Supplementary Figures

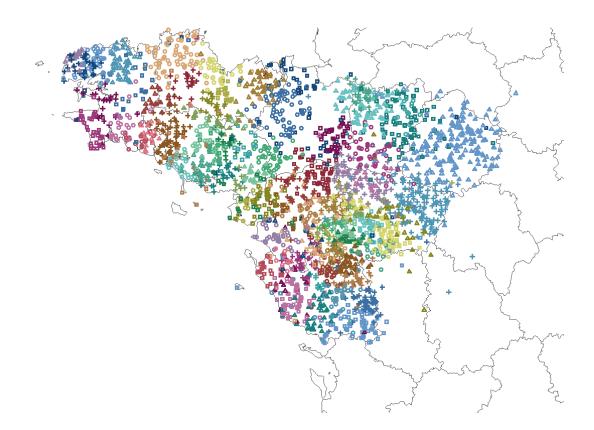


Figure S1. fineSTRUCTURE results for k=154. But only clusters with >10 individuals (78 clusters) are shown. Map generated with *R statistical package* using *sp* package and the "*départements*" boundary dataset from the French platform of open data (<u>https://www.data.gouv.fr/</u>).

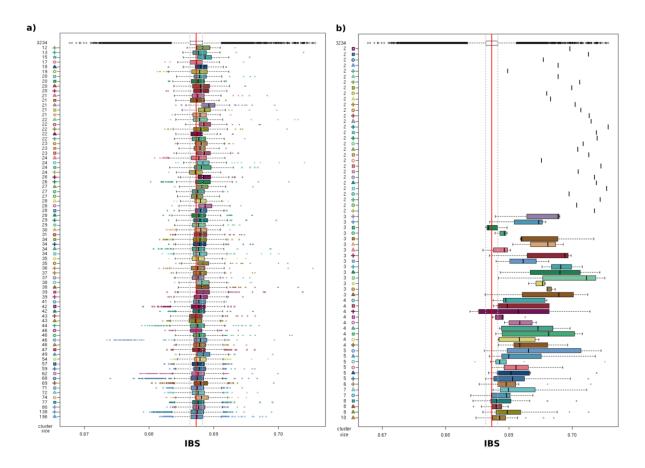


Figure S2. Identity-by-state (IBS) distances computed between pairs of individuals within the 154 clusters for the finest level of population structure. The white box plot represents the distribution of pairwise IBS values, computed with plink *--genome* option, across the whole dataset. Pairwise IBS values are shown in the x-axis while the y-axis shows the 78 clusters with n > 10 (a) and the remaining clusters with n <= 10 (b). Boxplots are coloured according to the cluster colours in Fig. S1. The distributions of relatedness, here captured by the IBS distances, within clusters globally match that computed across the whole sample (median distribution within the 25th-75th percentiles). This occurs for most clusters with a n > 10 (a). However, a departure from the whole distribution is observed for many of the clusters with a n < 10 (b). Given that fineSTRUCTURE detects groups of individuals with higher identity-by-descent, the smaller groups with increased IBS likely represent very recent shared ancestry (e.g., family relationships undetectable in the questionnaire, inbreeding).

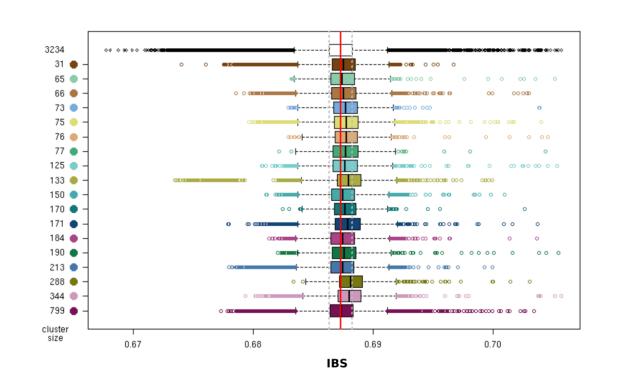


Figure S3. Identity-by-state (IBS) between pairs of individuals belonging to the 18 clusters inferred based on the TVD-based tree on fineSTRUCTURE clustering results. The distributions of relatedness within the 18 clusters (discussed in the main text) match that computed across the whole sample (white boxplot), indicating that individuals within clusters (n > 10) do not exhibit significantly more IBS than pairs of randomly selected individuals in the whole sample. Nevertheless, relatedness distributions slightly shift towards higher values as expected given that population structure is caused by shared ancestry. Such shared ancestry should be reflected over a large timescale and not only restricted to very recent periods (IBS \sim 1). Boxplots are coloured according to the cluster colours in Fig. 1d (main text).

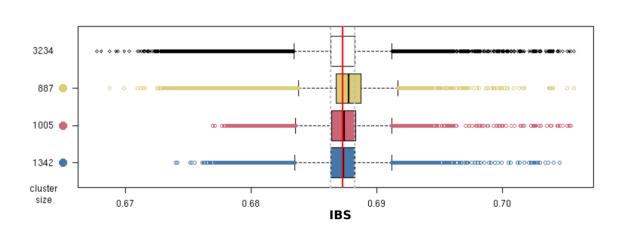


Figure S4. Identity-by-state (IBS) between pairs of individuals within the three clusters. The distributions of relatedness within the three clusters (Fig. 1a) match those computed across the whole sample (white boxplot), indicating that individuals within clusters (n > 10) do not exhibit significantly more IBS than pairs of randomly selected individuals in the whole sample. Overall, we conclude that fine-scale structure found in our study is not due to an overrepresentation of recently related samples.

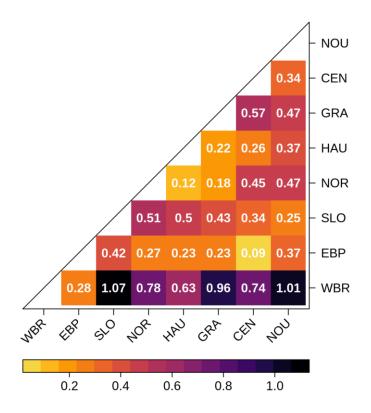


Figure S5. Pairwise average Weir and Cockerham's F_{ST}s within France x 1000. F_{ST} values were computed on the WGS dataset after pruning for LD and using n=25 samples/population with the exception of Normandy whose n=19. Cluster names: Western Brittany (WBR), Eastern Brittany/*Pays-de-la-Loire* (EBP) and South Loire (SLO). Population acronyme: NOR, *Normandie*; HAU, Hauts-de-France; GRA, Grand Est; CEN, Centre-Val de Loire; NOU, Nouvelle-Aquitaine.

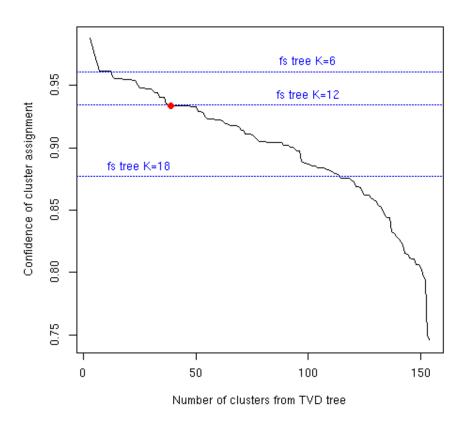


Figure S6. Confidence of cluster assignment from fineSTRUCTURE and TVD-based trees. FS-tree has lower confidence of cluster assignment than TVD-based tree for the same levels of number of clusters. For example, cluster assignment confidence retrieved for k=39 in TVD-based tree (red point) is similar for k=12 in FS-tree. See Material and Methods for details on the computation of cluster confidence (ChromoPainter and fineSTRUCTURE).

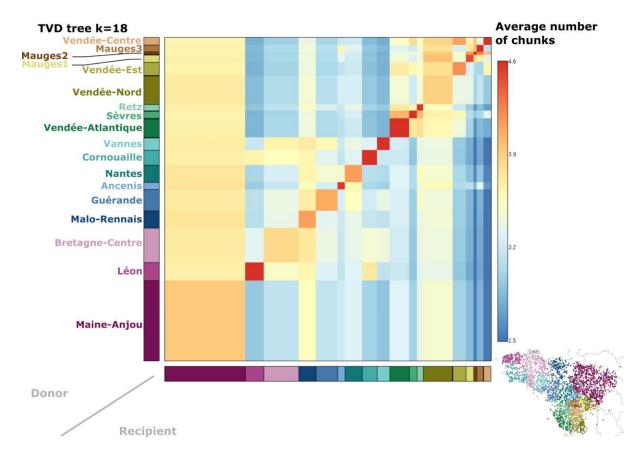


Figure S7. Population-level coancestry matrix for k=18. This matrix was based on the individual-level coancestry (chunkcount matrix) used in cluster inference. Along the columns is shown the average chunk counts a recipient cluster (bottom) received from the donor clusters (on the left) while across the rows is shown the average chunk count that donor clusters (on the left) have contributed to the recipient clusters (bottom). The diagonal represents the average chunk count between pairs of individuals assigned to the same cluster, i.e. the "drift component". In order to visualise the bulk of the variation, only values between 1-99 percentile were considered and those exceeding the range were coloured according to the closer 1 or 99 percentile value. Warmer colours indicate higher levels of ancestry sharing. Map generated in *R statistical package* using *sp* package and the "*départements*" boundary dataset from the French platform of open data (<u>https://www.data.gouv.fr/</u>).

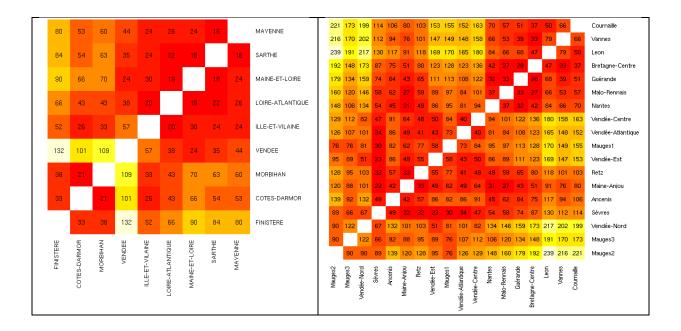


Figure S8. Pairwise F_{ST} values multiplied by 100,000 between the *départements* of Brittany and *Pays-de-la-Loire* (left panel) and the 18 clusters in Northwestern France (right panel) identified in PREGO dataset with fineSTRUCTURE (Fig. 1d, main text).

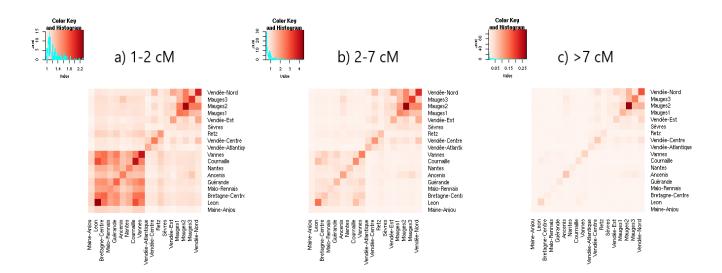


Figure S9. Identity-by-descent (IBD) sharing between pairs of individuals belonging to the 18 clusters identified with the TVD-based tree on fineSTRUCTURE (Fig. 1d main text) results for different chromosome segment sizes (a-c). Segment sizes are indicated over the heatmaps. Heatmap columns (from bottom to up) and rows (left to right) are ordered similarly to the coancestry matrices (see above).

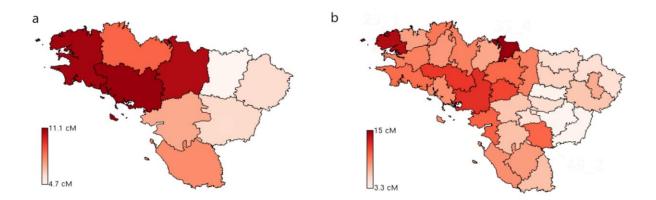


Figure S10. Heatmap displaying average length of runs of homozygosity (ROH) across individuals within a) *départements* and b) *arrondissements* of Northwestern France. The regions of Brittany and Mauges display large ROH indicating smaller effective population sizes Map generated with *R statistical package* using *sp* package and the "*départements*" and "*communes*" boundary dataset from the French platform of open data (<u>https://www.data.gouv.fr/</u>) and <u>OpenStreetMap</u>.

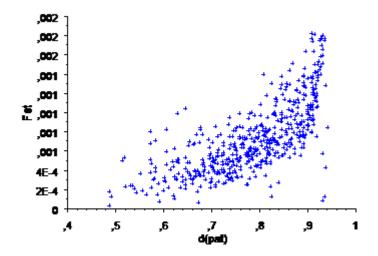


Figure S11. Correlation between surname distance and F_{ST} (negative F_{ST} removed), partial spearman correlation = 0.68.

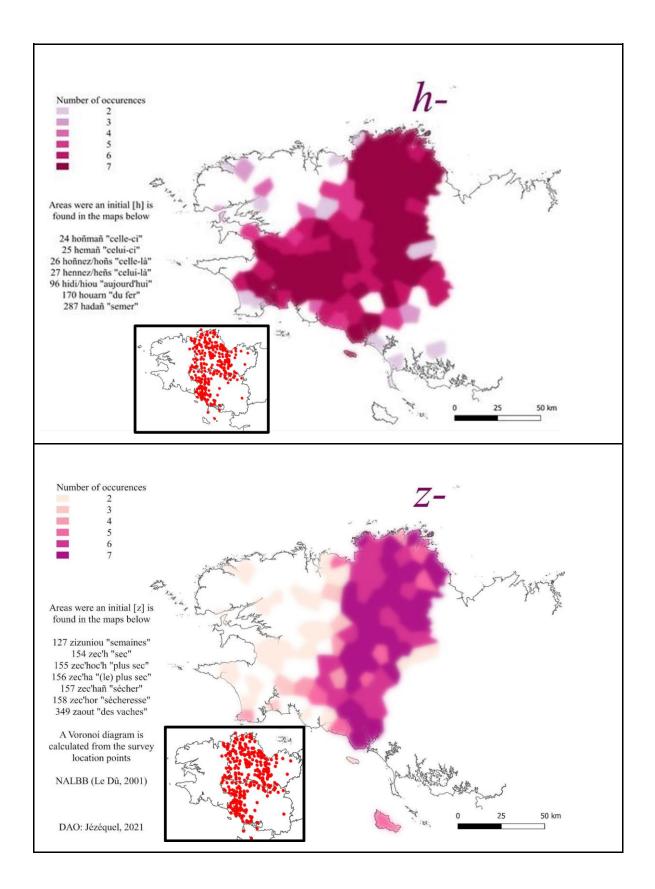


Figure S12. Distribution of the initials h- (upper) and z- (lower) outlining a central dialectal Breton area in Lower Brittany (See Supplementary Discussion). Both linguistic features overlap, at least to some extent, with the distribution of the cluster "Bretagne-Centre" (inset).

The usage of two initial consonants: the aspirated [h] instead of an unaspirated and the alveolar fricative [z] instead of [s], are linguistic features associated with dialectal areas of Breton as described in the *Nouvel Atlas Linguistique de Basse-Bretagne* (Le Dû 2001). The maps were obtained by overlapping word-specific maps of presence/absence across regions. The number of occurrences within polygons were computed using seven words (described on the left) in both cases. For instance, in the case of the occurrences of the initial *h-,* whose pronunciation changes are observed in words like *honman, heman, honnez, hennez, hidi, houarn, hadan,* darker fuchsia polygons mean that these seven words are present therein. Map generated with QGIS 3.22.11 (http://www.qgis.org/). Background map data from Diffusion.shom (https://diffusion.shom.fr/multiproduct/product/configure/id/93) is released under a CC BY-SA 4.0 license (https://creativecommons.org/licenses/by-sa/4.0/). Histolitt® Coastline data ©IGN-Shom 2009.

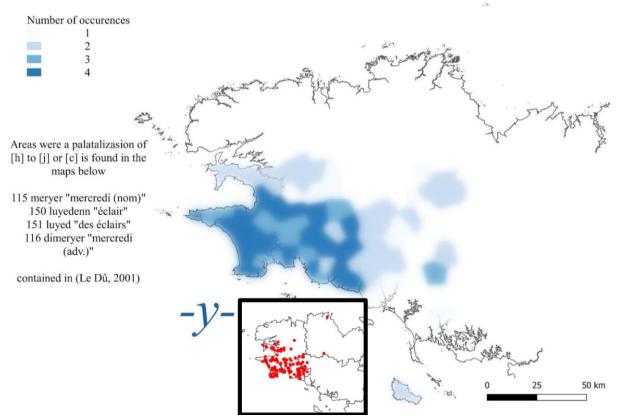
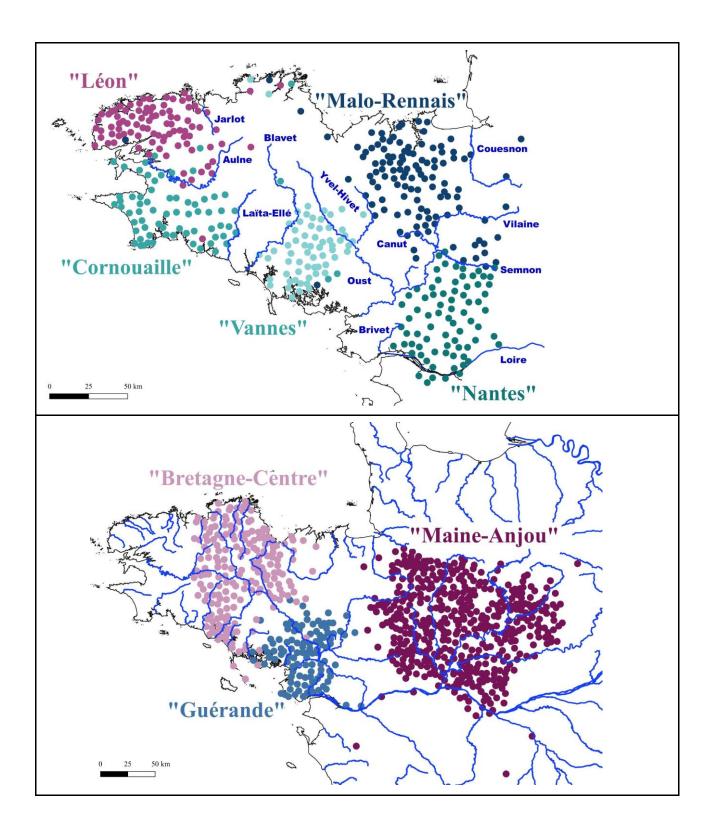


Figure S13. Distribution of the palatalisation of the *-h-* into *-y-* outlining a south-western dialectal area in Lower Brittany (See Supplementary Discussion). This linguistic feature was found to overlap with the distribution of the cluster "Cornouailles" (inset). The map was generated similarly to those in Fig. S12 (above), but focus on four words. Map generated with QGIS 3.22.11 (<u>http://www.qgis.org/</u>). Background map data from Diffusion.shom (<u>https://diffusion.shom.fr/multiproduct/product/configure/id/93</u>) is released under a CC BY-SA 4.0 license (<u>https://creativecommons.org/licenses/by-sa/4.0/</u>). Histolitt® Coastline data ©IGN-Shom 2009.



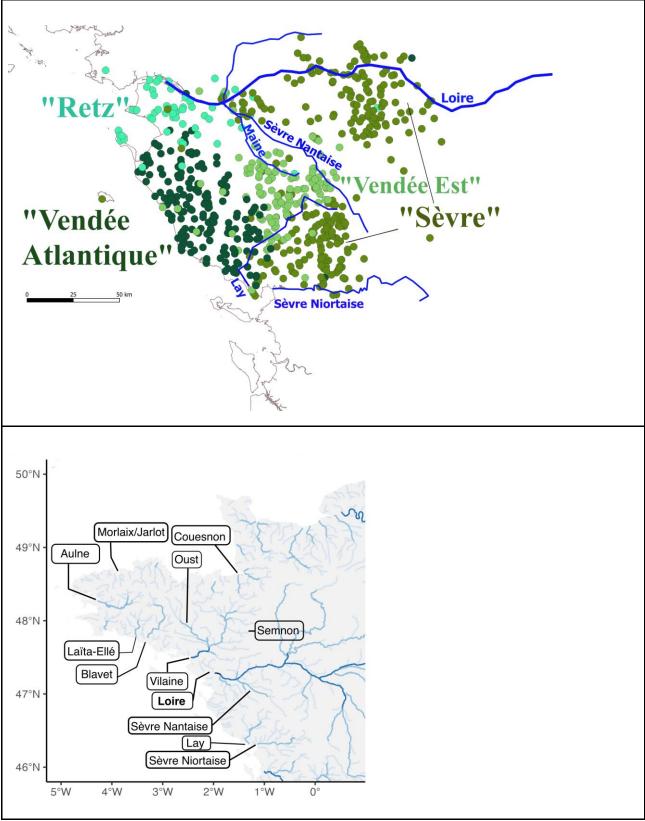


Figure S14. Clusters inferred with fineSTRUCTURE (k=18 of the TVD-tree) and the
distribution of local watercourses. The figure is split in three parts and clusters in the Mauges
region ("Mauges 1", "Mauges 2" and "Mauges 3") were not displayed to facilitate its
visualisation. The first three maps were generated with QGIS 3.22.11 (http://www.qgis.org/).BackgroundmapdatafromDiffusion.shom

(<u>https://diffusion.shom.fr/multiproduct/product/configure/id/93</u>) is released under a CC BY-SA 4.0 license (<u>https://creativecommons.org/licenses/by-sa/4.0/</u>). Histolitt® Coastline data ©IGN-Shom 2009.

The lower map represents the distribution of watercourses (blue) in Northwestern France. The darker the blue the higher the long-term average discharge of the river patch showing that most of the rivers overlapping with the borders of the genetic clusters are the major watercourses. The map was generated using the R statistical package together with the libraries: *ggplot2, sf, dyplr.* Hydrological data was obtained from: <u>https://www.hydrosheds.org</u>.

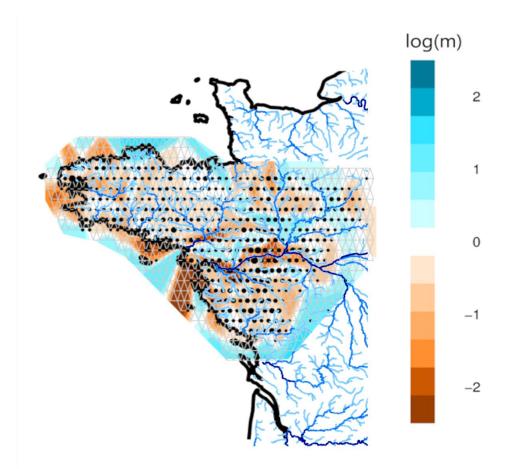


Figure S15. Effective migration surface inferred with EEMS overlapped with geographical location of waterbodies whose colour is proportional to long-term average discharge. Map generated with R statistical package together with the libraries: *ggplot2, sf, dyplr and rEEMSplots*. Hydrological data was obtained from: <u>https://www.hydrosheds.org</u>.

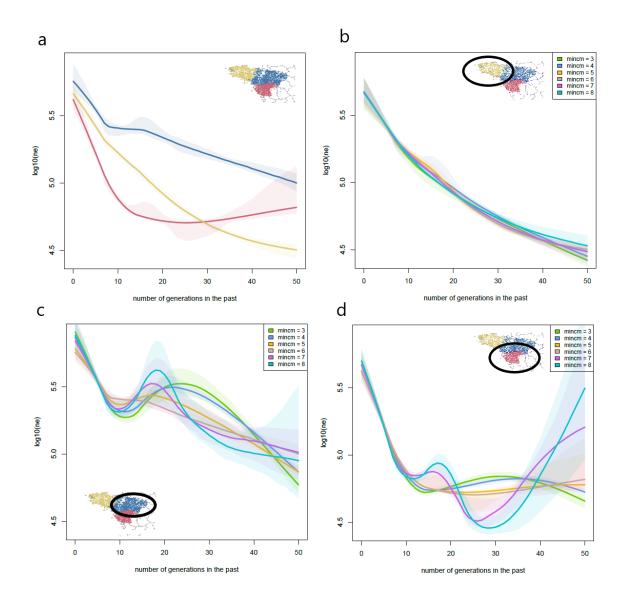


Figure S16. Effective population size trajectories for the three clusters inferred by fineSTRUCTURE (Fig. 1a, main text) obtained with IBDNe. a) IBDNe results for the three clusters together using minimum fragment length (*mincm*) of 6 cM. b-d) IBDNe results for each of the clusters shown in the inset across multiple *mincm* values.

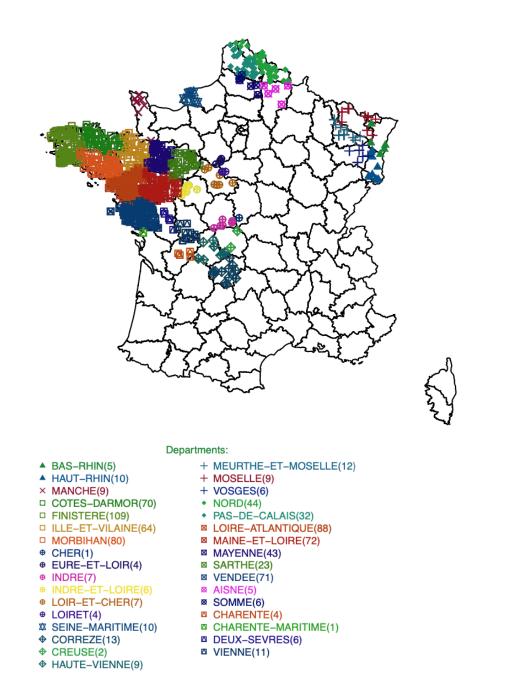
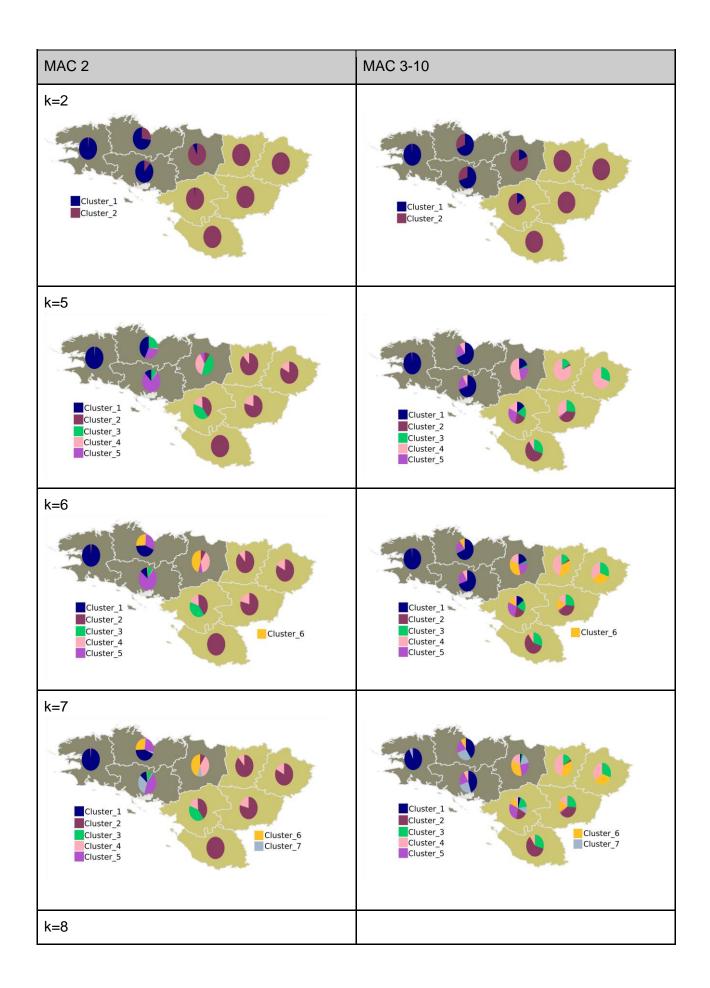


Figure S17. Sample distribution. Out of the 856 samples initially present in the WGS dataset we excluded related samples (PI_HAT > 0.10, three samples) and samples whose four grandparents were not born in the same *département* (10 samples). The 843 samples are distributed per regions as follows: Brittany (n=323), *Pays-de-la-Loire* (n=297), Normandy (n=19), *Hautsde-France* (n=87), *Grand Est* (n=42), *Centre* (n=29), *Nouvelle-Aquitaine* (n=46). Map generated with *R statistical package* using *sp* package and the "*départements*" boundary dataset from the French platform of open data (<u>https://www.data.gouv.fr/</u>).



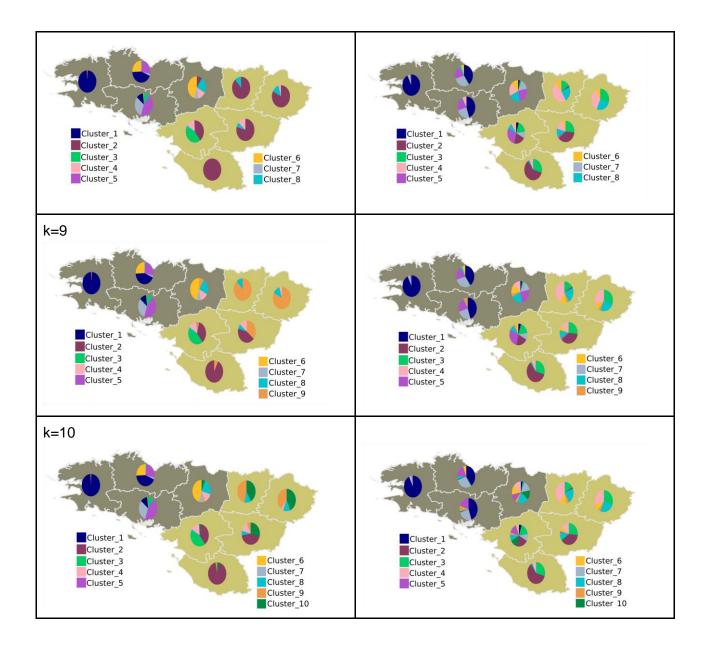
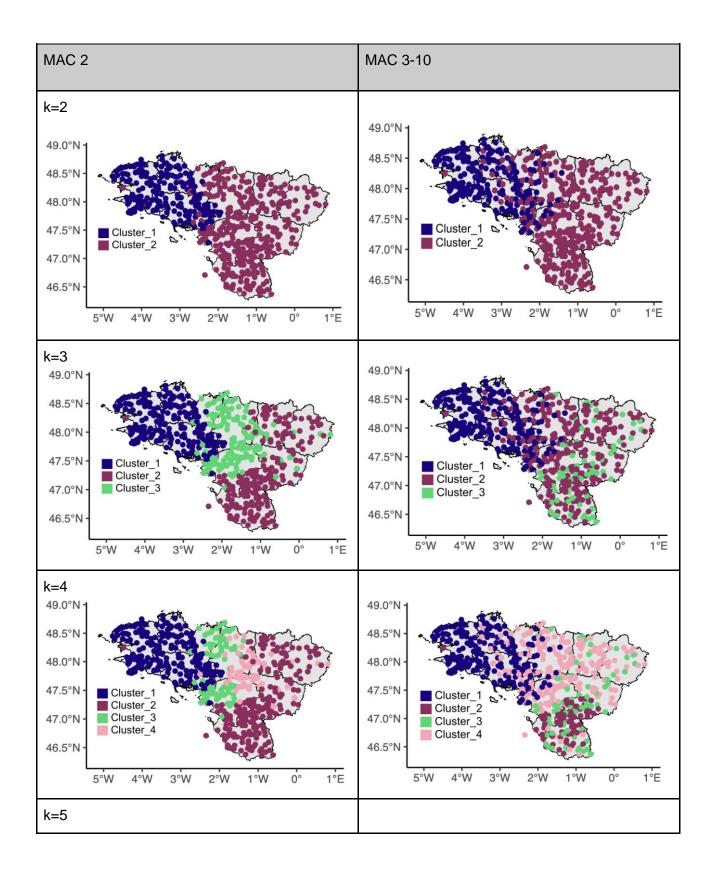
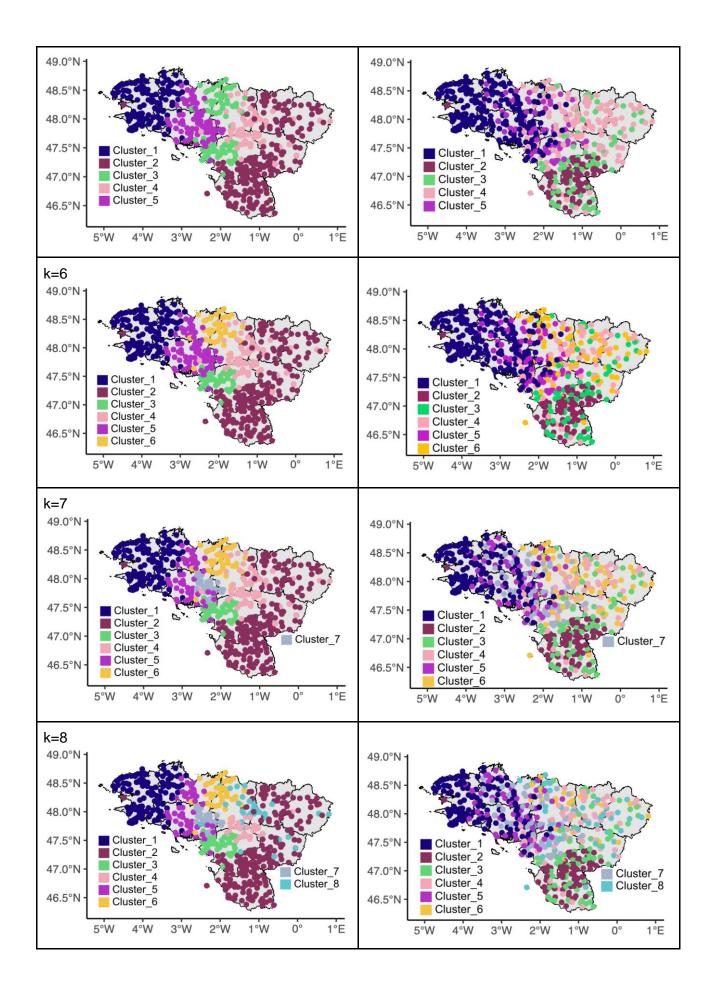


Figure S18. Distribution of individuals based on the hierarchical clustering performed on the allele-sharing matrices. Rows show different *k* values used in the hierarchical clustering and columns show the results for different allele count classes. Allele sharing matrices were obtained by randomly selecting 1 million variable sites within each of the two allele count categories and 620 individuals from Northwestern France. Map generated with *R statistical package* using *rgdal, sp, broom, ggplot2 and scatterpie* packages and the "*départements*" boundary dataset from the French platform of open data (https://www.data.gouv.fr/).





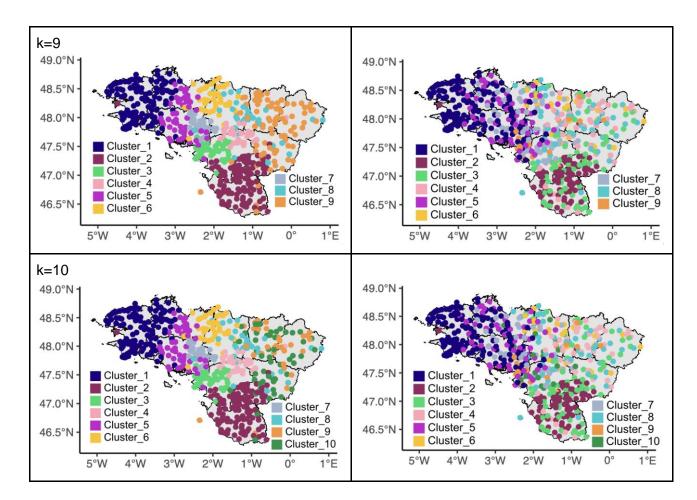


Figure S19. Individual cluster assignment based on the hierarchical clustering performed on the allele-sharing matrices. Rows show different *k* values used in the hierarchical clustering and columns show the results for different allele count classes. Allele sharing matrices were obtained described in Fig. S18 (above). Map generated with *R statistical package* using *rgdal, sp, broom,* and *ggplot2* packages and the "*départements*" boundary dataset from the French platform of open data (https://www.data.gouv.fr/).

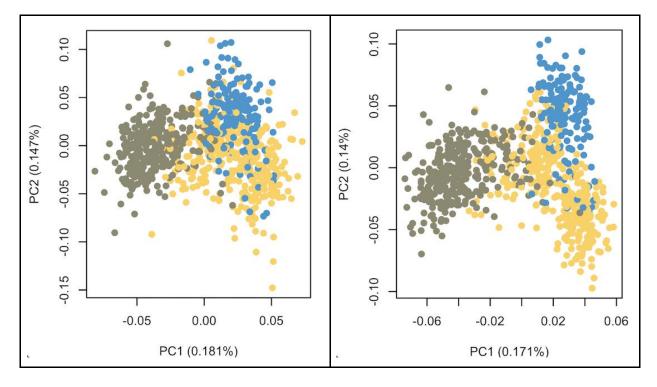


Figure S20. PCA on the WGS dataset containing only French populations for which all the four grandparents belong to the same *commune* (843 samples, Fig. S17). On the left panel, PCA was performed on common sites, MAF > 10% (~443,933 sites). The right panel shows the PCA low-frequency variants (1% < MAF < 10%, ~863,141 sites). Samples from Brittany are coloured in dark green and samples from Northeastern and Southwestern France are coloured in blue and yellow, respectively.

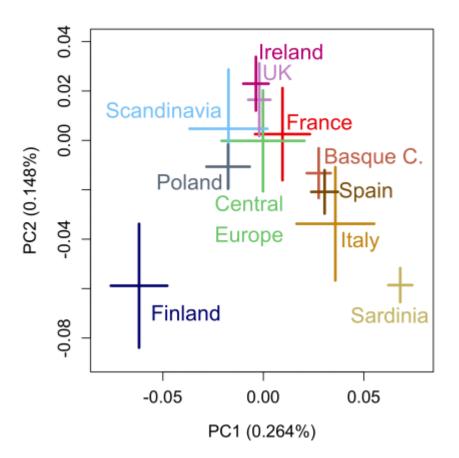


Figure S21. The two first principal components of genetic variation in the "modern merged dataset", which includes 843 French WGSs (red) for which the birth places of the four grandparents are situated in close proximity, and genome-wide data from 20 central and western European populations: Basque Country (HGDP), Belgium, Cornwall (UK, POBI), Denmark, Dyfed (UK, POBI), Finland, Germany, Gwynedd (UK, POBI), Ireland, Italy, Kent (UK, POBI), Norfolk (UK, POBI), Northern Ireland, Norway, Orkney Islands (HGDP), Poland, Sardinia (HGDP), Spain, Sweden, United Kingdom. For simpler visualisation samples from Norway, Sweden and Denmark were labelled as "Scandinavia", samples from Belgium and Germany were labelled as "Central Europe". All samples from Great Britain are represented by "UK". Only French samples from the FranceGenRef are shown in the plot. Crosses represent 2*standard deviations of the PC distribution for each group.

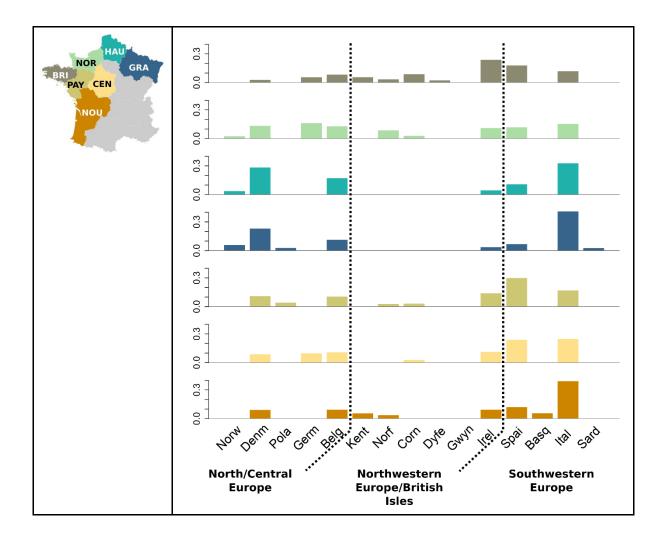


Figure S22. Ancestry profiles obtained with GLOBETROTTER for each of the French regions shown on the left panel (rows). Ancestry contributions shown in the y-axis from each of the European samples in the x-axis. French regions are coloured according to the map on the left pane. Population acronyme: BRI, Brittany; PAY, Pays-de-la-Loire; NOR, Normandy; HAU, Hauts-de-France; GRA, Grand Est; CEN, Centre-Val de Loire; NOU, Nouvelle-Aquitaine. Map generated with *R statistical package* using *sp* packages and the "*régions*" boundary dataset from the French platform of open data (<u>https://www.data.gouv.fr/</u>).

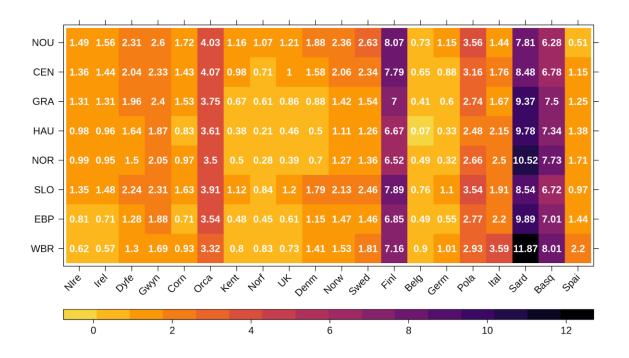


Figure S23. Pairwise average Weir and Cockerham's F_{ST}s between French and other European populations x 1000. French clusters and populations are: WBR, <u>W</u>estern <u>Br</u>ittany; EBP, <u>Eastern Brittany and Pays-de-la-Loire; SLO, South Loire; NOR, *Normandie*; HAU, Hauts-de-France; GRA, Grand Est; CEN, Centre-Val de Loire; NOU, Nouvelle-Aquitaine. Non-French population acronyms: Basque Country (Basq), Belgium (Belg), Cornwall (Corn), Denmark (Denm), Dyfed (Dyfe), Finland (Finl), Germany (Germ), Gwynedd (Gwyn), Ireland (Irel), Italy (Ital), Kent (Kent), Norfolk (Norf), Northern Ireland (NIrel), Norway (Norw), Orkney Islands (Orca), Poland (Pola), Sardinia (Sard), Spain (Spai), Sweden (Swed), United Kingdom (UK).</u>

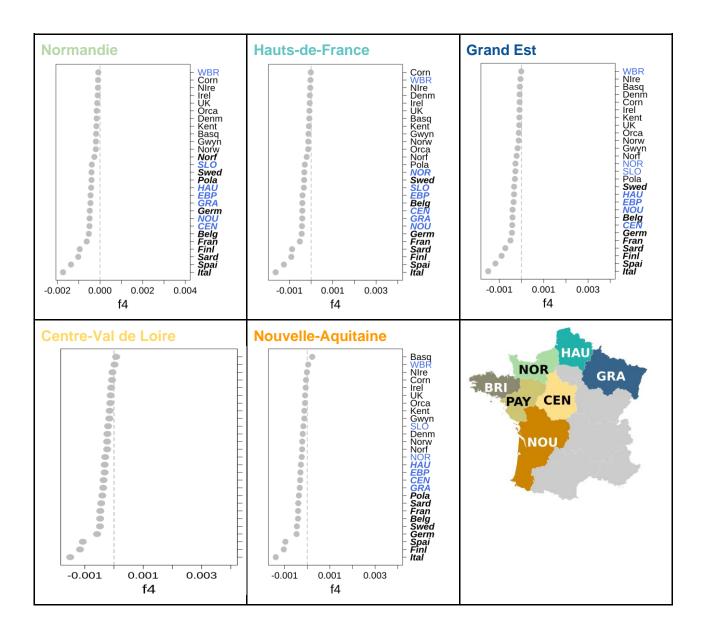


Figure S24. f_4 -statistics of the form f_4 (Mbuti, French region; Dyfed, X), where the French region is indicated on top of the plot and X is each of the populations on the right side of the plot. F_4 -statistics values are shown together with one standard deviation. French clusters and populations are: WBR, Western Brittany; EBP, Eastern Brittany and Pays-de-la-Loire; SLO, South Loire; NOR, Normandie; HAU, Hauts-de-France; GRA, Grand Est; CEN, Centre-Val de Loire; NOU, Nouvelle-Aquitaine. Ranges correspond to the standard deviation estimated from weighted block jackknife as implemented in Admixtools. Map generated with R statistical package using rgdal, sp, broom, and ggplot2 packages and the "départements" boundary dataset from the French platform of open data (https://www.data.gouv.fr/).

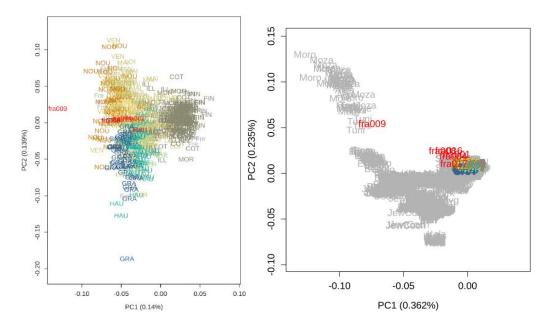
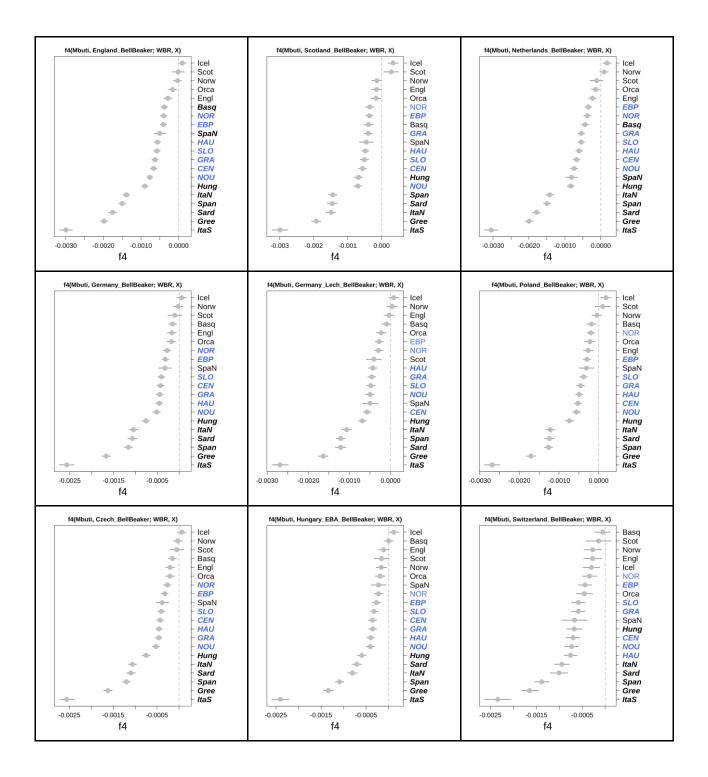


Figure S25. Mediaeval French samples projected onto the two first principal components from present-day Europeans and north Africans. Modern European genotypes present in the Human Origins Array (Reich's Lab, vs.42.2 March 2020 release, (49)) are coloured in grey. Modern French samples analysed in this study are coloured according as in Fig. S24 (above) whereas ancient samples are represented in red. North African sample acronyms: Tuni - Tunisia; Moza - Mozabites; Moro-Morocco. The pca was performed with smartpca (*Isqproject: YES*).



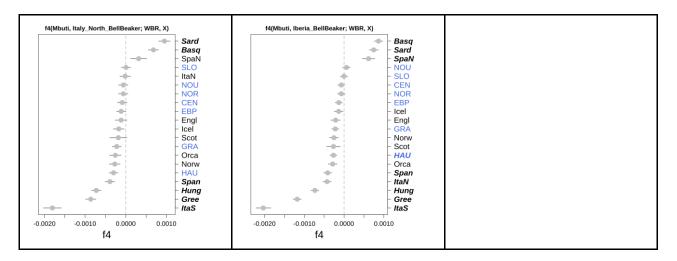


Figure S26. f_4 of the form f_4 (*Mbuti, Bell Beaker population; WBR, X*), where Bell Beaker population is indicated on top of the plot and X is each of the populations on the y-axis (right side of the plot). French populations are coloured in blue and comparisons with |Z| > 3 are indicated in bold on the y-axis. Ranges correspond to the standard deviation computed from the weighted block jackknife implemented in *Admixtools*.

Supplementary Tables

Table S1. Summary of the admixture composition, sources and times for the French regions obtained with GLOBETROTTER

	Admixture event 1				1	Admixture event 2			
French region	Best Fitting Admixture Model	Goodness of fit (R ²)	Time* of admixture in ya [95% CI]	Minor Component	Major Component	Ratio	Minor Component	Major Component	Ratio
Brittany (BRI)	One-date- multiway	0.62	1047 [858-1244]	Italy	Kent	0.32:0.68	Kent	Kent	0.43:0.57
Pays-de-la-Loire (PAY)	One-date- multiway	0.70	1337 [1163-1554]	Italy	Kent	0.49:0.51	Kent	Spain	0.48:0.52
Centre et Val-Loire (CEN)	Unclear signal	0.19	NA	NA	NA	NA	NA	NA	NA
Normandy (NOR)	One-date- multiway	0.30	1035 [733-1293]	Denmark (Denm)	Italy (Ital)	0.47:0.53	Ireland (Irel)	Norfolk (Norf)	0.24:0.76
Hauts-de-France (HAU)	One-date	0.65	1105[951-1354]	Italy	Kent	0.49:0.51	NA	NA	NA
Grand Est (GRA)	One-date	0.50	1148 [925-1438]	Kent (Kent)	Italy (Ital)	0.47:0.53	NA	NA	NA
Nouvelle-Aquitaine (NOU)	One-date- multiway	0.35	1540 [887-1914]	Kent	Spain	0.31:0.69	Kent	Italy	0.42:0.58

*Times were converted from generations to years ago using a generation time of 29 years/generation.

Samples ID	Dates*	Place	% endog.	Mapped reads	3' and 5' damage %	Average read length	Genome cov.**	Gender***	X-contam. (SE)	mtDNA contam.	total sites HOA	mean cov
fra001	375-541 cal. AD	Saint Lupien Rezé	0.199312	271,093,228	16.9-17.3	75.38	0.48	xx	NA	0.01	247020	0.551608
fra004	414-553 cal. AD	Saint Lupien Rezé	0.057764	15,241,917	14.2-15.0	72.26	0.28	XY	0.04367 (0.009)	0.02	169051	0.344645
fra008	943 - 1024 cal. AD	Chaussé Saint Pierre - Angers	0.018284	5,983,204	25.1-25.6	55.97	0.08	consistent with XY but not XX	0.22336 (0.000)	0.01	64438	0.116075
fra009	414-548 cal. AD	Chaussé Saint Pierre - Angers	0.276266	26,781,466	23.6-23.8	70.51	0.36	XY	0.03787 (0.007)	0.01	199735	0.421043
fra016	600-700 AD	Chémeré	0.480058	234,557,159	24.3-24.5	56.46	3.43	consistent with XY but not XX	0.014 (0.001)	0.01	574582	4.153501
fra017	600-700 AD	Chémeré	0.020224	6,559,927	9.1-11.1	80.88	0.14	XY	0.10887 (0.000)	0.01	92736	0.170474
* cal. AD	= Radiocarbor	n calibrated	l Anno Dor	mini (AD); A	nno Domin	i based on th	ne archaec	logical cont	ext			
** after M	apQ 30 filterin	g										
*** estima	ated biological	sex from a	lignment d	ata <u>(1)</u> .								
%endog.	- % of endoge	nous DNA										

	WBR (n=25)	EBP (n=25)	SLO (n=25)	NOR (n=19)	HAU (n=25)	GRA (n=25)	CEN (n=25)	NOU (n=25)	Mediaeval French (fra001, fra004, fra008, fra016, fra017)
England_Roman	0.9362	0.4472	0.0008	0.4504	0.3048	0.0004	0.0017	0.0000	0.1011
England_Saxon	0.0002	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0002
Germany_EMedieval	0.0744	0.1378	0.0000	0.0872	0.0054	0.0000	0.0000	0.0000	0.0756
Iberia_Celtiberian	0.0001	0.0000	0.0008	0.0002	0.0004	0.0000	0.0004	0.0028	0.0470
lberia_Medieval_publishe d	0.0000	0.0004	0.0366	0.0004	0.0009	0.0277	0.0219	0.0696	0.2434
Iberia_Carolingian	0.2219	0.4450	0.5731	0.5007	0.5409	0.4385	0.5462	0.6125	0.9324
Iberia_Visigoth_Barcelona	0.0689	0.1855	0.3636	0.1046	0.3221	0.3306	0.3678	0.4429	0.7020
Iberia_Visigoth_Girona	0.0363	0.2358	0.9075	0.3527	0.3833	0.7244	0.5947	0.7063	0.9538
Iceland_Early_Christian	0.3617	0.2317	0.0280	0.1455	0.1442	0.0442	0.0219	0.0009	0.2354
Iceland_Pre_Christian	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Italy_Medieval_EarlyMod ern_oCentralEuropean	0.0000	0.0002	0.4682	0.0003	0.0002	0.6033	0.3087	0.0695	0.8164
Italy_Medieval_EarlyMod ern	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000
Italy_North_EarlyMedieval _Langobards	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.4315
Vikings	0.0039	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0000	0.0027

Present-day population/cluster acronyms: Western Brittany (WBR), Eastern Brittany/Pays-de-la-Loire (EBP), south Loire (SLO), Normandy (NOR); Hauts-de-France (HAU); Grand Est (GRA); Centre-Val de Loire (CEN); Nouvelle-Aquitaine (NOU). Ancient sample sizes are shown in Fig. 4. Non-significant p-values are shown in bold.

Table S4. *qpAdm* results for the three-way model assuming Steppe pastoralists (SP), Early Farmers (EF) and Western Hunter-Gatherers (WHG) as sources populations of each modern and Mediaeval French populations (FRMedieval)

	SP (n=17)	EF (n=73)	WHG (n=42)				
	Prop (std. error)	Prop (std. error)	Prop (std. error)	d.f	X2	p-value(X ²)	
WBR (n=25)	0.460 (0.021)	0.436 (0.020)	0.104 (0.010)	10	77.265	1.72E-12	
EBP (n=25)	0.435 (0.021)	0.476 (0.020)	0.089 (0.009)	10	69.372	5.86E-11	
SLO (n=25)	0.372 (0.021)	0.545 (0.020)	0.083 (0.010)	10	63.73	7.09E-10	
NOR (n=19)	0.437 (0.021)	0.480 (0.020)	0.083 (0.010)	10	51.303	1.54E-07	
HAU (n=25)	0.422 (0.021)	0.483 (0.020)	0.095 (0.010)	10	67.78	1.19E-10	
GRA (n=25)	0.389 (0.020)	0.548 (0.019)	0.063 (0.009)	10	64.295	5.53E-10	
CEN (n=25)	0.384 (0.020)	0.538 (0.020)	0.079 (0.010)	10	70.677	3.28E-11	
NOU (n=25)	0.329 (0.019)	0.578 (0.019)	0.093 (0.009)	10	59.818	3.92E-09	
Mediaeval French (n=5)	0.326 (0.057)	0.608 (0.054)	0.067 (0.029)	10	9.248	0.508768613	

	 f₄-statistics of the form f₄(Mbut French population) 	i; anciel	nt EUR s	ample; V	VBR, oti	her		
Pop1 (W)	Pop2 (X) :	Pop3 (Y)	Pop4 (Z)	D-stat	Z	BABA	ABBA	# of SNPs
Mbuti	Russia_EBA_Yamnaya_Samara	WBR	NOU	-0.0091	-11.381	27968	28481	464380
Mbuti	Russia_EBA_Yamnaya_Samara	WBR	CEN	-0.007	-8.586	28029	28422	46438
Mbuti	Russia_EBA_Yamnaya_Samara	WBR	SLO	-0.0069	-8.816	28020	28407	46438
Mbuti	Russia_EBA_Yamnaya_Samara	WBR	GRA	-0.0058	-7.646	28068	28398	46438
Mbuti	Russia_EBA_Yamnaya_Samara	WBR	HAU	-0.0053	-7.279	28085	28383	46438
Mbuti	Russia_EBA_Yamnaya_Samara	WBR	NOR	-0.0041	-4.895	28093	28326	464380
Mbuti	Russia_EBA_Yamnaya_Samara	WBR	EBR	-0.0041	-5.3	28108	28341	464380
Mbuti	Germany_EN_LBK_published	WBR	HAU	-0.0004	-0.47	28132	28152	463062
Mbuti	Germany_EN_LBK_published	WBR	NOR	-0.0003	-0.429	28103	28122	463062
Mbuti	Germany_EN_LBK_published	WBR	EBR	0.0003	0.37	28139	28123	463062
Mbuti	Germany_EN_LBK_published	WBR	GRA	0.0013	1.678	28189	28115	463062
Mbuti	Germany_EN_LBK_published	WBR	CEN	0.0021	2.66	28213	28096	463062
Mbuti	Germany_EN_LBK_published	WBR	NOU	0.0025	3.267	28231	28089	463062
Mbuti	Germany_EN_LBK_published	WBR	SLO	0.0026	3.384	28218	28070	463062
Mbuti	lberia_EN	WBR	HAU	-0.0009	-1.089	27436	27484	452920
Mbuti	Iberia_EN	WBR	EBR	-0.0001	-0.179	27443	27451	452926
Mbuti	Iberia_EN	WBR	NOR	0.0006	0.721	27451	27417	452926
Mbuti	Iberia_EN	WBR	GRA	0.0013	1.654	27508	27434	452926
Mbuti	Iberia_EN	WBR	CEN	0.0015	1.831	27512	27428	452926
Mbuti	Iberia_EN	WBR	SLO	0.0021	2.638	27515	27402	452926
Mbuti	lberia_EN	WBR	NOU	0.0032	3.944	27569	27392	452926
Mbuti	Luxembourg_Loschbour_published.DG	WBR	GRA	-0.0064	-5.314	28402	28769	466881
Mbuti	Luxembourg_Loschbour_published.DG	WBR	NOU	-0.0041	-3.444	28477	28713	466881
Mbuti	Luxembourg_Loschbour_published.DG	WBR	CEN	-0.0034	-2.883	28494	28689	466881
Mbuti	Luxembourg_Loschbour_published.DG	WBR	HAU	-0.003	-2.565	28504	28677	46688 ⁻
Mbuti	Luxembourg_Loschbour_published.DG	WBR	SLO	-0.0026	-2.17	28513	28661	46688
Mbuti	Luxembourg_Loschbour_published.DG	WBR	NOR	-0.0021	-1.702	28508	28627	46688
Mbuti	Luxembourg_Loschbour_published.DG	WBR	EBR	-0.0019	-1.557	28533	28641	46688

Table S5 f statistics of the form f (Mbuti: ansight EUD complex M/PD, other

Note: significant values (|Z| > 3) are indicated in bold; |Z| > 4 are indicated in yellow and |Z| > 36 are indicated in red.

Table	S6 - F4-statistics of th	e form <i>f₄(M</i>	buti; Mega	alithic samp	oles; WE	BR,		
other Pop1 (W)	modern French popula Pop2 (X) :	etions) Pop3 (Y)	Pop4 (Z)	D-stat	Z	BABA	ABBA	# of SNPs
Mbuti	Ireland_Megalithic.SG	WBR	English	-0.000207	-1.648	28628	28726	471404
Mbuti	Ireland_Megalithic.SG	WBR	HAU	-0.000153	-1.59	28687	28759	471404
Mbuti	Ireland_Megalithic.SG	WBR	GRA	-0.000106	-1.092	28705	28755	471404
Mbuti	Ireland_Megalithic.SG	WBR	NOR	-0.000054	-0.527	28681	28707	471404
Mbuti	Ireland_Megalithic.SG	WBR	EBP	-0.000031	-0.325	28707	28722	471404
Mbuti	Ireland_Megalithic.SG	WBR	SLO	0.00009	0.916	28745	28703	471404
Mbuti	Ireland_Megalithic.SG	WBR	CEN	0.000095	0.96	28759	28714	471404
Mbuti	Ireland_Megalithic.SG	WBR	NOU	0.000196	2.023	28788	28696	471404
Mbuti	Scotland_Megalithic.SG	WBR	HAU	-0.000144	-1.007	9333	9355	154275
Mbuti	Scotland_Megalithic.SG	WBR	GRA	-0.000104	-0.72	9340	9356	154275
Mbuti	Scotland_Megalithic.SG	WBR	English	-0.000133	-0.684	9317	9338	154275
Mbuti	Scotland_Megalithic.SG	WBR	NOR	-0.000088	-0.577	9328	9342	154275
Mbuti	Scotland_Megalithic.SG	WBR	EBP	-0.000079	-0.555	9332	9344	154275
Mbuti	Scotland_Megalithic.SG	WBR	CEN	-0.000014	-0.091	9350	9352	154275
Mbuti	Scotland_Megalithic.SG	WBR	SLO	0.000103	0.695	9350	9334	154275
Mbuti	Scotland_Megalithic.SG	WBR	NOU	0.000316	2.098	9376	9327	154275

Note: Significant differences in allele sharing would result in |Z| > 3, which was not found for any of the above comparisons.

	S7 - F ₄ -statistics of the modern French populati	•	ıti; ancien	nt EUR sa	ample; V	VBR,		
Pop1 (W)	Pop2 (X) :	Pop3 (Y)	Pop4 (Z)	D-stat	Z	BABA	ABBA	# of SNPs
Mbuti	Germany_CordedWare	WBR	GRA	-0.00640	-8.235	28300	28666	466547
Mbuti	Germany_CordedWare	WBR	NOR	-0.00480	-5.766	28320	28592	466547
Mbuti	Germany_CordedWare	WBR	CEN	-0.00730	-9.113	28269	28687	466547
Mbuti	Germany_CordedWare	WBR	EBR	-0.00460	-5.987	28343	28604	466547
Mbuti	Germany_CordedWare	WBR	SLO	-0.00650	-8.191	28285	28653	466547
Mbuti	Germany_CordedWare	WBR	NOU	-0.00880	-10.708	28224	28724	466547
Mbuti	Germany_CordedWare	WBR	HAU	-0.00620	-8.223	28302	28658	466547

Table	S8- f ₄ -statist	ics of the fo	orm f ₄ (Mbuti; mode	rn French p	opulatic	on;		
Iberiar	n Early Neoli	thic, Early	Neolithic LBK)					
Pop1 (W)	Pop2 (X) :	Pop3 (Y)	Pop4 (Z)	D-stat	Z	BABA	ABBA	# of SNPs
Mbuti	EBP	Iberia_EN	Germany_EN_LBK_pu blished	-0.000084	-0.43	26520	26558	44769
Mbuti	WBR	Iberia_EN	Germany_EN_LBK_pu blished	-0.000141	-0.716	26504	26567	44769
Mbuti	NOR	Iberia_EN	Germany_EN_LBK_pu blished	-0.000251	-1.257	26470	26583	44769
Mbuti	HAU	Iberia_EN	Germany_EN_LBK_pu blished	-0.000063	-0.317	26501	26529	44769
Mbuti	GRA	Iberia_EN	Germany_EN_LBK_pu blished	-0.000136	-0.692	26512	26572	44769
Mbuti	SLO	Iberia_EN	Germany_EN_LBK_pu blished	-0.000056	-0.279	26529	26553	44769
Mbuti	CEN	Iberia_EN	Germany_EN_LBK_pu blished	-0.000076	-0.388	26523	26557	44769
Mbuti	NOU	Iberia_EN	Germany_EN_LBK_pu blished	-0.000214	-1.098	26493	26589	44769

Note: Significant differences in allele sharing would result in |Z| > 3, which was not found for any of the above comparisons.

Supplementary Notes

Supplementary archaeological details

Rezé (Loire-Atlantique) - Saint-Lupien area

Rezé is located in the *département* of *Loire Atlantique* on the southern shore of the Loire river facing the city of Nantes to the north. It is located at ~55km from the Atlantic coast. The excavation was carried out by the team of Mikaël Rouzic between 2005 and 2016 and the archaeological description of the sites remains unpublished.

<u>Site:</u> MR 4825 <u>Samples:</u> SP 4903 (fra001) <u>Radiocarbon dating:</u> 375-541 cal AD

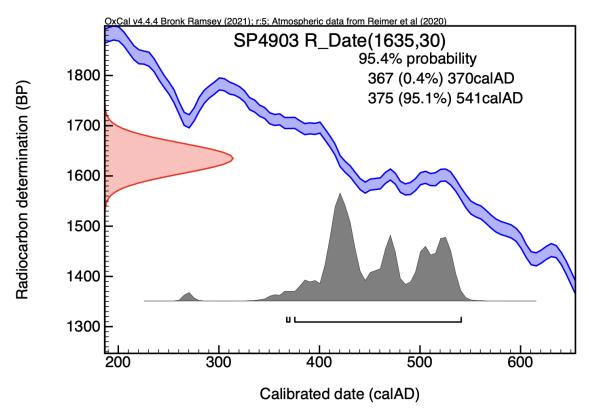


Figure S27: age graph of individual fra001 radiocarbon dating.

The wall of the site MR 4825 was pitted on several layers in order to install the SP 4903 grave with no signs of excavation. The grave was filled with a compact and heterogeneous brown silt, very rich in TCA, lime, gravel and coal. The grave contained the individual SP4903 and two ceramic shards were found inside. The individual was an adult who died at an age of 20-59 years.

<u>Site:</u> US 20380; US 20381; US 20382; US 20383 <u>Samples:</u> SP 20380 (fra004) <u>Radiocarbon Dating:</u> 414-553 cal AD

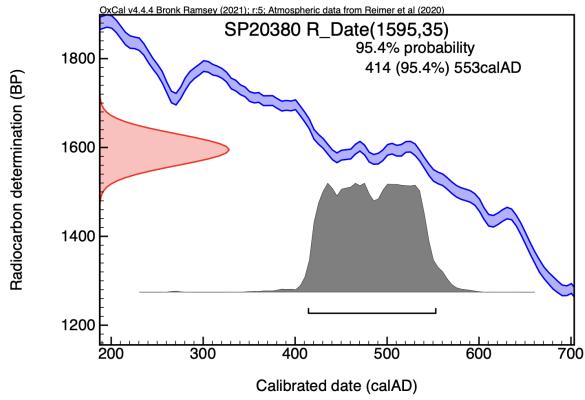


Figure S28: age graph of individual fra004 radiocarbon dating

The site consists of a 2.40 m long and 0.72 m wide burial pit, oriented east/west. It was dug in the ancient road and lies on top of an ash and charcoal layer. The fill is a compact and heterogeneous medium brown sandy loam containing numerous inclusions of architectural terracotta fragments, small blocks, some mortar fragments, several shards of ancient ceramics, glass and faunal bones. Six nails and eight shanks of carpentry nails were taken from the entire thickness of the filling. The skeleton remnants found inside belonged to a male individual 20-29 years old at death.

Chéméré (Loire-Atlantique)

<u>Samples:</u> SP 46 and SP 55 (fra016 and fra017) <u>Dates based on the archaeological context:</u> 600-700 AD

The Merovingian necropolis of Chéméré (*Loire Atlantique*, south Nantes) is located in an area peripheral to the present village. Its occupation is dated to the 6th-7th centuries by the furniture deposited in the tombs and by radiocarbon-based analyses. The total surface area of the burial space is estimated to be around 7700 m². Approximately 1500 m² of land containing 416 tombs have been cleared since the 1960s. The latest excavations in 2007 yielded a population sample of 181 individuals (155 adults and 26 children) recognised by anthropological analysis as a morphologically homogenous group. The two men 46 and 55 are from this group. They were buried in the same row, at the southern end of the cemetery.

Angers (Maine-et-Loire) - Chaussé Saint Pierre

Samples: SP 1046 and SP 1049A (fra008 and fra009) Radiocarbon calibrated (cal.) dates: 943 - 1024 cal. AD and 414-548 cal. CE

The site of Chaussé Saint-Pierre is located right in the modern city-center of Angers. The city of Angers is located on the intersection of Maine and the Loire rivers and on the northern shore of the latter. It is situated approximately 100km upstream from Nantes. The excavations in this site, which were led by the team of Martin Pithon, from the INRAP of west France, took place between July and August 2009.

The large density of archaeological remains around the Chaussé Saint-Pierre suggests it was densely occupied from the first period of the Roman Empire (27 BCE-250 CE). Until the 4th century CE the area was occupied by commercial buildings and thermes, nevertheless by the 5th century archaeological remains associated with a necropolis indicate a shift in the pattern of occupation. The excavation of Rue Chaussée-Saint-Pierre focused on the north-eastern end of the hopper where the project was the deepest and where the remains were best preserved. The excavations investigated a stratigraphic sequence of almost 1 m thick between the modern-day earthworks and the natural terrain over an area of ~100 m². The analysis of this sequence shows five main phases of occupation which lie within a chronological interval between the beginning of the Roman Empire (20 BC to 15 AD) and the modern period. Funerary-associated occupation begins in phase three.

SP 1049A (fra009)

The burial SP 1049 is among the earliest among those belonging to the phase three of occupation and the dating ranges from the beginning of the 5th century to the first half of the 6th century. This tomb, which has been badly damaged by Medieval/modern construction, seems to be part of a homogeneous group of six burials very similar in nature. The dating of the tomb shows repeated burial and skeletal remains indicate the presence of a total of three individuals, two adults and one child. The child's skeleton and one skull not belonging to the child have been dated to 401-539 cal AD and 414-548 cal. AD, respectively (https://dolia.inrap.fr/flora/ark:/64298/0121736 in French). The skeleton remains indicate the child was buried at the age of 1-4 years old. The other skeletal remains seem to belong to a female and an individual of indeterminate gender aged of >25 and >30 years, respectively. The time proximity of the burials within the tomb are likely indicative of a voluntary grouping and may represent a family management of the deposits.

SP 1046 (fra008)

The tomb SP 1046 together with a set of the other tombs dating from the 13-15th century CE indicate the practice of reuse of the tombs. The burial site is composed of heterogeneous loose deposits of fine sandy sediment including charcoal, fragments of tiles/bricks, blocks of slate shale as well as a batch of ceramic sherds mixing of Antiquity and Medieval (High Middle Ages? and Classical Middle Ages) elements. A full skeleton (fra008), belonging to a male aged >30 years, and other skeletal remains likely to come from three different individuals were buried in this site. Radiocarbon calibrated dating was retrieved under the scope of our study and indicates the skeleton dates to 943 - 1024 cal. AD.

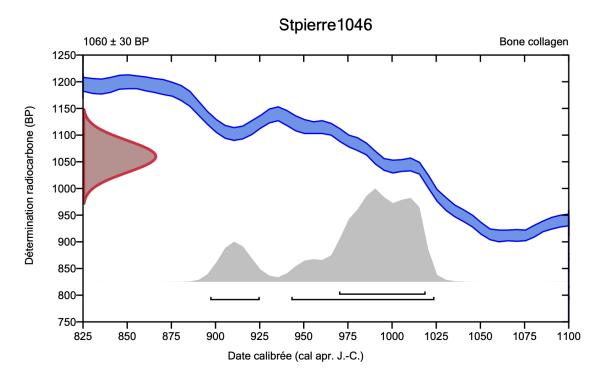


Figure S29: age graph of individual fra008 radiocarbon dating

Supplementary Discussion

1. Geolinguistics and genetic clusters

Below we describe in detail the overlap we found between dialectal areas and genetic clusters. However, these are not considered as biological relationships as they might reflect long-term inertia and socio-economic complex and cultural factors. Below the linguistic data come from the *Nouvel Atlas Linguistique de Basse-Bretagne* (1) and the places' names are extracted from the IGN BDtopo® database. A deeper understanding of such overlaps requires a higher resolution with respect to the genetic data.

1.1. Cluster "Bretagne-Centre", cluster "Cornouaille" and cluster "Vannes" together

The cluster "Bretagne-Centre" overlaps with the dialectal area featured by the usage of two initial consonants: the aspirated [h] instead of an unaspirated and the alveolar fricative [z] instead of [s], stretching respectively from the northern coast to the southern sea-side of the central area (Fig. S12). The river Blavet overlaps with the south-eastern boundaries of the dialectal areas and it separates, to some extent, the cluster "Bretagne-Centre" from the cluster "Vannes" (Fig. S14).

Fig. S12 'h-' shows seven analysed NALBB maps considering the treatment of the initial [h]. Fig. S12 'z-' represents the area where the initial [s] is voiced as [z]. The [z] area extends in a central crossing zone from the North to the South. It gradually disappears towards the southwest¹.

These two phonological features extend over an area between the southern coast of Morbihan and the north of the Côtes-d'Armor, excluding a north-western and a south-eastern area without the initial [h] and where the initial [s] is pronounced. Such phonological regions match with the distribution of "Bretagne-Centre" and "Cornouaille" clusters. The 'z-' map seems to correlate better than the 'h-' one. The articulation [z] for the initial [s] is also observed on the other side of the Channel, in the south-western dialects of England: *zome* 'some', *Zomerzet* 'Somerset', *to zwaip* for *to* sweep' fr. 'balayer', *zwell* for 'swell', *zwate / zweet* 'sweet' fr. 'doux' (Tristram 1995, Markus 2021). At this point, it would be interesting to compare the data from the People of the British Isles project (2,3) which shows a differentiation between the Cornwall and Devon clusters.

1.2. Cluster "Cornouaille"

If we would look for a coherence between the distribution of dialectal features in Lower-Brittany and "Cornouailles" cluster, we could consider the palatalization of -*h*- [h] into -*y*- [j] given their broad overlap (Fig. S13). This phenomenon can be found in the following words: *meryed* "girls", *luyadenn* "lightning", *luyed* "lightnings" and *meryer / dimeryer* 'Wednesday (noun/adv)'.

¹ A first analysis of the initial [z], according to NALBB data, was carried out by François Falc'hun, 1981, *Perspectives nouvelles sur l'histoire de la langue bretonne*, Union Générale d'Editions, fig. 67 p. 204.

1.3. Clusters 'Léon', 'Cornouaille', 'Bretagne-Centre', 'Vannes' and 'Guérande' and the Celto-Romance limit in eastern Brittany

All together the clusters "Léon", "Cornouaille", "Bretagne-Centre", "Vannes" and "Guérande" correlate with the Celto-Romance limit attested between the 16th and the 18th centuries. The distribution of place-names Ker- attests the progressive withdrawal towards the west of this eastern limit of Celtic dialects (Fig. S26). According to Bernard Tanguy, the oriental limit of toponyms Ker- (Fig. S30) does not show the maximum extension of Breton spoken varieties but testifies the retreat of Armorican Gaulish dialects facing the gradual advance of the Gallo-Romance dialects in formation during the Early Middle Ages (4). The same toponymic element can be found on both sides of the Channel spelled *Ker-*, *Car-*, *Gear* in Cornwall (Joanne Pye 2019), and *Caer* in Wales (National Library of Wales 2021), Great Britain. So far, there is no evidence that the presence of *Ker-* place names in Brittany is due to a migration from Great Britain. If one considers the density of place-names as indicating a point of departure, it would indicate the opposite. At this stage it is more cautious to simply consider this toponymic element as a celtic linguistic and cultural mark.

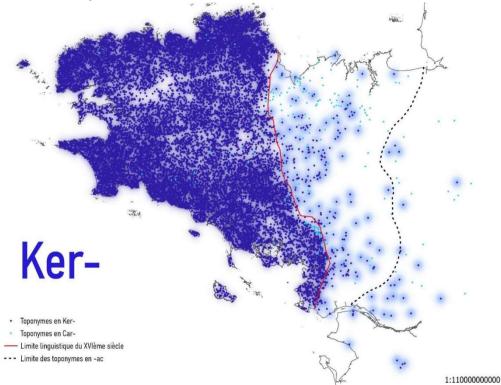


Figure S30. Distribution of Ker- "inhabited place". Data : BDtopo®, National Geographic Institute (IGN). *Ker-* (royal blue) and *Car-* (turquoise) toponyms.

2. The role of rivers in the distribution of the 18 clusters

Rivers and streams could play a role in the diffusion of linguistic features. In France, the Loire initially formed an important border between the Oïl Gallo-Romance dialects in the North, and the Oc Gallo-Romance dialects in the South. Later, it became a vector for the diffusion of new traits coming from the North to the inner country (5). In Lower Brittany, on a smaller scale, the river Blavet seems to be at the origin of a bundle of isophones separating the different

treatments of the interdental spirants (6). The linguistic frontier separating the Celtic and Gallo-Romance areas is not determined by a natural barrier. Nevertheless, the hydrography seems to mark the territories within these zones. By using a geographical information system and the file of the main rivers proposed by the IGN's BDcarthage®, we assessed their role in the organisation of the k=18 genetic clusters.

We first discuss the rivers which seem to correspond to the limits of the genetic clusters. Cluster "Léon " is bordered in the north-east by the River of Morlaix and the Jarlot. In the South, the same cluster is separated from cluster "Cornouaille" by the river Aulne. The Laïta, extended by the Ellé until the north of Le Faouët, seems to border the cluster "Cornouaille". The Carnoët forest could have reinforced its natural limit in the South. Cluster "Vannes" is enclosed on its western side by the Blavet and on its north side by the Oust. Cluster "Nantes" is circumscribed by the Semnon in the north and the Vilaine in the west. Cluster "Malo-Rennais" is surrounded by the Gouessant, then by the Yvel and the Hivet in the west, by the Canut and the Semnon further south. Finally, the Loire delimits the clusters "Guérande", "Nantes" and "Ancenis" in the north and the clusters "Retz", "Mauges 3" and "Mauges 1" in the south. In South Loire Vendée, we observe a role in the cluster landscape for rivers like "Le Lay" and "Sèvre Nantaise", although it appears to be less clear. The "Maine-Anjou" cluster is crossed by several rivers and, when focusing at a finer, 78 clusters, scale, we also observe structuration along rivers like Mayenne, Vègre and Loir (data not shown).

The difference between navigable and non-navigable waterways does not seem to be decisive when considering the role of rivers by hampering gene flow. However, it has to be taken into account when studying their role as vectors. The geographical distribution of "Maine-Anjou" includes the rivers Loire, Sarthe, Mayenne and Oudon, all of them are navigable waterways according to the *Service d'administration nationale des données et référentiels sur l'eau* (Sandre and IGN 2017). The cluster "Guérande" seems to be contained by the Vilaine mainly, the Oust, the Arz and the Brivet in the south. When we consider river geography, the distribution of clusters seems to be more complex than a simple cluster/bishoprics correlation.

3. Computer simulations to access the power to detect a short bottleneck with IBDNe

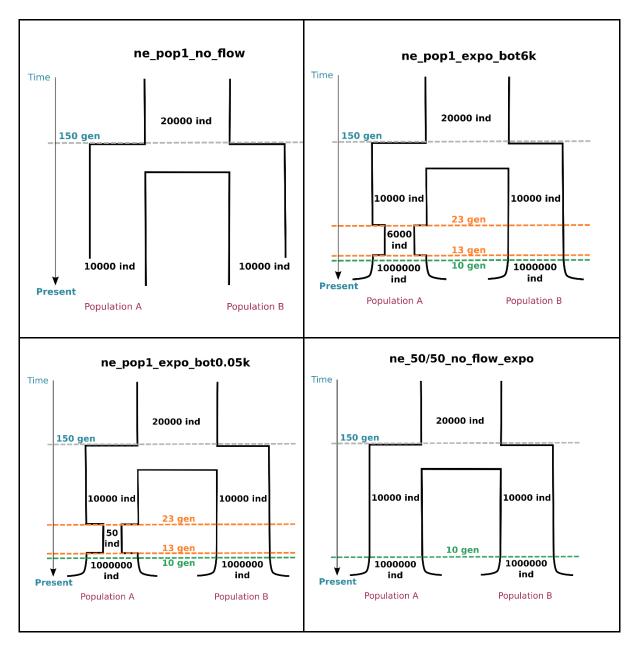
Identity-by-descent (IBD) sharing in population samples can be used to estimate recent effective population size changes. IBDNe (7) assumes an idealised random mating population with a constant effective size that has similar random changes in allele frequencies over time. Shorter IBD segments represent coalescent events that occurred further back in time, while longer segments represent coalescent events that occurred in the past few generations. If the number of IBD segments is high, a larger number of coalescent events have occurred, indicating a high coalescence probability meaning a low effective size. Similarly, if the number of IBD segments is low, the effective size is high.

The effective population size trajectories inferred in this study for the three main genetic clusters inferred with fineSTRUCTURE indicate a slight and short population decline, starting around the years 1,230-1,350 CE (13 generations and assuming a generation time ~29 year/generation) and lasting for almost ~300 years (~10 generations). Given the assumption of random mating within samples and our knowledge of extensive fine-scale structure within Northwestern France, this motivated us to perform a simulation study to assess whether a bottleneck could be detected if the evolutionary history of the population involves other scenarios other than a bottleneck.

More specifically, we investigated: 1) The performance of IBDNe in detecting a recent

bottleneck; 2) The effect of having admixture (punctual migration), sample structure and exponential growth in the estimation.

We used ARGON (8), a discrete time Wright-Fisher model simulator. Data was simulated for 50 diploid individuals (per population) according to a model containing 2 populations (see below). The data was simulated for each chromosome according to its respective size and we used the human recombination map (1000Genomes project). We ran the IBDNe software on our data using only IBD segments greater than 4cM. Simulated scenarios are shown below (Fig. S31).



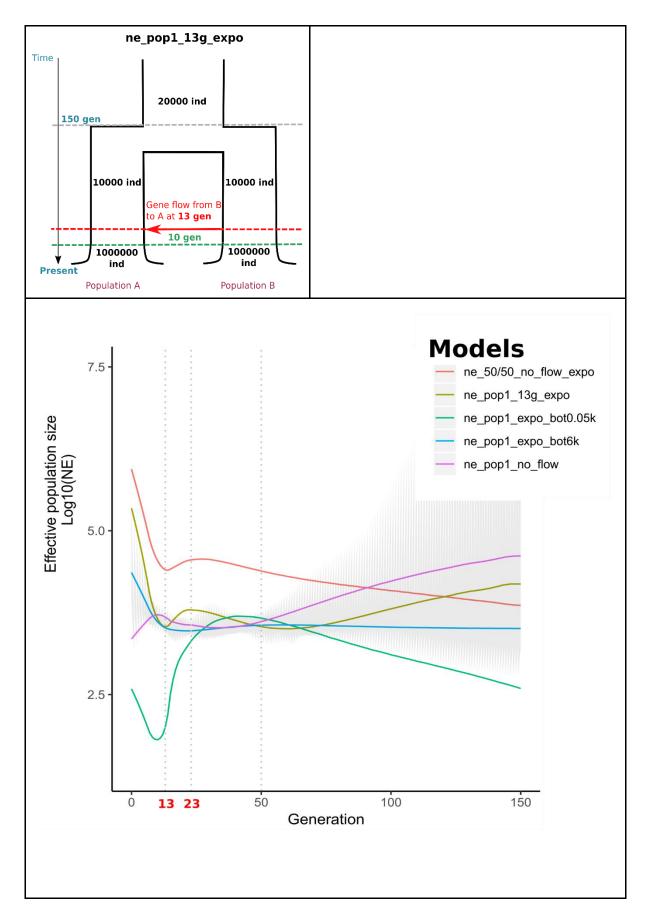


Figure S31. Simulation study performed to assess the power of IBDNe to infer shortlasting bottlenecks. We simulated models involving two populations that diverged 150

generations ago. Ne trajectories were inferred with IBDNe only from Population 1. The upper left panel represents the simplest model, in which after divergence Population 1 and 2 kept constant *Ne*. ne_pop1_expo_bot6k and ne_pop1_expo_bot0.05k models are variations of the simplest model, in which Population 1 undergoes 10 generations a long bottleneck after diverging from Population 2. Bottleneck starting times were set to 23 generations and the reduction in *Ne* were set to 6000/10000 and 50/10000, respectively. In model ne_50/50_no_flow_expo, Population 1 did not suffer a reduction in *Ne* but the sample analysed with IBDNe resulted from randomly sampling 50 individuals from both Population 1 and 2 in order to mimic population structure within the sample. Finally, the model ne_pop1_13g_flow_expo does not include a bottleneck but rather a one-generation event of gene flow. According to this model at generation 13 Population 1 has 10% of its lineages going to Population 2 (backwards in time). All models but the simplest one (upper left panel) include exponential growth in the last 10 generations as previously inferred in European populations (7,9).

We see that both the presence of stratification within the analysed sample represented here by the model ne_50/50_no_flow_expo (coral, Fig. S31) and the presence of admixture at generation 13 represented here by the model ne_pop1_13g_expo (kakhi, Fig. S31) induces a signal of a slight bottleneck similar to that found in the real dataset, with the drop in Ne and its recovery takes place in ~10 generations. In the case of an actual bottleneck, with a severe bottleneck represented here by the model ne_pop1_expo_bot0.05k (green colour, Fig. S31) and the bottleneck model (ne_pop1_expo_bot6k, turquoise colour, Fig. S31) with a reduction in size of ~1/3 of the original Ne, which is thought to have occurred during the black death period, we see that the actual start of the bottleneck is offset compared to the real timeframe (starting 23 generations ago). Therefore, we caution that short-lasting bottleneck signals from IBDNe trajectories may represent population structure or recent gene flow rather than a real bottleneck, which are inferred to exhibit a more progressive decrease and recovery than the reality.

4. References "merged ancient dataset"

The "merged ancient dataset" contains a subset of ancient (10–83) (84–90) and present-day samples (91–93) from the Allen Ancient DNA Resource (AADR) available in: <u>https://reich.hms.harvard.edu/allen-ancient-dna-resource-aadr-downloadable-genotypes-present-day-and-ancient-dna-data</u>. To the ancient samples we also added a set of ancient Vikings (94).

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