1   **Supplementary Materials for**

2   **Genetics and material culture support repeated expansions into**

3   **Paleolithic Eurasia from a population Hub out of Africa**

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13 Supplementary Materials include:

14 - Supplementary Information

20 - Supplementary Figures S1 to S8

21 - Supplementary Tables S1 to S4

22   **Supplementary Section 1: Material Culture**

23   Many of the DNA samples discussed in this paper come from sites without stratigraphic association with diagnostic archaeological materials.

24   To overcome this issue, we use layers of sites located in proximity of and coeval to DNA sampling sites (Table S1).

25   **Table S1**. **Cultural proxies for the ancient genomes analysed in the current work**. M: Mousterian; S: Szeletian; IUP: Initial Upper
26   Paleolithic; UP: Upper Paleolithic; [a] indicates an attribution based on nearby coeval sites, which are specified in the subsequent rows. The
27   coordinates of the sites are taken from the reference articles or from the ROCEEH Out of Africa Database (ROAD) (http://www.roceeh.org).

DNA Sample/Proxy	Sample/Site name	Techno-complex	Country	Coordinates	Broad chronological range (cal kyr BP)	References
DNA Sample	**Vindija**	**M**	**Croatia**	**46.29 N, 16.07 E**	~50	*
Pre-expansion sites in the Levant	Ksar_Akil_XXII-XXV	IUP	Lebanon	33.91 N, 35.64 E	~45-43	(Leder 2014, 2017; Bosch et al. 2015)
Pre-expansion sites in the Levant	Üçagizli_F,Fa,Fb-c,G,H,H1-3,I	IUP	Turkey	35.98 N, 35.96 E	~45-40	(Kuhn 2004; Kuhn et al. 2009)
Pre-expansion sites in the Levant	Umel'Tlel	IUP	Syria	35.27 N, 38.89 E	~40	(Eric Boëda and Bonilauri 2006)
Pre-expansion sites in the Levant	Boker_Tachtit_layer_4	IUP	Israel	30.84 N, 34.78 E	~45-40	(Marks and Kaufman 1983; Kadowaki, Omori, and Nishiaki 2015)

DNASample	UstIshim	IUP[a]	Russia	57.7 N, 71.1 E	~44	*
Ust_Ishim	Kara_Bom_OH_5_OH6	IUP	Russia	50.72 N, 85.57 E	>45	(N. Zwyns et al. 2012)
Ust_Ishim	Tolbor-4_layer_4-5-6	IUP	Mongolia	49.29 N, 102.97 E	~45-35	(Derevianko et al. 2013; Nicolas Zwyns et al. 2019)
Ust_Ishim	Tolbor-16_layer_6	IUP	Mongolia	49.23 N, 102.92 E	~45-40	(Nicolas Zwyns et al. 2019)
Ust_Ishim	Kamenka_A	IUP	Russia	51.44 N, 108.17 E	~45-40	(N. Zwyns and Lbova 2019)
DNASample	Tianyuan	IUP[a]	China	39.66 N, 115.87 E	~40	*
Tianyuan	Suindonggou_1	IUP	China	38.38 N, 106.51 E	<41	(Morgan et al. 2014; E. Boëda et al. 2013)
Tianyuan	Suindonggou_2	IUP	China	38.38 N, 106.51 E	<41	(Li et al. 2019; Peng et al. 2020)
DNASample	Kostenki14	UP[a]	Russia	51.23 N, 39.3 E	~38	*
Kostenki14	Kostenki_12_Vokov	UP	Russia	51.39 N, 39.05 E	<40	(A. A. Sinitsyn and Hoffecker 2006)
Kostenki14	Kostenki_1	UP	Russia	51.39 N, 39.05 E	<40	(Hoffecker et al. 2016)

DNASample	Oase1	IUP-UP[a]	Romania	45.12 N, 21.9 E	~38	*
Oase1	Bacho_Kiro_IUP_layer_11	IUP	Bulgaria	42.95 N, 25.43 E	>45	(Tsanova 2008; Hublin et al. 2020)
Oase1	Ořechov_IV_–_Kabáty	IUP	Czech Republic	~49.06 N, 16.31 E	~41-35 (underestimate)	(Yuri E. Demidenko, Škrdla, and Rychtaříková 2020)
Oase1	Brno-Bohunice	IUP	Czech Republic	~49.12 N 16.37 E	>45	(Richter, Tostevin, and Skrdla 2008)
Oase1	Românesti-Dumbravita	UP	Romania	45.49 N, 22.19 E	~45-40	(Anghelinu and Niţă 2014; Sitlivy et al. 2012)
Oase1	Cosava	UP	Romania	45.51 N, 22.19 E	~45-40	(Anghelinu and Niţă 2014; Sitlivy et al. 2014)
Oase1	Tincova	UP	Romania	45.33 N, 22.9 E	~45-40	(Anghelinu and Niţă 2014; Sitlivy et al. 2014)
DNASample	GoyetQ116-1	UP[a]	Belgium	50.45 N, 5.01 E	~35	*
GoyetQ116-1	Maisières-Canal	UP	Belgium	50.48 N, 3.98 E	<40	(Pirson et al. 2012)
GoyetQ116-1	Spy_Ossiferous_Horizon_2	UP	Belgium	50.48 N, 4.67 E	<40	(Pirson et al. 2012)

DNASample	**Sunghir**	**UP**	**Russia**	**56.18 N, 40.50 E**	**~34**	(Dobrovolskaya, Richards, and Trinkaus 2012; Trinkaus and Buzhilova 2018)
DNASample	**Yana**	**UP**	**Russia**	**70.72 N, 135.42 E**	**~31**	(Pitulko, Pavlova, and Nikolskiy 2017)
DNASample	**Mal'ta (MA1)**	**UP**	**Russia**	**52.9 N, 103.5 E**	**~24**	(Khenzykhenova et al. 2019; Lbova 2019)
DNASample	**Bacho_Kiro_IUP_layer_11**	**IUP**	**Bulgaria**	**42.95 N, 25.43 E**	**~45**	(Hublin et al. 2020; Tsanova 2008)
DNASample	**Zlatý_Kůň**	**S-IUP[a]**	**Czech Republic**	**49.37 N, 16.72 E**	**>45**	*
Zlatý Kůň	Szeleta	S	Hungary	48.06 N, 20. 37 E	~45-40	(Adams 2009; Zs Mester 2010; Hauck, Rethemeyer, and Rentzel 2016)
Zlatý Kůň	Pod_Hradem	S	Czech Republic	49.37 N, 16.72 E	~45-40	(Nejman et al. 2017)
Zlatý Kůň	Moravský_Krumlov_IV	S	Czech Republic	49.05 N, 16.41 E	~43-42	(Nerudová and Neruda 2017)
Zlatý Kůň	Stranska_Skala_III-IIIc	IUP	Czech Republic	49.11 N, 16.40	~45-40	(Tostevin 2003)

Zlatý Kůň	Brno-Bohunice	IUP	Czech Republic	~49.12 N, 16.37 E	>45	(Richter, Tostevin, and Skrdla 2008)
DNASample	BK1653	UP	Bulgaria	42.95 N, 25.43 E	~35	(Fewlass et al. 2020; Hublin et al. 2020)

1.1 IUP-UP definition

29 The definition of a techno-complex is based on the association among specific traits
30 identified and recorded in archaeological assemblages (e.g. typology and technology of
31 stone tools). Depending on the degree of magnification used to define a lithic assemblage,
32 each techno-complex could be seen as more/less similar to others. A broad definition of
33 techno-complexes could be compatible with models of population structure/movement, but
34 this requires a high degree of simplification and the loss of finer-grained differences across
35 the examined sites. On the other hand, a strict definition of a techno-complex could focus on
36 the specificities of each assemblage, hampering the possibility of gaining a more general
37 perspective. Strict definitions of lithic assemblages are in fact more compatible with
38 scenarios of independent development (N. Zwyns and Lbova 2019). In order to develop a
39 model which considers a wide geographical and chronological scale as the one under
40 consideration in this work, it is crucial to describe the techno-complexes choosing the key
41 criteria that are both able to: i) maintain site-specificity; and ii) identify the general
42 tendencies. The question is how the data are collected and compared and what meaning
43 can be given to them concerning the question of the origin of technical innovations. New
44 technologies could be related to demic movements, cultural diffusion/exchange without
45 implying migration, or parallel convergence, i.e. the same response to common adaptive
46 challenges. Distinguishing homoplasy from homology requires testing a hypothesis on the
47 diversity of cultural traits and understanding it through a comprehensive framework of
48 technological evolution (E. Boëda et al. 2013; Morgan et al. 2014; Peng et al. 2020; Kuhn
49 2019; Boyd and Richerson 1988).

50 The terms Initial Upper Paleolithic, Early Upper Paleolithic and Upper Paleolithic, are
51 commonly used in the scientific literature with different meanings. They could have either a
52 chronological connotation or a technological one, or both of them. This paper refers to the
53 term Initial Upper Paleolithic (IUP) to indicate specific techno-complexes characterised by
54 volumetric blade productions and Levallois reduction sequence (Kuhn 2019; Kuhn and
55 Zwyns 2014) (Table S2). The term Upper Paleolithic (UP) is used to group the lithic
56 industries characterised by the production of several standardised blades and bladelets
57 often together with ornaments and bone tools (Table S2). The non-Mousterian and non-IUP
58 technologies appeared during the Middle to Upper Palaeolithic transition, comprising
59 Uluzzian (Benazzi et al. 2011; Moroni et al. 2018; Peresani et al. 2019; Collina et al. 2020;
60 Marciani et al. 2020; Riel-Salvatore 2009), Châtelperronian (Morgan Roussel 2013;
61 Ruebens, McPherron, and Hublin 2015; M. Roussel, Soressi, and Hublin 2016), Szeletian

62   (Nerudová and Neruda 2017; Zsolt Mester 2018; Neruda and Nerudová 2019) and

63   Lincombian-Ranisian-Jermanowician (LRJ) (Flas 2011; Kot et al. 2020; Krajcarz et al. 2018).

64   **Table S2.** Main criteria adopted in this work to characterise Initial Upper Paleolithic (IUP)

65   and Upper Paleolithic (UP) sites.

Definition	**IUP** **Initial Upper Paleolithic**	**UP** **Upper Paleolithic**
Lithic technology	The IUP is a blade-based production. The reduction sequences use direct hard hammer percussion, platform faceting, and mostly flat-faced or semi-tournant cores. The blades and some cores resemble products of Levallois reduction sequence (Kuhn and Zwyns 2014; Kuhn et al. 2009; Kuhn 2019). Sites in Siberia and Mongolia are also characterised by "burin-cores" for producing small blades and the exploitation of the narrow face cores (Slavinsky et al. 2019; N. Zwyns et al. 2012; N. Zwyns and Lbova 2019).	Blade and bladelet based industries usually used in complementary tools. The broad category of UP includes a great diversity of techno-complexes which share the production of standardised bladelets and blades with a wide range of technical options (e.g. unidirectional debitage by prismatic core, carinated core, burin-cores, among others) and percussion techniques (Zilhão et al. 2006; Le Brun-Ricalens, Bordes, and Eizenberg 2009; Teyssandier, Bon, and Bordes 2010a; Moreau 2012a; Kozłowski 2015b; Bataille 2016; Falcucci 2018; Kadowaki, Suga, and Henry 2021b).
Included techno-complex	Emiran (Kuhn, Stiner, and Güleç 1999), Bokerian (Leder 2014, 2017), Bohunician (Richter, Tostevin, and Skrdla 2008; Škrdla 2017; Yuri E. Demidenko, Škrdla, and Rychtaříková 2020), Bachokirian (Hublin et al. 2020).	Protoaurignacian and Aurignacian (Teyssandier, Bon, and Bordes 2010b; Sitlivy et al. 2012; Bataille 2016; Y. E. Demidenko and Škrdla 2017; Tafelmaier 2017; Falcucci 2018; Riel-Salvatore and Negrino 2018), Spitsynian (Vishnyatsky and Nehoroshev 2004; Usik, Monigal, and Kulakovskaya 2006), Ahmarian (Kadowaki, Omori, and Nishiaki 2015; Barzilai, Hershkovitz, and Marder 2016; Goring-Morris and Belfer-Cohen 2018), Early Upper Paleolithic (Hoffecker 2011; Kadowaki, Suga, and Henry 2021a), Gravettian (Andey A. Sinitsyn 2007; Dobrovolskaya, Richards, and Trinkaus 2012; Moreau 2012b; Kozłowski 2015a; Klaric 2013).

Other material evidence	In some IUP assemblage is documented the use of personal ornament and formal bone tools e.g. in Levant (Kuhn et al. 2009), Bulgaria, Siberia, and Mongolia (Rybin 2014; Kuhn and Zwyns 2014; Kaifu et al. 2014; Derevianko and Rybin 2003).	Habitual use (especially in the Gravettian) of portable art, graphic representations, musical instruments, and various bone tools (Zilhão et al. 2006; Conard 2003; Conard, Malina, and Münzel 2009). Impressive burials sites e.g. Sunghir (Trinkaus et al. 2014).
Chronology	Chronologically, the IUP is a long phenomenon comprised between approximately 50 kya and 35 kya (calibrated). Its stratigraphic position follows the Middle Paleolithic assemblages and is before UP assemblage (Nerudová and Neruda 2017; Zsolt Mester 2018; Neruda and Nerudová 2019; Kuhn 2019; Kuhn and Zwyns 2014).	The various stages of the UP make their appearances approximately 42 kya. Their stratigraphic position follows the IUP, Uluzzian, Szeletian, Chatelperroian, LRS.

Supplementary Section 2: Genetic dataset

Table S3: Individuals used in qpGraph trees. A green background indicates samples
present in the "Allen Ancient DNA Resource" v44.3 database while an orange background
indicates samples available from ENA.

Version ID	Master ID	Publication	GroupID	PopID in qpGraph	SNPs
GoyetQ116-1_published	GoyetQ116-1	FuNature2016	Belgium_UP_GoyetQ116_1_published_all	GoyetQ116-1	765579
Chimp.REF	Chimp	Genome	Chimp.REF	Chimp	1102442
Tianyuan	Tianyuan	YangCurrentBiology2017	China_Tianyuan	Tianyuan	885079
HGDP00449.SDG	HGDP00449	BergstromScience2020	Mbuti.SDG	Mbuti	1140413
HGDP00462.SDG	HGDP00462	BergstromScience2020	Mbuti.SDG	Mbuti	1137821
HGDP00463.SDG	HGDP00463	BergstromScience2020	Mbuti.SDG	Mbuti	1137064
HGDP00467.SDG	HGDP00467	BergstromScience2020	Mbuti.SDG	Mbuti	1136730
HGDP00474.SDG	HGDP00474	BergstromScience2020	Mbuti.SDG	Mbuti	1140529
HGDP00476.SDG	HGDP00476	BergstromScience2020	Mbuti.SDG	Mbuti	1140439
HGDP00478.SDG	HGDP00478	BergstromScience2020	Mbuti.SDG	Mbuti	1137743
HGDP00982.SDG	HGDP00982	BergstromScience2020	Mbuti.SDG	Mbuti	1126932
HGDP00984.SDG	HGDP00984	BergstromScience2020	Mbuti.SDG	Mbuti	1137625
HGDP01081.SDG	HGDP01081	BergstromScience2020	Mbuti.SDG	Mbuti	1135761
B_Papuan-15.DG	HGDP00546	PrueferNature2013	Papuan.DG	Papuan	1117740
S_Papuan-1.DG	HGDP00550	SkoglundNature2015	Papuan.DG	Papuan	1118074
S_Papuan-10.DG	HGDP00553	SkoglundNature2015	Papuan.DG	Papuan	1118955
S_Papuan-11.DG	HGDP00555	SkoglundNature2015	Papuan.DG	Papuan	1118110
S_Papuan-12.DG	HGDP00556	SkoglundNature2015	Papuan.DG	Papuan	1116429
S_Papuan-13.DG	HGDP00552	SkoglundNature2015	Papuan.DG	Papuan	1118089
S_Papuan-14.DG	HGDP00554	SkoglundNature2015	Papuan.DG	Papuan	1118823
S_Papuan-2.DG	HGDP00540	SkoglundNature2015	Papuan.DG	Papuan	1118158
S_Papuan-3.DG	HGDP00541	SkoglundNature2015	Papuan.DG	Papuan	1118510
S_Papuan-4.DG	HGDP00543	SkoglundNature2015	Papuan.DG	Papuan	1118633

S_Papuan-5.DG	HGDP00545	SkoglundNature2015	Papuan.DG	Papuan	1119621
S_Papuan-6.DG	HGDP00547	SkoglundNature2015	Papuan.DG	Papuan	1118542
S_Papuan-7.DG	HGDP00548	SkoglundNature2015	Papuan.DG	Papuan	1119624
S_Papuan-8.DG	HGDP00549	SkoglundNature2015	Papuan.DG	Papuan	1118670
S_Papuan-9.DG	HGDP00542	SkoglundNature2015	Papuan.DG	Papuan	1118992
Oase1_d	Oase1	FuNature2015	Romania_Oase	Oase1	195345
Kostenki14	Kostenki14	FuNature2016	Russia_Kostenki14	Kostenki14	1068326
MA1.SG	MA1	RaghavanNature2013	Russia_MA1_HG.SG	MA1	805960
Sunghir1.SG	Sunghir1	SikoraScience2017	Russia_Sunghir1.SG	Sunghir	795499
Sunghir2.SG	Sunghir2	SikoraScience2017	Russia_Sunghir2.SG	Sunghir	1136035
Sunghir3.SG	Sunghir3	SikoraScience2017	Russia_Sunghir3.SG	Sunghir	1149730
Sunghir4.SG	Sunghir4	SikoraScience2017	Russia_Sunghir4.SG	Sunghir	1133177
Ust_Ishim_published.DG	Ust_Ishim	FuNature2014	Russia_Ust_Ishim_HG_published.DG	Ust_Ishim	1147829
Yana_old.SG	Yana1	SikoraNature2019	Russia_Yana_UP.SG	Yana	1150093
Yana_old2.SG	Yana2	SikoraNature2019	Russia_Yana_UP.SG	Yana	1147981
baa001.SG	baa001	SchlebuschScience2017	South_Africa_1900BP.SG	South_Africa_2000BP	1149601
I9133.SG	UCT386	SkoglundCell2017	South_Africa_1900BP.SG	South_Africa_2000BP	1009127
bab001.SG	bab001	SchlebuschScience2017	South_Africa_2000BP.SG	South_Africa_2000BP	719140
I9028.SG	KhoesanLeipzigHunter	SkoglundCell2017	South_Africa_2200BP.SG	South_Africa_2000BP	718296
Vindija_snpAD.DG	Vindija	Pruefer2017	Vindija_Neanderthal.DG	Vindija Neanderthal	1061414
	ZKU002	Pruefer2021		ZlatyKun	653677
	BB7-240	Hajdinjak2021		Bacho_Kiro	866466
	CC7-335	Hajdinjak2021		Bacho_Kiro	783301
	F6-620	Hajdinjak2021		Bacho_Kiro	1068449
	BK_1653	Hajdinjak2021		BK_1653	1025353

70 **Supplementary Section 3: qpGraph analyses**

71 **3.1 Base graph with Zlatý Kůň and Eurasian trifurcation**

72 The position of Ust'Ishim with respect to Paleolithic Eastern and Western Eurasian is unclear
73 and described as a near-trifurcation (Yang et al. 2017; Lipson and Reich 2017) with D-
74 statistics in the form of (Tianyuan, Sunghir, Ust'Ishim, Chimp), (Tianyuan, Ust'Ishim,
75 Sunghir, Chimp) and (Sunghir, Ust'Ishim, Tianyuan, Chimp) all not significantly different from
76 zero (Table S4).
77 We started with the simple population tree proposed by Prufer and colleagues (Prüfer et al.
78 2021) (Fig 2c) with Zlatý Kůň as the most basal non African and placed Ust'Ishim either
79 basal to the split between Western and Eastern Eurasians, as a sister of East Eurasians or
80 as a sister of West Eurasians and found that all options are supported with no outlier f4s,
81 being the highest f4 |Z-score| < 3. We used Tianyuan as representative ancient "East Asian"
82 and either Sunghir or Kostenki14 as ancient "West Eurasian".

83 **3.2 Adding Bacho Kiro**

84 We then tried to add the IUP Bacho Kiro individuals in different positions of these graphs,
85 downstream of the Zlatý Kůň split. We assumed Zlatý Kůň to be more basal than Bacho Kiro
86 because of the following reasons:
87 • The Bacho Kiro individuals, set aside their higher Neanderthal admixture proportion,
88 have been shown to share more alleles with East Asians compared to Europeans,
89 while Zlatý Kůň is basal to the split between the two (Hajdinjak et al. 2021).
90 • f4 in the form (X, Zlatý Kůň, Bacho Kiro, Chimp) with X a later Eurasian individual
91 including Ust'Ishim tend to be positive, even if not always significant (Table S4).

92 As a first attempt we placed Bacho Kiro basal to Ust'Ishim and all other Eurasians except
93 Zlatý Kůň (Figure S1.A): the resulting graph is rejected since it has several f4 with |Z-scores|
94 > 3, highlighting the fact that the genetic distances expected from the tree designed by the
95 user do not match the ones computed from the real data and hence implying a poor fit to the
96 data for the examined tree. f2- statistic results, which are indicators of which samples should
97 be closer in the tree to account for the observed genetic distance, suggest that Bacho Kiro
98 should be placed closer to Tianyuan (f2 Z-score = -3) and further from Sunghir (f2 Z-score =
99 2.6). A similar result is obtained when placing Ust'Ishim basal to Bacho Kiro and Bacho Kiro

100 basal to the split between Europeans and East Asians, here represented by Tianyuan and
101 Sunghir (Figure S1.B).
102 Rather than adding an admixture event from a Bacho Kiro related population to Tianyuan we
103 tried to minimize admixture events and instead placed Bacho Kiro as a sister of Tianyuan,
104 hence leaving Ust'Ishim basal to the split Europe/East Asia. This returned a graph with the
105 worst (i.e. highest in absolute value) Z-score = -2.7 (Figure S1.C), which hence provides a
106 satisfactory fit to the data.
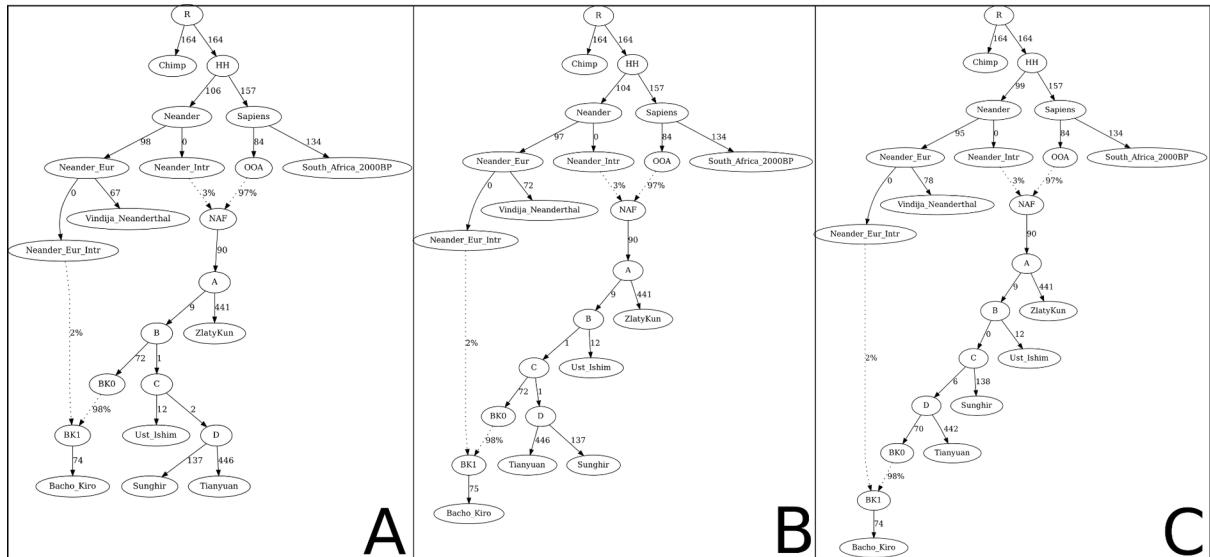
107 **Figure S1 Placement of Bacho Kiro within the tree proposed in Prufer et al. 2021.**
108 When taking into account the excess Neanderthal contribution, Bacho Kiro still yields outlier
109 f4s when placed soon after the split of Zlatý Kůň (**A**) or after the separation of Ust'Ishim (**B**).
110 Placing Bacho Kiro as a sister of Tianyuan (**C**) yields instead no f4 outliers. (Graphs based
111 on 423445 SNPs. Highest |Z-score| = 4.3(A, B), 2.7(C))

112 The placement of Bacho Kiro with respect to other Eurasians that we are proposing differs
113 from the one proposed by Hajdinjak and colleagues (Hajdinjak et al. 2021) in their Figure
114 S6.2. We speculate this may be due to the presence of Zlatý Kůň in our tree who may inform
115 the genetic drift that characterizes a basal OoA landscape, or to the availability of
116 Neanderthal from the early phases of our qpGraph construction (whose absence, by virtue of
117 the additional Neanderthal ancestry compared to other non Africans, can have the effect of
118 making Bacho Kiro appear more basal if not immediately accounted for)(Table S4).
119 For the sake of completeness we also tried to position Bacho Kiro as a sister of Sunghir
120 (geographically it could make sense). The graph has several f4 |Z-scores| > 3 and is
121 therefore rejected.

3.3 Ust'Ishim as an early leaf of the IUP branch

We added Kostenki14, the oldest "genetically European" individual sequenced to date to "populate" more the European branch (Figure S2.A – final score 11964). Since we noticed that the drift between nodes B and C was 0 and bearing in mind that D-statistics (Table S4) and previous graphs supported Sunghir/Tianyuan/Ust'Ishim to be a trifurcation, we tried to place Ust-Ishim either as a sister of Tianyuan/Bacho Kiro (Figure S2.B – score 11630) or as a sister of Kostenki14/Sunghir (Figure S2.C – score final 11721). All three graphs produce no f4 outliers and, when not basal, Ust'Ishim shares only one unit of drift with its sisters before branching off (hence showing a near-trifurcation). While all three topologies provide no outlier Z scores, the tree where Ust'Ishim is a sister of Tianyuan/Bacho Kiro (Figure S2.B), has the lowest final score and so is the most supported.
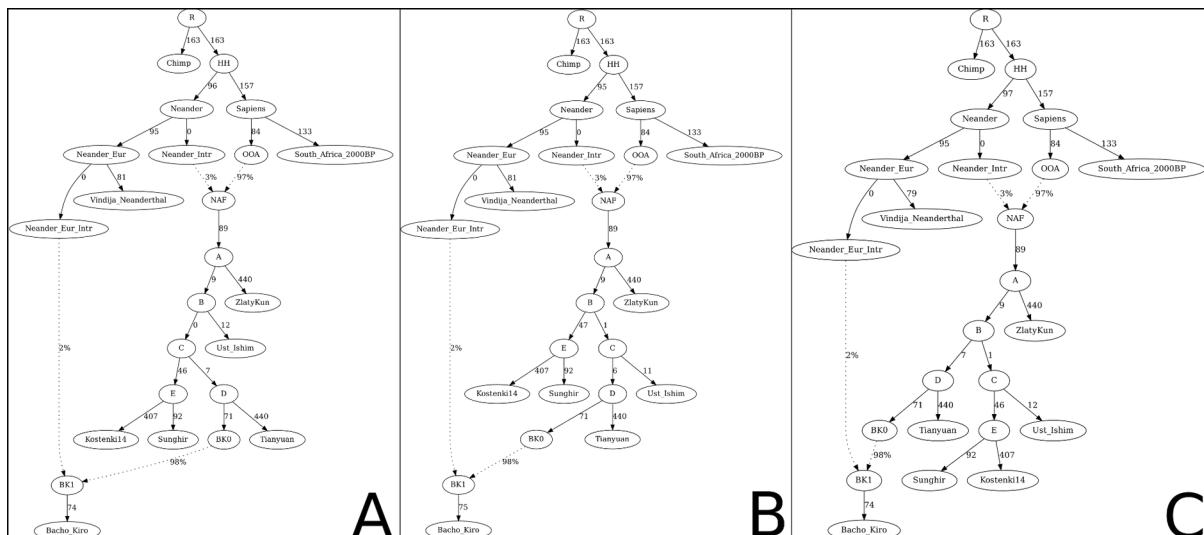
Figure S2 Relationship of Ust'Ishim to other Eurasians. After adding Kostenki14 as a crucial reference sample, we tested all possible positions of Ust'Ishim as either basal to East and West Eurasians (**A**), sister of Bacho Kiro and Tianyuan (**B**) and sister of Kostenki and Sunghir (**C**). All three graphs produce no f4 outliers and Ust'Ishim shares only one unit of drift with its sisters before branching off (hence showing a near-trifurcation). The tree where Ust'Ishim is a sister of Tianyuan/Bacho Kiro (Figure S2.B), however, has the lowest final score and so is the most supported. (Graphs based on 408905 SNPs. Highest |Z-score| = 2.7)

3.4 Oase1 could be simply a descendant of Bacho Kiro, remnant of the IUP movement

We then tried to place on the emerging tree of Figure S2.B the ~40 ky old Oase1 individual, recovered from a site less than 400 km away from Bacho Kiro cave which, like and more than the individuals from Bacho Kiro, has been shown to have a Neanderthal relative in its

145 recent genealogy.

146 Since it has not been possible, based on simple f4s, to determine a higher affinity of Oase1
147 for either East Asians or Europeans (retrospectively, that might be caused by the
148 combination of low number of SNPs and high Neanderthal admixture proportion), we started
149 by making Oase1 a sister of Zlatý Kůň that later admixed with Neanderthals (Figure S3.A).
150 The resulting graph showed several |Z scores| > 3, an excessive proportion of Neanderthal
151 in Eurasians and a clear tendency for Oase1 and Bacho Kiro to share more drift (f2 Z-score
152 = -6.0), incidentally showing that the quality and quantity of Oase1 genetic information is
153 nevertheless suitable for a qpGraph analysis. We then placed Oase1 as a sister of Bacho
154 Kiro with an additional Neanderthal pulse and obtained no outlier f4 and a parsimonious
155 placement for this elusive sample (Figure S3.B).

156 Our proposed graphs illustrate the claim made by Fu and colleagues (Fu et al. 2015) that
157 Oase1 experienced an additional pulse of Neanderthal admixture between the one shared
158 by all non Africans and the one that occurred 4-6 generations before it lived, and identify
159 said event in the one occurred in the Bacho Kiro population or in a closely related one.
160 A similar graph where the node Oase0 splits from BK0 instead of BK1 (Figure S3.C) is also
161 not rejected but does not explain this additional event of Neanderthal admixture.

162 To further support the placement of Oase1 and rule out the possibility that its attraction to
163 Bacho Kiro is driven by the excess of Neanderthal ancestry they share, we proceeded to
164 mask out the most recent Neanderthal introgressed segments of Oase1 (we used the
165 genomic coordinates reported in Table S5.1 of Fu et al 2015 (Fu et al. 2015)) and re-run the
166 analysis: Oase1 holds its position and can be described without the last admixture event with
167 Neandethal (inferred to be 0) hence confirming its connection with the Bacho Kiro population
168 or a related one is genuine and not a simple attraction introduced by their shared excess
169 Neanderthal fraction (Figure S3.D).

170 Finally, it is worth mentioning that while the low coverage, high contamination and high
171 Neanderthal ancestry of Oase1 prevented the direct assessment of its closer relationship to
172 either Western or Eastern Eurasians, an individual from the same site and with similar age
173 (Oase2) showed a clearly higher affinity for East Asian and Native American populations
174 than with Western Eurasians. The closest sample to Oase2 in outgroup f3 analyses, after
175 Oase1, was reported to be Tianyuan (the individuals from Bacho Kiro cave were not
176 available at the time of those analyses), supporting our claim of its placement in the
177 "genetically East Asian" branch (Siska 2019).
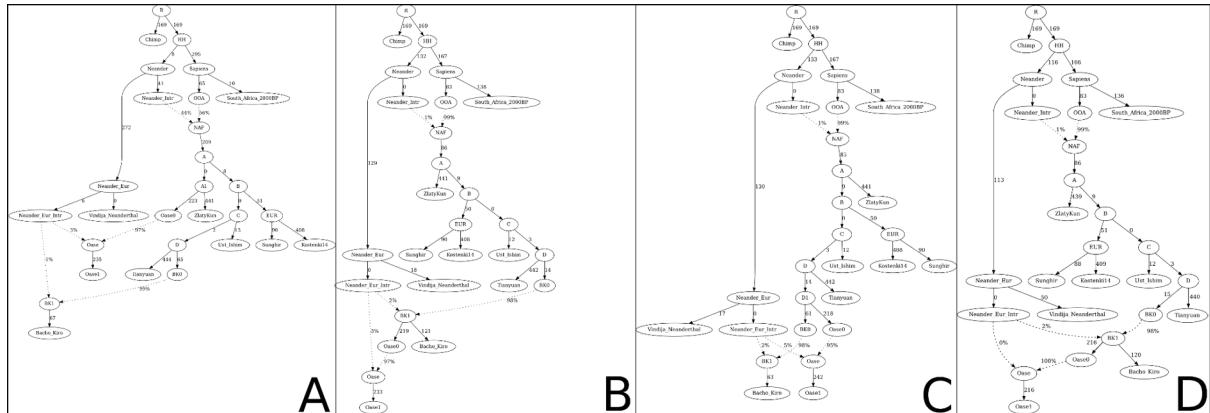
Figure S3 Placement of Oase along the IUP tree. After accounting for the reported recent pulse of Neanderthal admixture, Oase1 cannot be described as a simple basal lineage along the Eurasian tree (**A**) and is instead best fit as a descendant of the same population Bacho Kiro belonged to (**B**, **C**). When masking out the very recent Neanderthal introgressed segments present in Oase1, this individual can still be modeled as a sister of Bacho Kiro, confirming their attraction is genuine and not mediated by Neanderthal (**D**). (Graphs A, B, C based on 95587 SNPs, graph D based on 88793SNPs. Highest |Z-score| = 7.2(A), 2.6 (B, C, D)).

3.5 Interaction between Zlatý Kůň and Bacho Kiro

Given the geographical proximity of the two sites, we tested whether a contribution to the Bacho Kiro individuals by a population related to Zlatý Kůň can be accommodated within our so far most supported graph.

An admixture event contributing between 2% and 29% from a sister of Zlatý Kůň into the ancestors of Bacho Kiro, depending on the position of Ust'Ishim (Figure S4), yields no outliers. Although not rejected, these graphs are all less parsimonious than the graph in Figure S2.B and might pick up a signal of shared drift between Bacho Kiro and Zlatý Kůň that is caused by the reduced time purifying selection had to drive out introgressed Neanderthal segments (Zlatý Kůň is the oldest sample and Bacho Kiro has recent Neanderthal introgression), rather than a genuine gene flow.
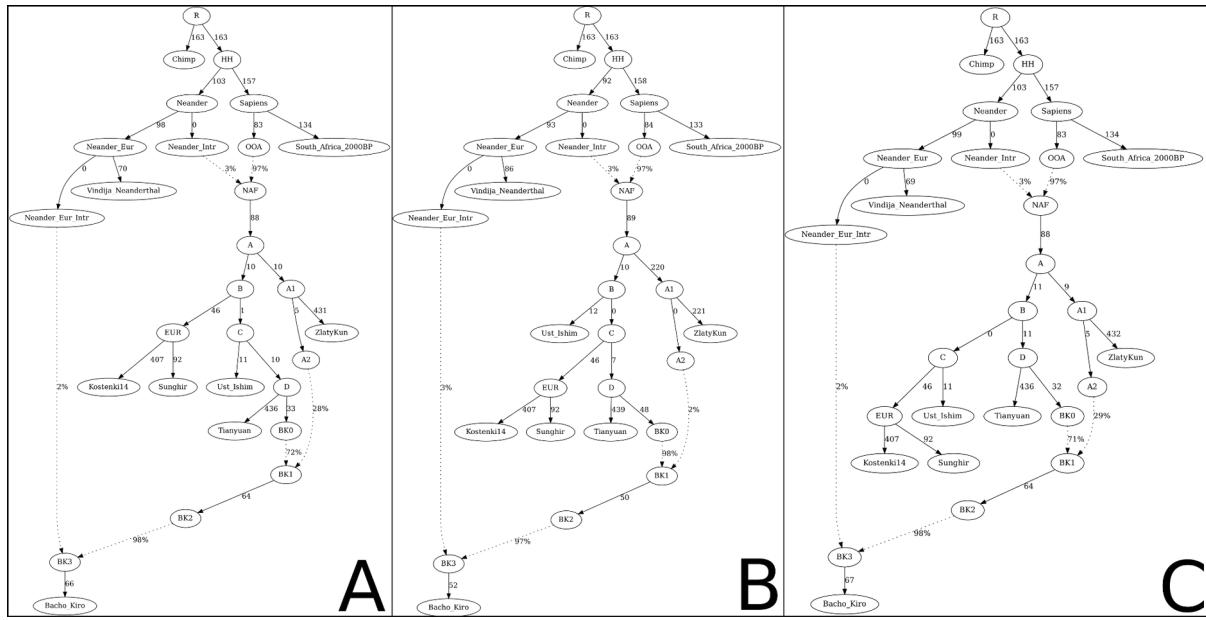
Figure S4 Interaction between Zlatý Kůň and Bacho Kiro. Given the geographic and partial chronological overlap of Zlatý Kůň and Bacho Kiro samples we allowed for a contribution of the former on the newly arrived IUP population. Such a contribution is acceptable, in varying amounts depending on the position of Ust'Ishim within the tree (**A,B,C**) but given the viability of the trees without this event we decided to exclude it from further analyses. (Graphs based on 408905 SNPs. Highest |Z-score| = 2.1 (A, C), 2.3 (B)).

3.6 Formation of Paleolithic Siberian ancestry profile

Paleolithic Siberian populations younger than 40 ky are consistently described as a mix of European and East Asian ancestries (Massilani et al. 2020; Sikora et al. 2019; Raghavan et al. 2014), although a comprehensive cultural or population dynamics to account for these admixture events is still lacking. Building on our dichotomy between Initial Upper Paleolithic (IUP) and Upper Paleolithic waves of expansions across Eurasia, we tried to model ancient Siberian individuals as a mixture of these waves into our best fitting parsimonious graph (Figure S2.B).

We started by creating a Siberian population that is ancestral to the ~32 ky old Yana individuals (Massilani et al. 2020; Sikora et al. 2019; Raghavan et al. 2014) as a mixture of the E and C nodes in the graph of Figure S2.B. The resulting graph has several f4 outliers and indicates that Yana may fit better as more closely related to Tianyuan (f2 Z-score = -6.1) and, to a lesser extent, Bacho Kiro (f2 Z-score = -2.1), suggesting that the node of origin of the East Asian component in Siberians needs to be moved further down along that branch. With the East Asian component of Siberians originating from node D the graph still has

17   17

several outliers and Yana needs to share more drift with Tianyuan (f2 Z-score = -3.8), so
219 after taking that into account we obtained a graph that is not rejected (Figure S5.A).
220 To better define the Siberian populations we included the ~24 thousands years old Mal'ta
221 (MA1) individual (Fu et al. 2015; Raghavan et al. 2014), we started by considering it as a
222 sister of Yana (Figure S4.B) and obtained two |Z-scores| ~3,1 (Chimp,
223 South_Africa_2000BP, ZlatyKun, Yana and ZlatyKun, Tianyuan, Sunghir, MA1). Since
224 qpGraph performs f-statistic analyses on a subset of SNPs that is shared by all the
225 individuals in a tree, this might sometimes severely limit the number of SNPs used in a test,
226 especially when ancient individuals are included. We tested whether the two outliers remain
227 significantly different from zero when including all SNPs available to each quadruple and
228 found they do not (Table S4), deeming this outcome as more robust.
229 Finally, in order to allow MA1 and Yana to have different proportions of East and West
230 Eurasian components, we added an EUR_Sib node and parallelized the admixture events of
231 Yana and MA1. The resulting graph (Figure S5.C) has one f4 test with Z-score 3.1 (Chimp,
232 South_Africa_2000BP, ZlatyKun, Yana) , however it results not significantly different from
233 zero when computing the D-statistic using the all the available SNPs (Table S4) . Since the
234 data we used has been produced over many years and with different strategies, we decided
235 to also verify the structure we found using only transversions. The core structure of the tree
236 (IUP and UP branches, with Siberians as a mixture) holds, however the East Asian
237 component of siberians needs to pick ancestry from different positions along the IUP
238 branch:we think this is due to the limited number of SNPs (55427) this graph is based on,
239 that confer enough power to identify siberians as a mixture of UP and IUP, but not sufficient
240 to pinpoint the exact position of the admixing source within the variegated IUP branch.
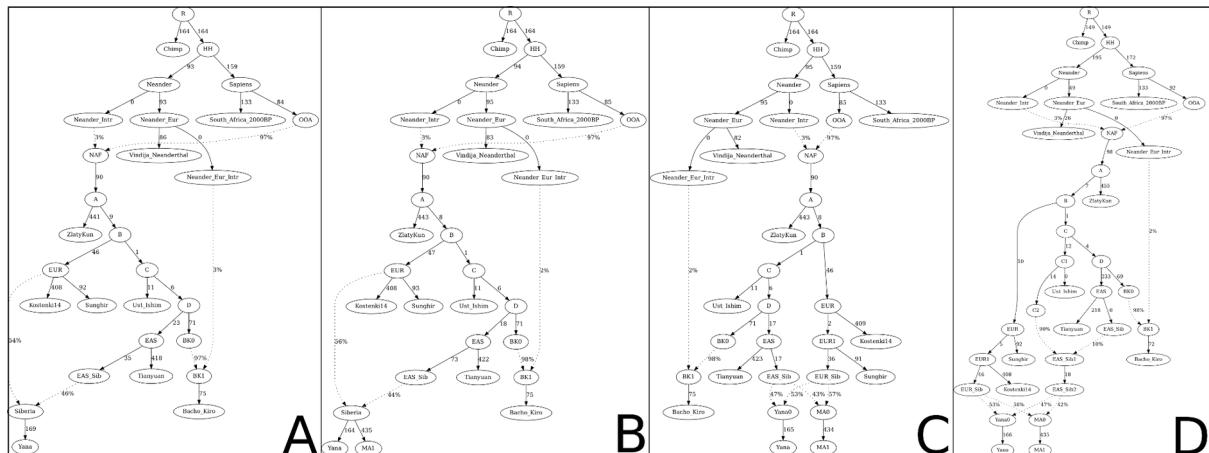
241 **Figure S5 Adding Paleolithic Siberians to the qpGraph of Figure S2.B.** We first placed
242 Yana1 on the graph as a putative mixture of West Eurasian (EUR) and East Asian (EAS)
243 components (**A**) and, after having obtained no outliers we proceeded with adding Mal'ta as a

3.7 Adding GoyetQ116-1 and BK-1653 to the emerging picture of IUP and UP interaction in Western Eurasia

The ~35000 years old GoyetQ116-1 individual from Belgium, although closer to Europeans than to East Asians, shares more alleles with the latter compared to other contemporary Europeans (Fu et al. 2016) and it has recently been described as a recipient of gene flow from a population related to Bacho Kiro (Hajdinjak et al. 2021).

We started from Figure S5.C and tried to describe Goyet as mixture between the Kostenki14/Sunghir branch and the Bacho Kiro branch (Figure S6.A); however doing so shows that Goyet needs to share more drift with Tianyuan (f2 Z-score = -2.5, plus several f4 outliers). On the other hand, sourcing the East Asian genetic component from the node upstream of Bacho Kiro and Tianyuan (node D) results in the Z scores of f2 with both Tianyuan and Bacho Kiro to be too low (-2.4 and -2.2 respectively), highlighting how its East Asian genetic component is more variegated than the one found in Bacho Kiro or Tianyuan alone (Figure S6.B).

When adding to Goyet a contribution from both the Bacho Kiro and Tianyuan branches (Figure S6.C) we obtained only two minor outliers (Chimp, South_Africa_2000BP, ZlatyKun, Yana = 3.4 and South_Africa_2000BP, ZlatyKun, ZlatyKun, Sunghir = -3.3), the first of which results not significant when computed using all SNPs (Table S4). Notably, the ~35 ky old Bacho Kiro individual: BK1653 (Hajdinjak et al. 2021), that is associated with Upper Paleolithic assemblages (Fewlass et al. 2020; Hublin et al. 2020) can instead be described more simply as a member of the UP genetic branch that received ~33% contribution from a population related to the older IUP Bacho Kiro individuals recovered from the same site with only a minor outlier (ZlatyKun, Tianyuan, Sunghir, MA1 with Z-score -3.3) which results as non significant when all SNPs are taken into account (Figure S6.D). In the modeling of Goyet we fixed the proportion of Neanderthal admixture in non Africans using the lock V option of qpGraph.
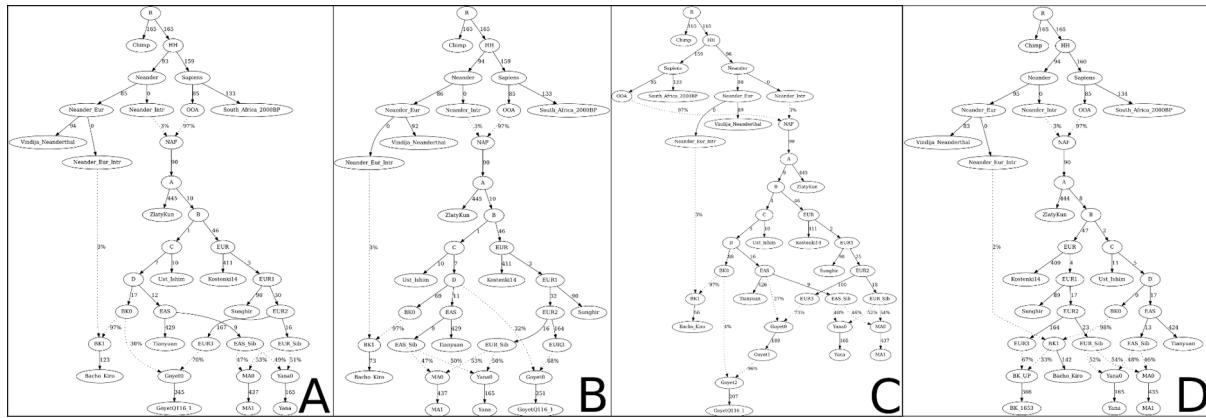
Figure S6 Goyet Q116-1 and BK1653 as a mixture of UP and IUP lineages. Modelling the East Asian component found in Goyet Q116-1 as a simple interaction between UP and the Bacho Kiro branch (**A**) or a node upstream of the Tianyuan-bacho Kiro split (**B**) yields an unexplained attraction between Goyet Q116-1 and Tianyuan. Modelling the IUP component as a mixture of Bacho Kiro- and Tianiyuan-related lineages (**C**) lineages provides a better fit to the data. A simple interaction between Bacho Kiro and the UP branch can instead explain the ancestry of BK1653(**D**). (Graphs A, B and C are based on 257802 SNPs, graph D is based on 300021 SNPs. Highest |Z-score| = 3.9 (A, B), 3.4 (C), -3.3 (D)).

3.8 Peopling of Oceania

The position of Oceanian populations with respect to the East and West Eurasian is still unclear, with Oceanians often seen as either an earlier split (Malaspinas et al. 2016; Choin et al. 2021) or a sister population to East Asians (Wall 2017; Mallick et al. 2016). We included Papuans in our model in order to address this.

We started from Graph S2.B and tried to have Papuans split from other Eurasians before, together or after the split of the Zlatý Kůň lineage; in all cases the graph was rejected as papuans needed to share more drift with Tianyuan (f2 Z-score: -11, -11 and -8). For this reason we then modeled them as a sister group of Tianyuan and obtained a graph whose only outlier (South_Africa_2000BP, Bacho Kiro, Zlatý Kůň, Papuan Z-score = -3.1) resulted non significant when taking all SNPs into account (Figure 1.B, Figure S7.A).

Subsequently we tried to model Papuans as a mixture of a Tianyuan sister population and a more basal lineage; the contribution from said lineage decreases the more that split is moved backward: it is 96% when splitting just before the split Tianyuan/Bacho Kiro (Figure S7.B), 42% when splitting before Ust'Ishim (Figure S7.C), 26% when splitting after Zlatý Kůň but before any other Eurasian branching (Figure S7.D) and only 2% when splitting before Zlatý Kůň (Figure S7.E). Finally, we decided to test whether a small contribution to Papuans from a population of Anatomically Modern Humans (AMH) that left Africa before the main

302 Out of Africa 70-60 thousands of years ago (kya) and otherwise extinct (xOoA) (Pagani et al.
303 2016) is rejected by the model (Figure S7.F). A 1% contribution from said population to
304 modern papuans is not rejected.
305 All the acceptable solutions for the placement of Papuans within the broader OoA tree
306 confirm Zlatý Kůň as the most basal human genome among the ones ever found Out of
307 Africa. In conclusion, Papuans can be described either as an almost even mixture (occurred
308 sometimes between 45 and 38 kya) between East Asians and a population splitting from
309 other Eurasians before the IUP expansion, or as a sister lineage of East Asians with or
310 without a minor basal OoA or xOoA contribution.
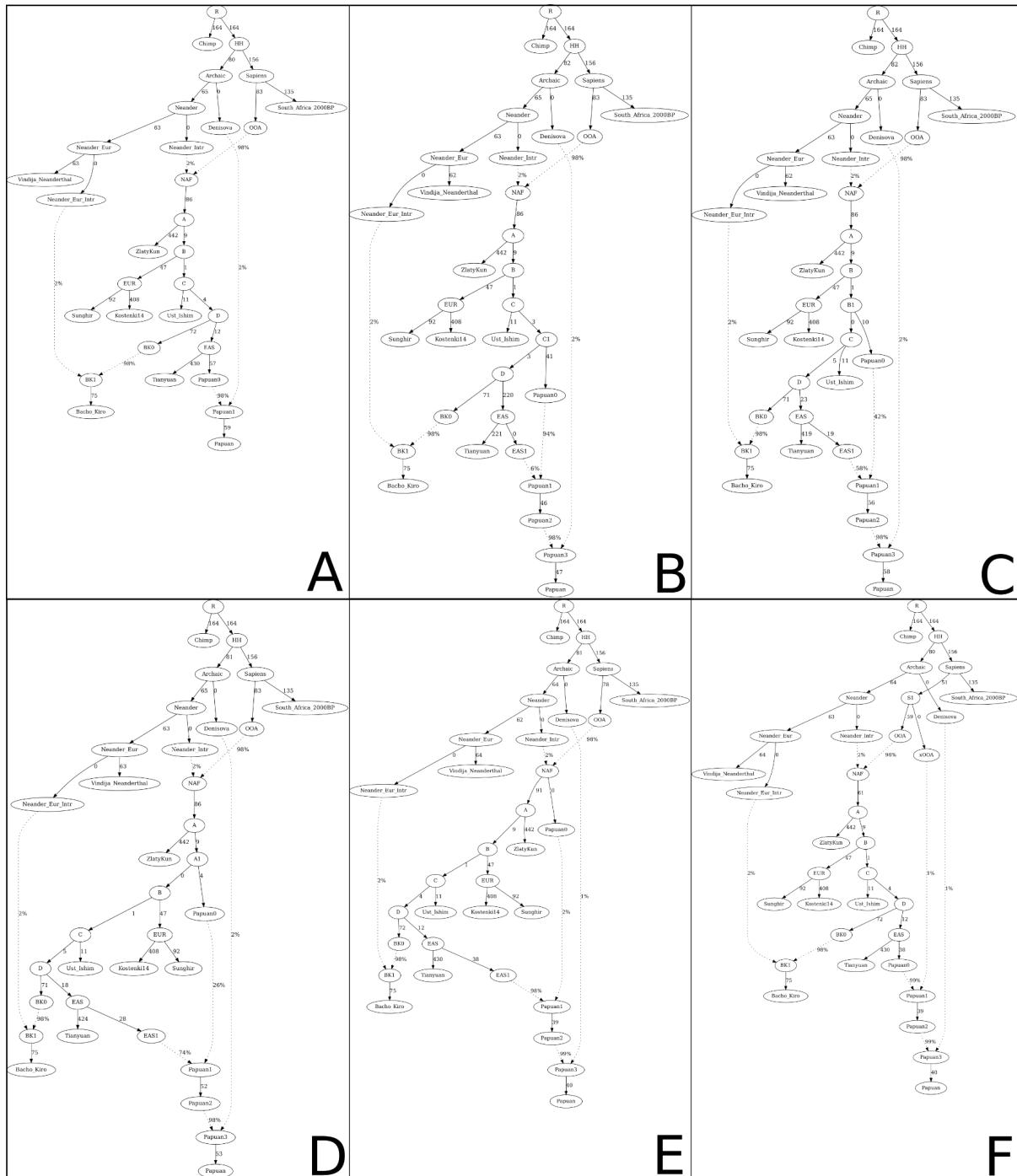
Figure S7 The place of Papuans within the out of Africa graph. Modern Papuans can be invariably described as a sister population of Tianyuan (**A**) with or without the addition of a more basal, *H. sapiens* lineage. The position of this deeper lineage along the population model influences the genetic contribution it gives to Papuans: 94% if placed within the IUP branch after Ust'Ishim (**B**), 42% before Ust'Ishim (**C**), 26% before the separation of IUP and UP branches (**D**), 2% as the most basal OoA branch (**E**) or even 1% as an extinct, extra OoA (xOoA) (**F**). (Graphs based on 418884 SNPs. Highest |Z-score| = 3.1 (A), 2.8(B, C, D, E, F)).

Table S4: Relevant f4 tests. The first seven columns report population and values as outputted by Admixtools. The eight column reports the Supplementary Section where a given f4 is mentioned.

Pop1	Pop2	Pop3	Pop4	D	Z	BABA	ABBA	SNPs	Section
Ust_Ishim	Kostenki14	Tianyuan	Chimp	0.0006	0.087	45718	45659	817238	3.1
Ust_Ishim	Sunghir	Tianyuan	Chimp	-0.0023	-0.34	47517	47732	847217	3.1
Tianyuan	Kostenki14	Ust_Ishim	Chimp	-0.0037	-0.523	45718	46054	817238	3.1
Tianyuan	Sunghir	Ust_Ishim	Chimp	0.0024	0.386	47517	47289	847217	3.1
Ust_Ishim	Tianyuan	Kostenki14	Chimp	0.0043	0.588	46054	45659	817238	3.1
Ust_Ishim	Tianyuan	Sunghir	Chimp	-0.0047	-0.758	47289	47732	847217	3.1
Bacho_Kiro	Kostenki14	Ust_Ishim	Chimp	-0.0276	**-4.337**	56016	59200	997593	3.2
Bacho_Kiro	Sunghir	Ust_Ishim	Chimp	-0.0194	**-3.581**	59440	61792	1052955	3.2
Bacho_Kiro	Tianyuan	Ust_Ishim	Chimp	-0.0228	**-3.628**	45780	47915	841030	3.2
Tianyuan	ZlatyKun	Bacho_Kiro	Chimp	0.0349	**5.005**	32014	29852	554809	3.2
Kostenki14	ZlatyKun	Bacho_Kiro	Chimp	0.0124	1.821	33982	33147	594315	3.2
Sunghir	ZlatyKun	Bacho_Kiro	Chimp	0.0144	2.352	35625	34612	620949	3.2
Ust_Ishim	ZlatyKun	Bacho_Kiro	Chimp	0.0194	2.856	35526	34172	619549	3.2
ZlatyKun	Tianyuan	Sunghir	MA1	0.0085	1.112	21533	21171	422908	2.6, 2.8
Chimp	South_Africa_2000BP	ZlatyKun	Yana	0.0122	2.864	25885	25262	625628	2.6, 2.8
South_Africa_2000BP	Bacho_Kiro	ZlatyKun	Papuan	0.0122	2.136	36159	35288	636119	3.8
Kostenki14	ZlatyKun	Papuan	Chimp	0.029	**4.028**	34260	32330	587236	3.8
Sunghir	ZlatyKun	Papuan	Chimp	0.027	**4.452**	35811	33929	614406	3.8
Ust_Ishim	ZlatyKun	Papuan	Chimp	0.0363	**5.213**	35842	33334	613032	3.8
Tianyuan	ZlatyKun	Papuan	Chimp	0.0736	**10.746**	33107	28566	547447	3.8

Supplementary Section 4: Unsupervised analysis with OrientAGraph/Treemix

Since qpGraph is a supervised method that relies on a topology provided by the user, we decided to test the robustness of our base model using an unsupervised method such as Treemix (Pickrell and Pritchard 2012), taking advantage of the Maximum Likelihood Network Orientation algorithm recently implemented in OrientAGraph (Molloy et al. 2021).

We selected the same set of samples used in supplementary section 3.3 and ran
328 OrientAGraph to heuristically identify the best fitting graph; our only input was specifying
329 Chimp as root and the gene flow from Neanderthal to Bacho Kiro. The Graph we obtained
330 (Figure S8) has the same topology of graph S2.B, with Bacho Kiro sister of Tianyuan and
331 Ust'Ishim splitting early from the IUP branch, although qpGraph appears to be a more
332 flexible option when multiple admixture events are required to properly explain the data.
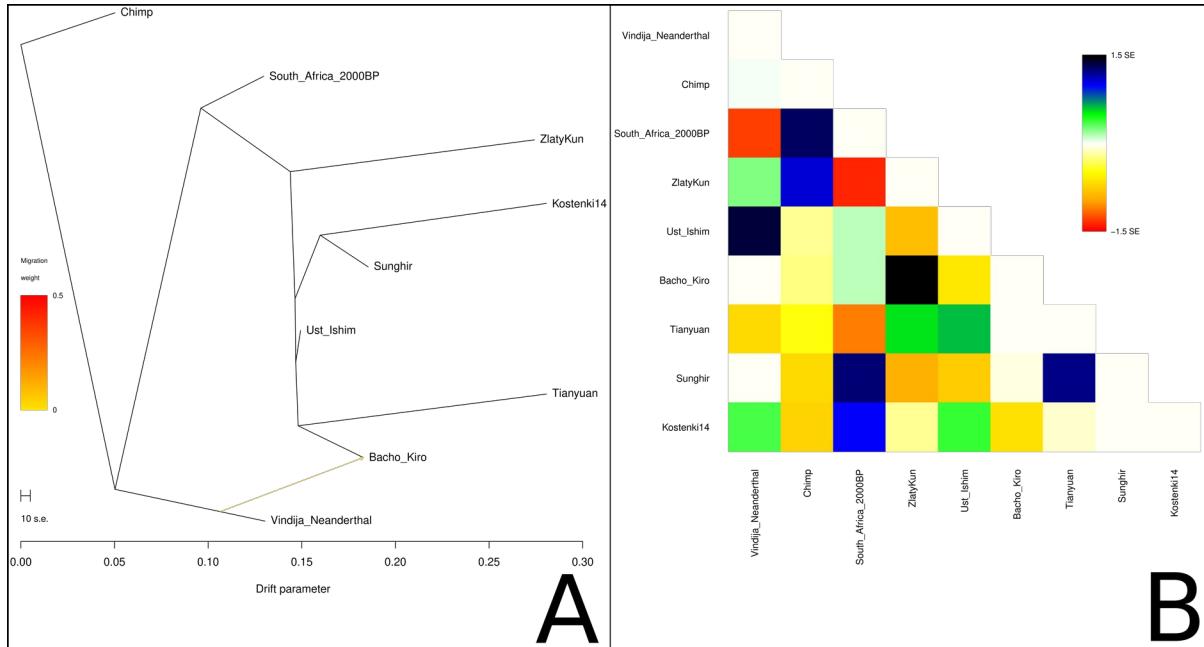
333 **Figure S8 Best fitting admixture graph with OrientAGraph/Treemix** Best fitting tree (A)
334 and associated residuals(B) obtained running OrientAGraph/Treemix while specifying only
335 chimp as root and the additional gene flow from Neanderthal to Bacho Kiro.

Supplementary Section 5: Paleomaps plotting

337 To plot the paleomaps, we used R (version 4.0.5 - "Shake and Throw"), the GUI RStudio
338 (version 1.4.1103 - "Wax Begonia") and the package "oce" (https://dankelley.github.io/oce/).
339 We downloaded the alti-bathymetric maps from the ETOPO1 dataset (Amante and Eakins
340 2009) and used the inferred sea level values at given times in the past from the "Global 1Ma
341 Temperature, Sea Level, and Ice Volume Reconstructions" dataset (Bintanja, van de Wal,
342 and Oerlemans 2005) available from the NOAA Paleoclimatology Program.
343 We plotted the maps applying Lambert conformal conic projections ("+proj=lcc +lat_1=25
344 +lat_2=50 +lon_0=97" in PROJ.4 format).

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345

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