# Phylogenetic history of patrilineages rare in northern and eastern Europe from large-scale re-sequencing of human Y-chromosomes

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## Abstract

The most frequent Y-chromosomal (chrY) haplogroups in northern and eastern Europe (NEE) are well-known and thoroughly characterized. Yet a considerable number of men in every population carry rare paternal lineages with estimated frequencies around 5%. So far, limited sample-sizes and insufficient resolution of genotyping have obstructed a truly comprehensive look into the variety of rare paternal lineages segregating within populations and potential signals of population history that such lineages might convey. Here we harness the power of massive re-sequencing of human Y chromosomes to identify previously unknown population-specific clusters among rare paternal lineages in NEE. We construct dated phylogenies for haplogroups E2-M215, J2-M172, G-M201 and Q-M242 on the basis of 421 (of them 282 novel) high-coverage chrY sequences collected from large-scale databases focusing on populations of NEE. Within these otherwise rare haplogroups we disclose lineages that began to radiate ~1-3 thousand years ago in Estonia and Sweden and reveal male phylogenetic patterns testifying of comparatively recent local demographic expansions. Conversely, haplogroup Q lineages bear evidence of ancient Siberian influence lingering in the modern paternal gene pool of northern Europe. We assess the possible direction of influx of ancestral carriers for some of these male lineages. In addition, we demonstrate the congruency of paternal haplogroup composition of our dataset with two independent population-based cohorts from Estonia and Sweden.



**Keywords:** Human population genetics, Y chromosome variety, North-East Europe, Y rare haplogroups


## Introduction

Genetic studies investigating uniparental and fine-scale autosomal variation in Estonia [1] and in its neighboring populations in NEE [2–6] observed that the regional genetic structure correlates closely with geography. In addition, recent ancient DNA studies have begun to uncover the settlement history of NEE, which is distinct from that of central and southern parts of the continent [7–9].

The four most common chrY haplogroups (hgs) with incidence above 5% (R1a-M198, N3-TAT, I-M170, R1b-M343) constitute over 90% of the chrY pool in NEE [3, 10–12]. Several studies have analysed these hgs in a wide phylogeographic context.

Besides the four most common hgs, several paternal lineages belonging on the basic level to hgs E2, J2, G and Q with frequency up to 5%, complement the pool of Y-chromosomes in NEE [3, 4, 13–15]. In Europe, hgs E2a, J2 and G are common in the southern Mediterranean populations and form 20-30 % of their chrY lineages. In NEE, the frequency of hg E2a'd is ~2-3%, hgs J2 and G respectively reach ~1-2% and ~1% of the total pool of chrY lineages [3, 5, 6, 15, 16]. Hg Q has a frequency of 1-3% in most European populations with the highest incidence in Sweden [3, 4]. Hg Q is otherwise widely spread in Siberian populations and is among the major founding male lineages in the peopling of the Americas [17, 18]. These rare hgs that make up less than 10% of NEE male lineages, are mostly left unexplained and are often regarded as recent scattered entries into populations. The small sample sizes and low phylogenetic resolution has not allowed separation of rare lineages beyond the major hg labels. The sequencing of complete Y-chromosomes provides a way to resolve the inner structure of lineages on the phylogenetic tree regardless of their prevalence in populations [13, 14, 19, 20]. Sequencing a considerable number of well dispersed samples from NEE

reveals the distribution of rare lineages on the entire phylogenetic tree and provides sufficiently granular data to estimate their split times. This builds the necessary geographic and chronological context for surveying patterns of uncovered lineage clusters stemming from a single node and hallmarking local expansions. The coalescence ages of ancestral internal nodes and phylogenetically well-defined clusters nested within disclose the geography and timeframe of local expansions as well as possible gene flow involving ancestral carriers of rare male lineages in Estonia, Sweden and their neighbouring populations.

Here we aim to analyze the previously understudied rare chrY lineages with a focus on Estonia and Sweden together with their NEE neighbors and Germany to account for the historic influence of the Baltic Germans. Additional populations are included to widen the geographic context. We combined full sequences of Y-chromosomes from populations inhabiting Estonia, Sweden, Finland, Latvia, Lithuania, Poland, Germany, Ukraine and the Russian Federation to build updated phylogenetic trees for haplogroups rare in NEE. In order to mitigate sampling bias that might influence any conclusions drawn from such a rare substratum present among the populations, we tested the representativeness of our two largest cohorts sampled from the Estonian and Swedish populations by comparing their frequency compositions with sample sets independently obtained from the same two populations.

## Materials and methods

### Samples

We screened the occurrence of rare hgs in a sample of 1,160 chrY sequences from male donors (selected randomly by county of birth) from the population-based Estonian

97 Biobank [21]. The Estonian chrY sequences are part of the whole genome sequencing
98 (WGS) data set autosomally first described in Mitt et al. (2017) [22] for constructing a
99 population-specifc imputation panel. Only chrY sequences of the haplogroups rare in
100 NEE (N=64) are included in the current study. Next, in scientific collaboration with the
101 commercial genetic testing company Gene by Gene (Houston, Texas, USA), we
102 screened the collection of customers who had provided informed consent for their data
103 to be used in scientific inquiry. This resulted in a total of 2018 male donors with self-
104 reported ancestry from Sweden, Finland, Latvia, Lithuania, Poland, Germany, Ukraine
105 and the Russian Federation. If the database contained more than 500 samples from a
106 respective country, individuals with identical self-reported paternal and maternal origin
107 were preferably selected. In case of smaller available sample sets, all samples with self-
108 reported paternal origin from the respective country were selected. From the resulting
109 set of 2,018 samples, we detected 222 Y-chromosomes belonging to the rare NEE
110 haplogroups and these samples were included in the current study. We collected
111 additional 139 chrY sequences from published sources resulting in the final set of 421
112 chrY sequences (Supplemental Table S1) used for reconstruction of phylogenetic trees
113 for rare hgs E2 (129 samples), J2 (136 samples), Q (83 samples) and G (71 samples)
114 (Figures 1-2 and Supplemental Figures S1-S7).

115 To test for possible sampling bias in the two largest sequencing cohorts, we screened
116 the haplogroup frequencies of two independent datasets – a total of 505 chrY sequences
117 available from the SweGen project (samples specifically selected to be representative of
118 the historic Swedish population [23]) and a randomly selected non-overlapping set of
119 genotyped 7,949 Estonian male donors from the Estonian Biobank.

## Data availability

The Estonian WGS data are available on demand through the Estonian Biobank: https://www.geenivaramu.ee/en/biobank.ee/data-access. In accordance to the consent form signed by the customers of Gene by Gene commercial genetic testing company, the sequencing data included in this study is used for the sole purpose of scientific inquiry and is reported here on an aggregate level in the form of phylogenetic trees. For both the Estonian Biobank and the Gene by Gene samples, summary-level data including variable positions and their frequency in the cohort population have been deposited to dbSNP with links to BioProject accession number PRJNA718714 in the NCBI BioProject database (https://www.ncbi.nlm.nih.gov/bioproject/). The Swedish data from the SweGen Project is available upon request from the original authors of the project [23].

## Sequencing, mapping and genotyping

ChrY sequences from the Estonian Biobank and the SweGen project were generated with Illumina Inc. (Illumina, San Diego, CA, USA) using HiSeq instruments (PCR-free protocol) and targeted 30x genome-wide coverage. The personal genetic testing company dataset was generated using the proprietary BigY Illumina-based targeted chrY capture sequencing service (https://learn.familytreedna.com/wp-content/uploads/2014/08/BIG_Y_WhitePager.pdf).

We used the same processing pipeline for all Illumina data. Fastq files were mapped with BWA-MEM (v0.7.12) [24] on the human reference hs37d5 (http://ftp.1000genomes.ebi.ac.uk/vol1/ftp/technical/reference/phase2_reference_assembly_sequence). Read duplicates were removed with Picard (v2.12.0) (http://broadinstitute.github.io/picard/) and remaining unique reads realigned around known indels, followed by base quality score recalibration (BQSR) using GATK (v3.8) [25]. Variant calling was performed with GATK tool HaplotypeCaller in haploid mode. All-sites VCF files were filtered with bcftools (v1.9) [26]. The Illumina data and previously filtered data from Complete Genomics (Supplemental Table S1) were merged with CombineVariants from GATK (v3.8) [25]. We extracted the effective overlap between the two datasets by masking out all positions with 5% or higher proportion of missing genotypes in either Illumina or Complete Genomics datasets. We additionally excluded regions with poor mappability as described previously [13] resulting in a total of 9.7 Mb of analysed sequence. Within this sequence, the resulting numbers of variant positions used for phylogenetic reconstruction in each haplogroup are given in Supplemental Tables S4-S7.

**Haplogroup assignment**

We assigned chrY haplogroups using yHaplo [27] for the Illumina capture and WGS data. We used SNAPPY [28] for chrY haplogroup assignment of the genome-wide array genotyping data.

**Comparisons with an independent Estonian cohort**

To validate the representativeness of sequenced Estonian chrY samples (N=1,160), we compared the hg frequencies of this cohort against a ~7 times larger cohort of 7,949 Estonian male samples genotyped with the Illumina Infinium Global Screening Array v2 (Illumina, San Diego, CA, USA) containing 6,638 Y-specific single nucleotide variants (SNVs). To do this, we first assessed the accuracy of haplogroup assignments obtained from this particular set of SNVs. We sub-sampled the 6,638 array-specific Y-SNVs from the Estonian WGS data and used SNAPPY software to determine the haplogroups from the extracted set of SNVs. We compared the results against those from the software yHaplo [27]. The latter utilises the full set of SNPs in the WGS samples. The results are identical on the highest level of the major branches and only differ slightly at the finest resolution due to the lower number of array-genotyped SNPs available to SNAPPY for detecting the haplogroups. However, this shows that hg assignments based on the 6,638 array-specific Y-positions are accurate enough to be compared to hg assignments based on full sequencing. The comparison of the hg frequencies of the WGS-based and array-based Estonian datasets was performed using a Wilcoxon signed rank test with continuity correction. We only used array-based data for comparing haplogroup frequencies between two independent cohorts. For the phylogeny reconstruction and phylogeographic analysis full sequencing data were used.

**Phylogeny reconstruction of rare paternal haplogroups**

We reconstructed phylogenies and estimated the coalescent times with the software package BEAST v.1.7.5 [29]. We used a Bayesian skyline coalescent tree prior, the general time reversible (GTR) substitution model with gamma-distributed rates and a relaxed lognormal clock. The run was performed with the piecewise-constant coalescent model. The mutation rate used was $0.74 \times 10^{-9}$ (95% CI: $0.63 - 0.95 \times 10^{-9}$) per base

pair per year [13]. The results were visualized and checked for effective sample size above 200 in Tracer v.1.4. Coalescence time estimates were computed with normally distributed age priors with 10% standard deviation from previously published phylogeny [13] and are in Supplemental Table S3. Lineages from hg R and I were used as outgroups for hgs Q and G, respectively. Each run had thirty million chains logged every 3000 steps and 10% discarded as burn-in. Two parallel with different random number seeds were combined with LogCombiner.

The manually annotated phylogenetic trees, mutation lists and coalescence age estimates are available in Supplemental Figures S1-S4 and Supplemental Tables S4-S11. This study's updated nomenclature follows the criteria set in Karmin et al. (2015) [13].

**Bayesian phylogeographic analysis**

To illustrate the potential direction of influx of the primarily Estonian subclades in hgs E2a1-CTS1273 and J2b2-L283, we performed Bayesian phylogeographic analyses in continuous space. For this we used available geographic coordinates for 59 sequences belonging to hg E2a1-CTS1273 and for 41 sequences belonging to hg J2b2-L283. This method has been originally developed and successfully used to reveal the ancestral location and spatial dynamics of viruses in continuous space [30, 31]. We conducted the analysis according to the publication exploring the history of Y-chromosomal hg J1 [32] in BEAST v1.10.4 [33] using BEAGLE library v3.1.2 [34] for accelerated likelihood evaluation. This statistically robust and absolutely data-driven method uses molecular sequence data and geographic coordinates of the samples to infer phylogeography in a continuous landscape while simultaneously reconstructing the evolutionary history in

time. It draws the confidence area of ancestral locations where the root and internal nodes originated together with the directions and the speed of the diffusion (Figure 3). The uncertainties of the maximum clade credibility tree node locations were visualised with SpreaD3 v0.9.7.1rc software [35]. This inference approach accounts for the coalescent, phylogenetic, molecular clock, location, and other uncertainties within a single framework. Additional details are provided in Supplemental Note 1.

## Ethics approval

All donors have provided informed consent and all experiments were performed in accordance with the relevant guidelines and regulations of collaborating institutions. Access to genetic data in Estonian Biobank was approved by the Research Ethics Committee of the University of Tartu (permission number 1.1.-12/659 granted by the Research Ethics Committee of the University of Tartu, Estonia). The chrY sequences included from customers of the commercial personal genetic testing service were only from individuals who had provided informed consent for the use of their data in scientific research and for publication in aggregated form. The list of IDs along with additional sample information is presented in Supplemental Table S1.

## Results

### *Phylogeny of rare lineages in NEE*

The studied 1160 high coverage sequences of Y-chromosomes from Estonia disclose 64 samples carrying male lineages rare in NEE (frequency of each under 3%), amounting to ~6% of the total paternal lineage pool in Estonia. The most frequent minor lineage in

231 Estonia belongs to hg E2 (2.5%), followed by hgs J2 (1.9%) and hg G (0.9%), whereas
232 hg Q is the rarest (0.3%) (Supplemental Table S2). Our second largest sample set
233 consists of a total of 746 males from Sweden and discloses 78 samples with rare NEE
234 chrY lineages. The most common minor haplogroup in the Swedish cohort is hg Q
235 (4.6%); followed by hgs G (3%), E2 (1.7%) and J2 (1.2%) (Supplemental Table S2).

236 To verify the robustness of our frequency estimates, we compared hg frequencies of our
237 Swedish sample set and the SweGen cohort (N=505) [23]. The Wilcoxon signed rank
238 test showed no statistically significant differences between the two, either considering
239 all hgs (p-value=0.4689) or minor hgs with major hgs collapsed (p-value 0.6602).
240 Similarly, a comparison of hg frequencies between the Estonian sample set and an
241 independent non-overlapping set of 7,949 genotyped male samples from the Estonian
242 Biobank yielded no statistically significant differences in their hg composition, either
243 considering all haplogroups (Wilcoxon signed rank test p-value=0.4896) or rare hgs
244 with major hgs collapsed (Wilcoxon signed rank test p-value=0.9219).

245 Hg E originated in Africa with its sublineage E1 distributed solely on the African
246 continent, whereas the neighbour-lineage hg E2 displays a notably wider distribution.
247 Subclade E2-V13 is common (~10-20%) among south-eastern European populations [4,
248 6, 14, 16], falling to 10% in Anatolia and the Middle East [36] and declining towards
249 northern Europe to 1-2% in Scandinavia [4].

250 Here we reconstruct the phylogeny of hg E2a'b'c'd-M35. Its subclade E2a-M78 is
251 largely confined to Europe with a coalescence time of ~14 kya (95% CI: 10,432-18,566)
252 (Figure 1a, Supplemental Table S8). Within this subclade, L618 marker unites almost
253 all European samples that split ~13 (95% CI: 9,682-17,360) kya from the neighbouring

clade E2a2-V22. The latter consists primarily of samples from the Middle East with deeper diversification times (Supplemental Figure S5). The absolute majority (25/29) of hg E samples from Estonia belong to subclade E2a1-V13 (Supplemental Table S2). The bulk of Estonian samples form clearly distinguishable clusters: lineage E2a1-S7461 contains an Estonian founding cluster that splits from the neighbour lineages with Swedish and Middle Eastern origin ~4 kya (95% CI: 3,146 – 5,752, Supplemental Table S8) and a radiation time of ~2 kya (95% CI: 1,398 – 2,999; Supplemental Table S8). Similar pattern can be seen in the hg E2a1-B409 that has lineages from Germany and Sweden and an exclusively Estonian cluster defined by marker Z37869 with a radiation time of ~2 kya (95% CI: 1,150 – 2,428; Supplemental Table S8).

Hg J is one of the most common haplogroups in Western Asia and in regions surrounding the Mediterranean Sea and thus was initially connected to the dispersion of male farmers from the Fertile Crescent. Phylogenetic studies of hg J have shown surviving ancient sublineages with radiation signs in the Bronze Age [37, 38]. Additionally, hg J2a and an unresolved hg J have been discovered in ancient DNA from hunter-gatherer samples excavated in the Caucasus [39] and Karelia [40]. In southern Europe, the most common hg J subclade is J2-M172, which, however, becomes rare throughout the northern latitudes [4, 16].

Here we reconstruct the phylogenetic tree of hg J2-M172 (Figure 1b and Supplemental Figure S6) with 134 individuals. A substantial part of NEE individuals belong to sublineages within hg J2b2-L283 (Figure 1b) which splits from its neighboring clade at ~16 kya (95% CI: 11,860 – 20,018; Supplemental Table S9). Hg J2b2-L283 itself split ~7 kya (95% CI: 5,000 – 8,912) into two major sublineages J2b2-Z2505 and J2b2-YP29. The latter is an exclusively Estonian cluster encompassing over half of all hg J

278 samples from Estonia (12 of 22) with an expansion time of ~2 kya (95% CI: 1,446 –
279 3,027) (Figure 1b, Supplemental Figure S6, Supplemental Table S9).

280 The other major hg J subbranch – J2a-M410 – contains samples from broad Eurasian
281 background which are distributed in subclades mostly coalescing during the early post-
282 Last Glacial Maximum – a much deeper time estimate than in the neighbouring hg J2b-
283 M12 phylogeny (Supplemental Figure S6). Lack of information on detailed geographic
284 or ethnic origin hinders any further conclusions regarding the single-origin clusters from
285 the Russian Federation (Supplemental Figure S6). Based on published research,
286 lineages of hg J2a-M67 are among the most common (~20%) paternal haplogroups of
287 the North Caucasus region [41], whereas in ethnic Russians this haplogroup amounts to
288 less than 2% [5, 6].

289 Hg Q is frequent in Siberian populations and is carried by over 85% of male Native
290 Americans [16–18, 42]. In Europe, the occurrence of hg Q is uneven and the general
291 frequency is low (~0.42%) [42], but hg Q is somewhat more frequent in the populations
292 of Sweden and Norway [3, 4]. It is the most numerous minor haplogroup in both of our
293 Swedish sample sets with frequencies of 2.6% and 4.6% (Supplemental Table S2). In
294 the datasets of Karlsson et al. (2006) [4] and Lappalainen et al. (2008) [3] the frequency
295 of hg Q fluctuates between 1% and 5% in different regions of Sweden. On the updated
296 phylogenetic tree, Swedish samples fall into two main clusters that separated from each
297 other around the peak of the Last Glacial Maximum ~20 kya (Figure 2). About a third
298 of the Swedish hg Q samples are defined by marker L804. Hg Q1a-L804 coalesces ~16
299 kya (95% CI: 12,456 – 19,874; Supplemental Table S10) with haplogroup Q1a-M3,
300 which today describes the overwhelming majority of Native American Y-chromosomes

301 [42]. The rapid diversification among Swedes in the L804-defined clade began ~3 kya
302 (95% CI: 1,961 – 3,917; Supplemental Table S10).

303 Haplogroup G-M201 is common in the Caucasus and the Middle East. Hg G is one of
304 the most prevalent male lineages in Sardinia and Corsica, but displays low frequencies
305 elsewhere in Europe [4, 14, 15]. Hg G splits into two basal lineages – hgs G1 and G2, of
306 which the former occurs infrequently in Western and Central Asia and is almost absent
307 in Europe [15]. Almost all hg G samples from NEE belong to hg G2-P287 that ~22 kya
308 (95% CI: 17,620 – 26,973) split into two main subclades – G2a-P15 and G2b-M377
309 (Supplemental Figure S7, Supplemental Table S11). The bulk of sampled European
310 individuals belong to subclade G2a2-P303 (Supplemental Table S2). Downstream, in hg
311 G2a2-Z727, the absolute majority of Swedish hg G samples forms localised clusters
312 with a variety of coalescence times (Supplemental Figure S7, Supplemental Table S11).

313

314 **Discussion**

315 In case of Estonia, our sequenced samples were collected across the country avoiding
316 large settlements with recorded extensive migration history. Considering a census size
317 of roughly 1 million, rare lineages amount to a total of 30,000 men evenly sampled
318 across the country and thus cannot be exclusively ascribed to any random influx of
319 recent migrants.

320 From the screened sample of 506 Finnish males we did not detect any rare NEE
321 lineages as almost all Finnish samples belong to hgs common among neighbouring

populations – a probable reflection of either differing migration history or of demographic bottleneck(s) that have affected the Finnish population [43, 44].

Hg E sublineages have been associated with Neolithic demic diffusion into Europe [16], but current ancient DNA data has shown this haplogroup to be uncommon among the first agriculturalists in Europe [40]. In the resolved phylogenetic tree, the primarily Middle Eastern neighbouring clade with deeply diverged lineages supports a possible Levantine source of the European hg E2a1-V13. However, the split time predates the Neolithic transition in Europe and matches better with the age of the Villabruna hunter-gatherer cluster that displays earliest autosomal affinities to the Middle Eastern populations detected in ancient samples from Europe [45]. The coalescence age of the primarily European clades of hgs E3a1-V13 and J2b2-Z2505 underpins mid-Holocene as the starting point of chrY variation growth in Western Europe (Figure 1) and indicates a possible influx of male lineages from the Levant or the Caucasus.

The coalescence ages of Estonia-specific clades J2b2-YP29, E2a1-Z37869 and E2a1-Y28220 broadly correspond to the Late Bronze Age and Iron Age period in Northern Europe (Supplemental Figure S8). Additional sampling might certainly affect the coalescence age of these clusters. However, the geographical spread across all Estonian counties and current age estimates suggest that these expansions are not the result of any migratory events from the recent recorded (last ~800 years) history of this region. To infer the potential directions of influx of the clades J2b2-YP29, E2a1-Z37869, and E2a1-Y28220, we conducted continuous Bayesian phylogeographic analysis of parent hgs J2b2-L283 and E2a1-CTS1273. The estimated diffusion rate of hg J2b2-L283 equals 0.27 (95% HPDs: 0.1992 – 0.3478) and for hg E2a1-CTS1273 0.231 (95%

HPDs: 0.175 – 0.295) kilometers/year. The 80% HPD of the putative geographic centre of diffusion for the hg J2b2 covers the area focused in present-day Poland, with a partial covering of central and southeastern Europe, spreading further north and south (Figure 3a). The area for hg E2a1 ancestral location similarly covers central and eastern Europe with a focus on Poland (Figure 3b), but the focal point appears to be more condensed.

From a conservative standpoint, all three subclades most probably arrived to present-day Estonia from the direction of central Europe. However, based on currently available data, it is not possible to say whether the evident local expansions initially began in Estonia or were the carriers already sufficiently diversified on arrival.

Within hg Q, clusters defined by L804 and Y4838 capture almost all of Swedish hg Q diversity, marking these lineages as an inherent, albeit scarce, part of the pool of male lineages in Sweden The scarcity of internal nodes on the branches leading to the two now predominantly Swedish clusters hinders any discussion regarding a potential direction of influx or ancestral center of diffusion. Due to the glacial coverage, the split between lineages Q1a-L804 and the Native American Q1a-M3 could not have happened in Scandinavia. Ancient DNA research confirms the presence of hg Q in the remains of hunter-gatherers (~8 kya ) from Latvia and Lower Volga Region in Russia [46]. Today, European Q1 lineages are restricted to NEE with occasional findings in other populations (single L804 derived English chrY sample in Grugni et al. (2019) [47]). Precursors to current European hg Q1 sublineages could have been widely present in North Eurasia during the Last Glacial Maximum and followed a primarily northern (Siberian) route of dispersal into Europe. The presumptively common ancient gene pool is reflected in the autosomal European affinity of 24,000-old Mal'ta sample from the vicinity of Lake Baikal [48]. Alternatively, the prevalence of hg Q in Sweden could

testify of a more recent Siberian influence deduced both from modern and ancient DNA analysis in northeastern Europe [2, 8]. Studies have demonstrated minor eastern affinities in the autosomes and in the maternal lineages of the modern Saami, but small sample sizes have not revealed Saami male lineages belonging to hg Q [2]. Further sampling across Northern Eurasia might provide additional insights about these peculiar North Eurasian hg Q lineages. A total of two out of the three Estonian hg Q samples form a subset of the Swedish Y4838-defined cluster. It is most parsimonious to assume that the paternal ancestors of the two Y4838-derived individuals arrived in Estonia around 1-2 kya from Scandinavia.

Hg G2a has become firmly associated with the early Neolithic farmers of Europe [40, 46, 49]. Most of European hg G2a inner lineages started to diverge around 5-7 kya (Supplemental Figure S7, Supplemental Table S11) – within the timeframe of the European agricultural transition. In Sweden, it is the second most frequent minor chrY haplogroup. The majority of Swedish carriers demonstrate a strong expansion signal approximated to ~1 kya (nodes 52 and 60 in Supplemental Figure S7), whereas Estonian samples are not part of the Swedish hg G2a diversity.

In conclusion, we demonstrate that in NEE, rare paternal lineages are not just single lineages scattered across different subclades in the phylogeny. We identified several population-specific clusters among less common haplogroups, which testify of radiation events that have occurred in various timeframes and can be used to tentatively suggest possible influx directions.

This study demonstrates the power of large-scale re-sequencing of Y-chromosomes to explore and compare the male demographic history of single populations. Current survey of rare lineages paves the way for future research involving large datasets of re-

sequenced genomes with a focus on those maternal and paternal lineages that have left a major demographic impact on modern populations in NEE and elsewhere.

## Conflict of Interest

D.M.B and C.B. declare stock ownership at Gene by Gene, Ltd. L.S. in an employee of Gene by Gene.

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## Titles and legends to figures

**Figure 1.** Schematic phylogenetic trees of hg E2a and J2b

The calibrated trees were constructed using BEAST v.1.7.5 software package. Internal nodes, sub-clade names and population names (numbers show the number of samples) are indicated. Internal nodes with posterior probabilities <0.73 are not shown. Samples from Estonia and Sweden are marked in blue and orange, respectively.

A) A schematic phylogenetic tree of hg E2a is based on 132 high coverage chrY sequences. Neighbor-clade E2b and its sublineages are marked in grey. Detailed tree can be found in Supplemental materials (Supplemental Figure S5). Age estimates can be found in Supplemental Table S8. All the subclade (node) defining mutations and marker names are presented in Supplemental Table S4.

B) A schematic phylogenetic tree of hg J2b is based on 136 high coverage chrY sequences. Neighbor-clade J2a and its sublineages are marked in grey. Detailed tree can be found in Supplemental materials (Supplemental Figure S6). Age estimates can be found in Supplemental Table S9. All the sub-clade (node) defining mutations and marker names are presented in Supplemental Table S5.

**Figure 2.** Detailed phylogenetic tree of hg Q

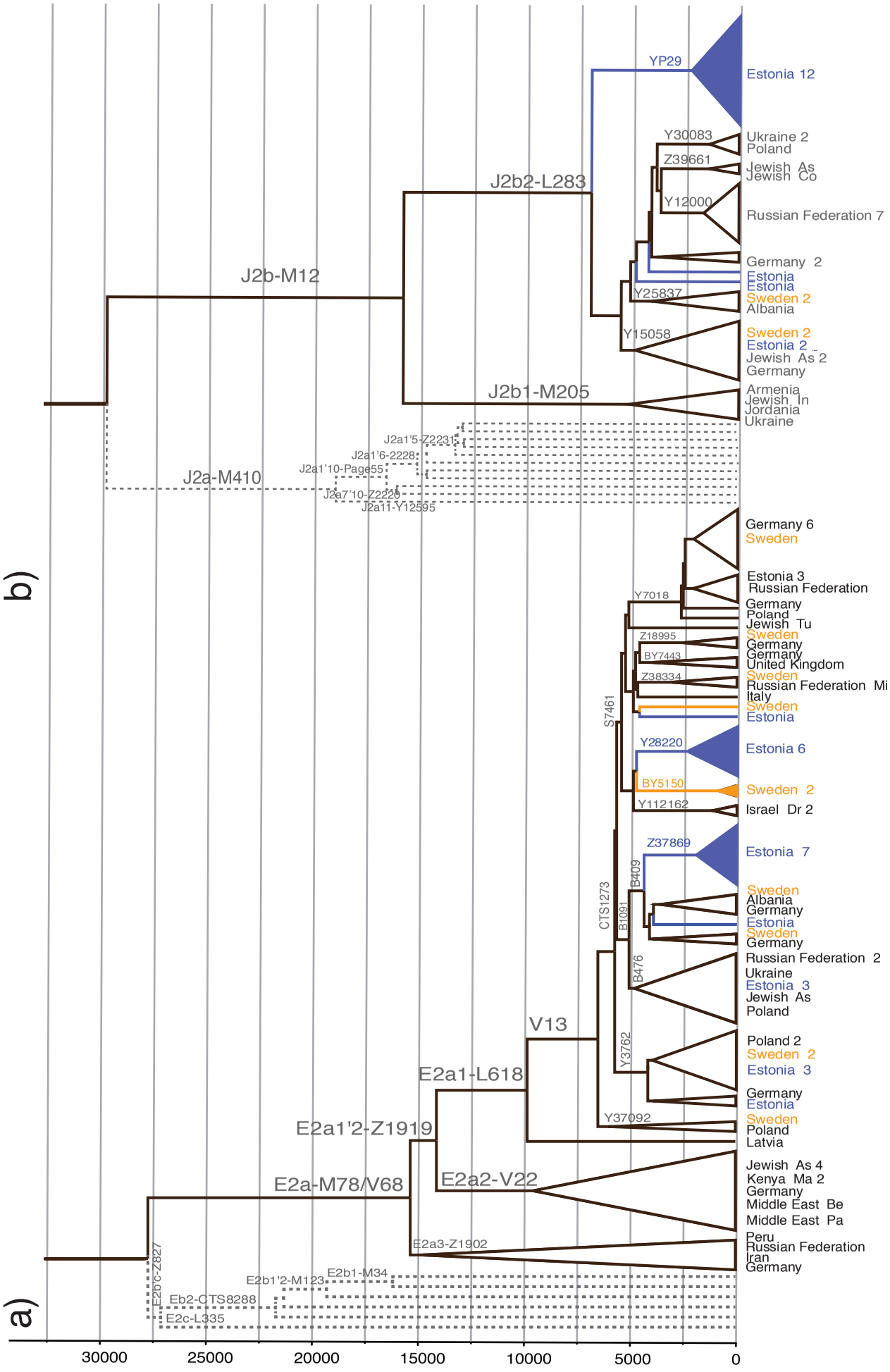
A detailed phylogenetic tree of hg Q-M242 is based on 84 high coverage chrY sequences. Two hg R1a sequences were used for an outgroup. The detailed calibrated tree was constructed using BEAST v.1.7.5 software package. Internal nodes, sub-clade names and population names are indicated. Internal nodes with posterior probabilities <0.73 are not shown. Age estimates can be found in Supplemental Table S10. All the subclade (node) defining mutations and marker names are presented in Supplemental

591 Table S6. Samples from Estonia and Sweden are marked in blue and orange,
592 respectively.

593

594 **Figure 3.** Phylogeographic spread maps of hgs J2b2-L283 and E2a1-CTS1273 in
595 Europe

596 Maps indicate the phylogeographic spread of A) J2b2-L283 around 6 kya, 4 kya, 2 kya
597 and in the present, and B) E2a1-CTS1273 around 5-6 kya, 4 kya, 2 kya and in the
598 present. Shaded in pink are the 80% HPD areas of the node locations inferred by
599 Bayesian continuous phylogeographic analysis with Beast v1.10.4 software. White
600 circles indicate the median locations of the nodes, while black lines indicate the
601 branches of the maximum clade credibility tree.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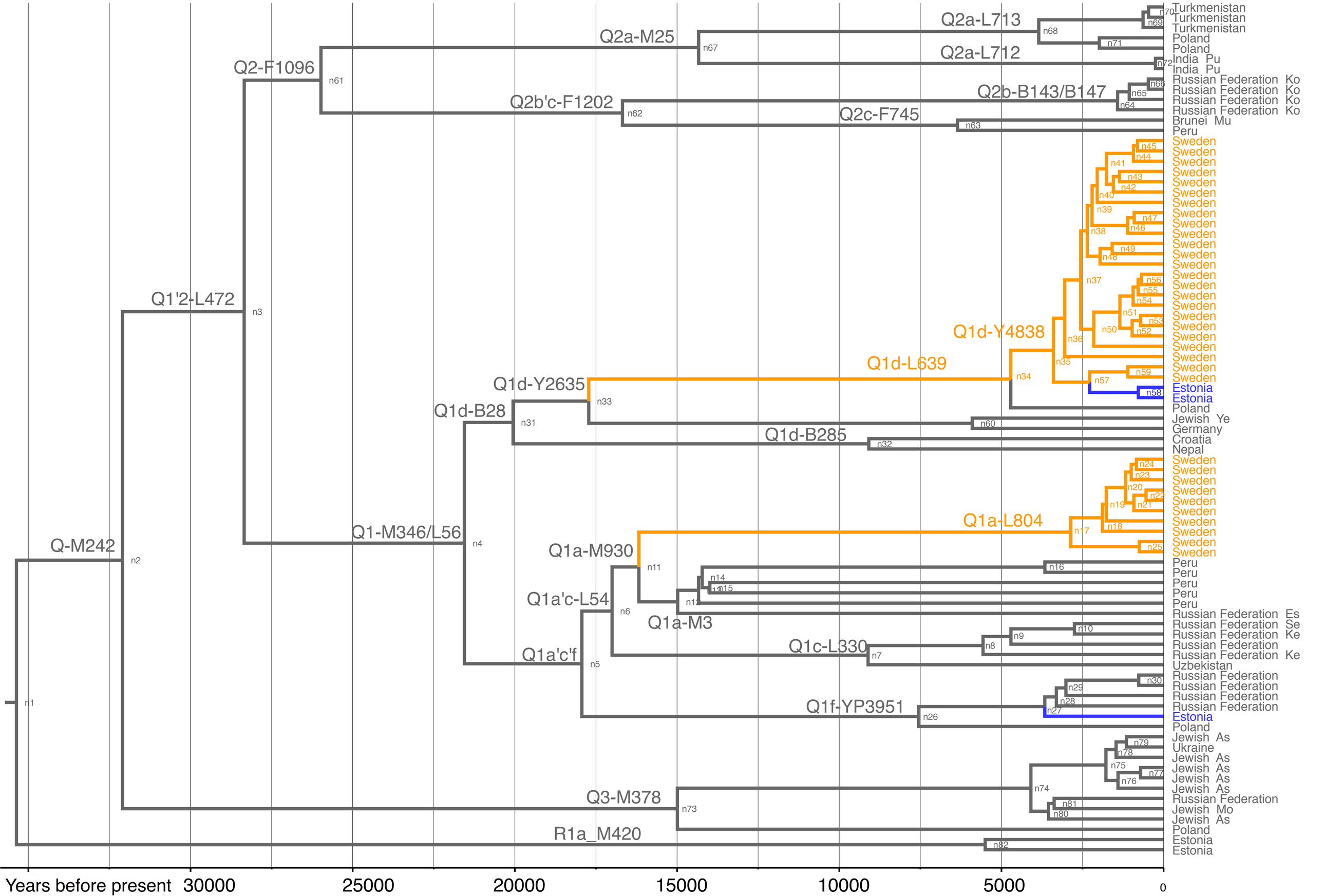
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Q2a-L713
Q2a-L712
Q2b-B143/B147
Q2c-F745
Q1d-Y4838
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Q1d-Y2635
Q1d-B28
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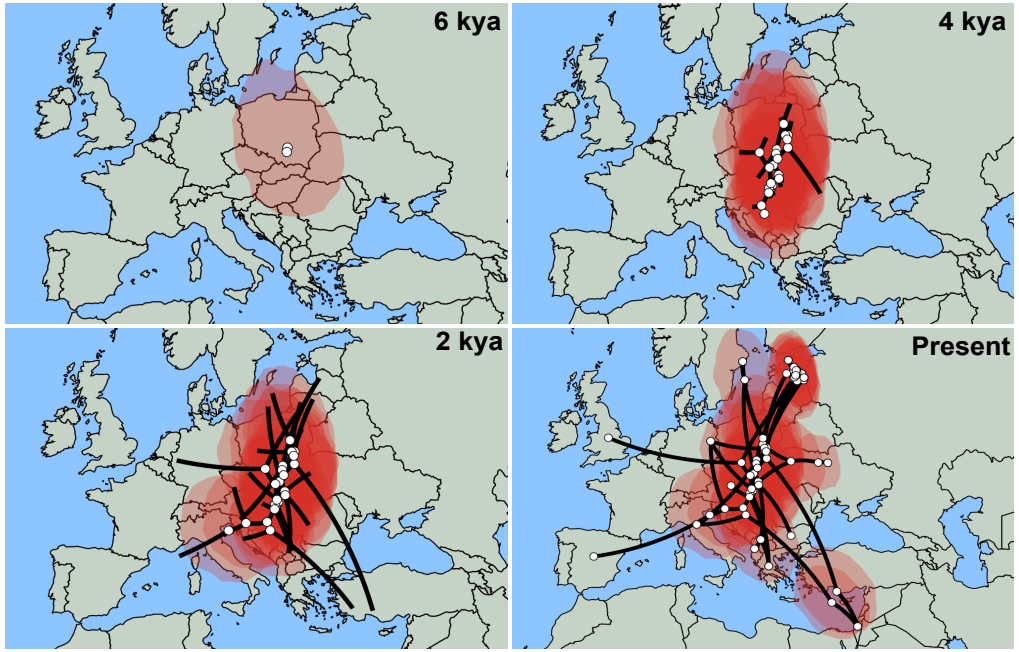


B) E2a1-CTS1273