.116	0.225 ± 0.165	0.140 ± 0.057	-0.003	0.189	-0.558
Greek	0.792	0.058	0.151	0.791	0.780-0.816	0.048	0.019-0.060	0.161	0.150-0.171	0.658 ± 0.098	0.255 ± 0.127	0.086 ± 0.039	1.357	-1.627	1.915
Hungarian	0.558	0.264	0.179	0.548	0.520-0.590	0.279	0.199-0.313	0.174	0.156-0.210	0.391 ± 0.109	0.454 ± 0.153	0.155 ± 0.050	1.437	-1.145	0.371
Icelandic	0.394	0.456	0.15	0.409	0.386-0.424	0.448	0.409-0.473	0.143	0.126-0.170	0.342 ± 0.102	0.476 ± 0.137	0.182 ± 0.045	0.654	-0.204	-0.861
Lithuanian	0.364	0.464	0.172	0.352	0.327-0.384	0.488	0.433-0.527	0.16	0.135-0.184	0.248 ± 0.117	0.548 ± 0.163	0.205 ± 0.052	0.886	-0.367	-0.864
Maltese	0.932	0	0.068							1.298 ± 0.185	-0.509 ± 0.248	0.211 ± 0.079			
Norwegian	0.411	0.428	0.161	0.417	0.388-0.438	0.423	0.383-0.450	0.16	0.140-0.181	0.273 ± 0.115	0.557 ± 0.161	0.170 ± 0.055	1.252	-0.831	-0.185
Orcadian	0.457	0.385	0.158	0.465	0.439-0.493	0.378	0.329-0.403	0.157	0.140-0.179	0.395 ± 0.088	0.437 ± 0.122	0.168 ± 0.041	0.798	-0.487	-0.264
Sardinian	0.817	0.175	0.008	0.818	0.791-0.874	0.141	0.058-0.182	0.041	0.026-0.068	0.883 ± 0.128	0.075 ± 0.166	0.042 ± 0.048	-0.510	0.400	-0.024
Scottish	0.39	0.428	0.182	0.408	0.387-0.424	0.421	0.384-0.448	0.171	0.149-0.201	0.286 ± 0.112	0.532 ± 0.156	0.182 ± 0.053	1.091	-0.712	-0.210
Sicilian	0.903	0	0.097							1.012 ± 0.149	-0.131 ± 0.199	0.119 ± 0.060			
Spanish	0.809	0.068	0.123	0.759	0.736-0.804	0.126	0.066-0.170	0.115	0.091-0.151	0.856 ± 0.126	-0.015 ± 0.165	0.160 ± 0.049	-0.769	0.855	-0.922
Spanish_North	0.713	0.125	0.163	0.612	0.561-0.660	0.292	0.214-0.365	0.096	0.072-0.126	0.581 ± 0.120	0.298 ± 0.158	0.121 ± 0.046	0.254	-0.038	-0.533
Tuscan	0.746	0.136	0.118	0.751	0.737-0.806	0.123	0.047-0.145	0.126	0.114-0.150	0.734 ± 0.118	0.153 ± 0.160	0.113 ± 0.054	0.141	-0.188	0.249
Ukrainian	0.462	0.387	0.151	0.463	0.445-0.491	0.376	0.322-0.399	0.16	0.148-0.187	0.259 ± 0.123	0.596 ± 0.173	0.145 ± 0.057	1.661	-1.269	0.269
Finnish										-0.299 ± 0.204	1.194 ± 0.296	0.105 ± 0.105			
Mordovian										-0.255 ± 0.173	1.151 ± 0.246	0.104 ± 0.090			
Russian										-0.303 ± 0.211	1.230 ± 0.301	0.072 ± 0.106			