Supporting Information

SI Materials and Methods

Microsatellite genotyping

Elephant microsatellite loci were selected from a range of studies on both African and Asian elephants (Nyakaana & Arctander 1998; Comstock *et al.* 2000; Eggert *et al.* 2000; Kongrit *et al.* 2008). Fifteen loci were chosen out of a total of 41, on the basis of simple repeating sequence, relatively short product length and variability in both elephant species (Table S1). These loci were screened for amplification success using five mammoth samples that previously had been successful for mtDNA analysis. PCR reactions for four loci that amplified successfully and displayed genetic variability were subsequently optimized using three different annealing temperatures (±2 °C from the calculated annealing temperature) as well as two different $MgCl_2$ concentrations (1.5 and 2.0 mM for loci EMU10 & EMU13; 2.0 and 2.17 mM for loci FH67 & FH71).

One sample from each locus was cloned, using the TOPO-TA cloning kit for sequencing (Invitrogen), and two individual clones from each sample were amplified. The resulting PCR-products were cleaned with Exonuclease I (Fermentas) and FastAP (Fermentas) and sequenced using the BigDye Terminator v.1.1 cycle sequencing kit (Applied Biosystems) and analyzed on an ABI 3130xl automated sequencer (Applied Biosystems). All clone sequences, both in terms of repeat type and flanking region sequence, were identical to the previously published microsatellite sequences for elephants (GenBank accession numbers: EF643832, EF643835, A206283, A206282). The only difference compared to the elephant sequences was in the number of repeats observed, which is to be expected.

SI Tables

Table S1 Microsatellite loci with repeat unit, primer sequence and size range in African and Asian

elephants. The four loci selected for this study are marked with an asterisk (*) and shows the size range observed in mammoth.

Locus	Repeat	Primer sequence (5'-3')	Size	Reference
FH67*	$(GT)_{15}$	F: GCTTCTCTAGAAATGTGTATGC R: GGCGTATAGGATAGTTCCAC	89-93	Comstock *et al.* 2000
FH71*	$(CA)_{14}$	F: GGGATTGGCTAAAATAG R: CTAAGCACATCAGGGAC	65-89	Comstock *et al.* 2000
EMU10*	$(CA)_{17}$	F: AATCGACTCAGCAGCAACAG R: CCAGTAAATCCATATCACTCGTC	94-104	Eggert *et al.* 2000
EMU13*	$(GT)_{17}$	F: GTATTTGGGCTGGCATGGT R: GTGGGGTCTGTGGTCAAGTG	100-110	Eggert *et al.* 2000
EMU01	$(GT)_{12}$	F: TTTTCTTTGGTCCCCATGAT R: AGACCTTGGGTCTTGTGCTG	78-82	Eggert *et al.* 2000
EMU04	$(TG)_{12}$	F: TGACTCTCCCTCTTCTGCATC R: GGCTGAGAGGGAAAGAAATTG	97-107	Eggert *et al.* 2000
EMU07	$(TG)_{15}$	F: GAGCAGTGCCTTTCGTGAC R: AGCCTGGGAGGTAAGTAGCA	100-124	Eggert *et al.* 2000
FH1	$(CA)_{12}$	F: GATCAGACCATGGCATGAG R: ACAGTCTCCCTTGGGAAGAC	81	Comstock *et al.* 2000
FH60	$(CA)_{13}$	F: CAAGAAGCTTTGGGATTGGG R: CCTGCAGCTCAGAACACCTG	148	Comstock *et al.* 2000
FH103	$(CA)_{13}$	F: TGTGCTGCCACTTCCTACAC R: GATGTTGAGACAGTTCTGTAAG	154	Comstock *et al.* 2000
LA3	$(CA)_{10}$	F: TACTCTGCTCCTCTGCCTATCC R: GCAGAATTTTGGTCTTGGAGG	165-171 166-172	Kongrit *et al.* 2008
LA5	$(CA)_{13}$	F: GGGCAGCCTCCTTGTTTT R: CTGCTTCTTTCATGCCAATG	130-154 142-144	Kongrit *et al.* 2008
LA6	$(CA)_{13}$	F: AAAATTGACCCAACGGCTC R: TCACGTAACCACTGCGCTAC	158-214 155-159	Kongrit *et al.* 2008
LafMS02	$(AC)_{16}$	F: GAAACCACAACTTGAAGGG R: TCGCTTGTAAGAAGGCGTG	149	Nyakaana & Arctander 1998
LafMS03	$(TG)_{15}$	F: CATATGAACATACCGGAAC R: GAAACTCCTCGAGTAGTAGAA	142	Nyakaana & Arctander 1998

Table S2 Estimated probability (*P)* of falsely accepting a homozygote, given the observed rate of allelic dropout (*K*), after three replicates for each locus according to Gagneux *et al.* (1997).

	EMU10	EMU13	FH67	FH71
K	0.14	0.45	0.26	0.33
P	0.0006	0.023	0.004	0.009

Table S3 Estimated mean likelihoods of data and posterior probabilities for each cluster (*K* = 1-5) for (a)

the complete data set and (b) where individuals containing missing data were excluded from the analyses.

	(a) n = 59		(b) n = 44	
K	Mean ln $Pr(X\backslash K)$	$Pr(X\backslash K)$	Mean ln $Pr(X\backslash K)$	$Pr(X\backslash K)$
1	-553.74	0.000	-436.68	0.000
2	-510.48	0.999	-402.63	0.999
3	-521.61	0.000	-414.57	0.000
4	-544.72	0.000	-436.81	0.000
5	-576.37	0.000	-493.44	0.000

Table S4 Genetic variation in each locus and time period, after excluding individuals containing missing data. H_O = observed heterozygosity, H_E = expected heterozygosity.

Locus	>62-12 ka (n = 20)		9-3.7 ka (n = 24)	
	H_O	H_E	H_O	H_E
EMU10	0.70	0.51	0.21	0.30
EMU13	0.60	0.70	0.46	0.37
FH67	0.45	0.48	0.42	0.38
FH71	0.95	0.93	0.71	0.71
Mean±s.d.	0.68±0.21	0.65±0.21	0.45±0.21	0.44±0.19

Table S5 The observed allele frequencies for each locus and time period.

Locus	Allele	>62-12 ka (n = 27)	9-3.7 ka (n = 32)
EMU10	78	0.042	-
	84	0.583	0.844
	86	0.375	0.094
	90	-	0.063
EMU13	100	0.220	0.038
	102	0.400	-
	104	0.300	0.731
	106	0.080	-
	110	-	0.231
FH67	89	0.048	-
	91	0.690	0.724
	93	0.262	0.276
FH71	65	0.037	-
	67	0.093	-
	69	0.019	-
	71	0.111	-
	73	0.130	-
	75	0.148	0.033
	77	0.093	0.217
	79	0.074	0.017

81	0.130	0.217
83	0.037	0.017
85	0.037	0.467
87	0.056	-
89	0.037	0.033

SI Figures

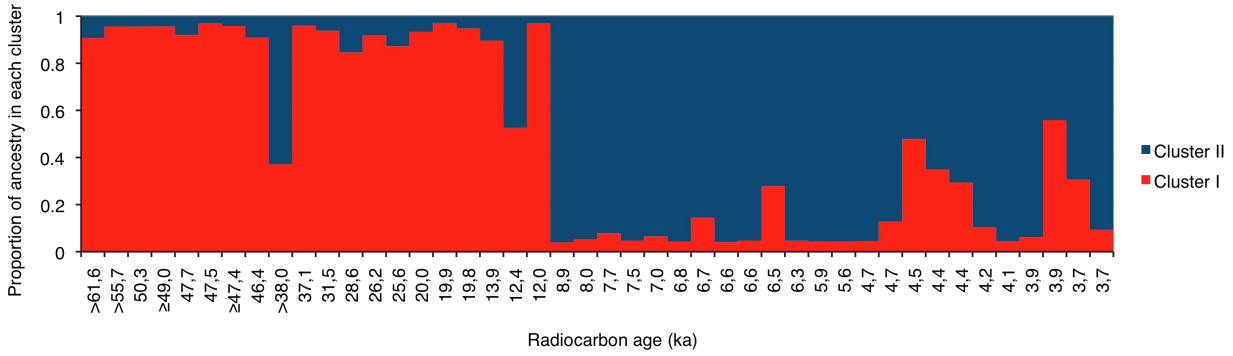

Fig. S1 Estimated population structure from analyses performed on individuals with complete genotypes (*n* = 44). The figure shown is based on the mean Q-values computed in CLUMPP from 10 clustering solutions obtained in STRUCTURE assuming two clusters. Each bar represents one individual and shows its cluster membership. The radiocarbon age (ka) is given below each individual.

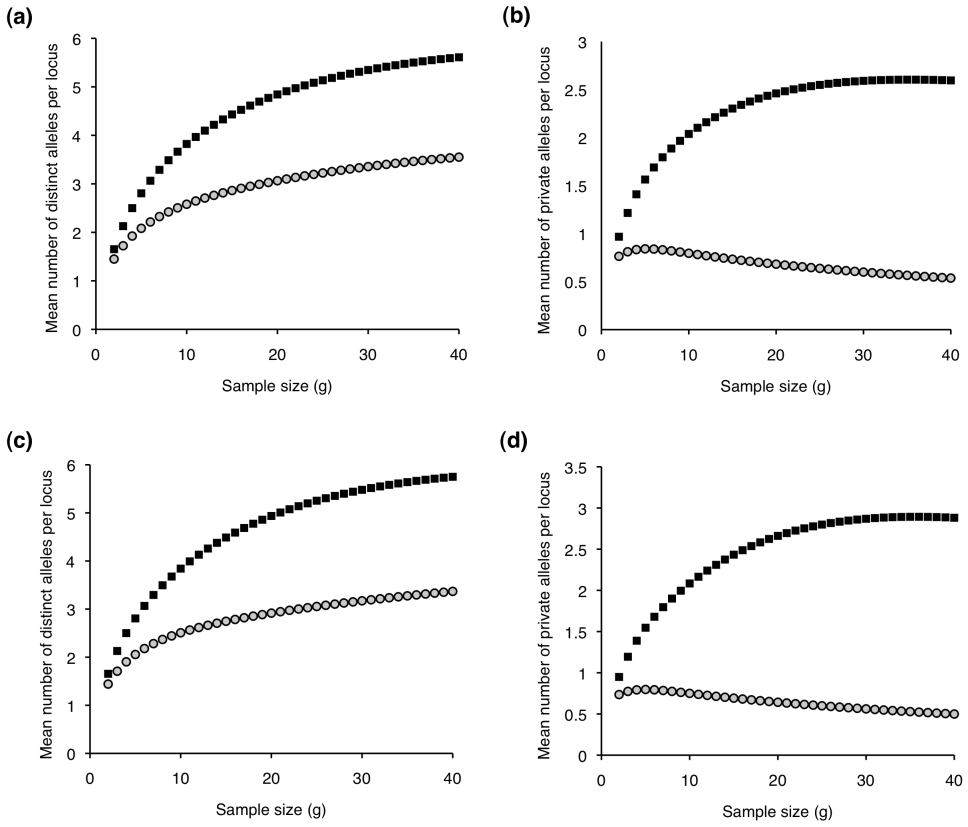

Fig. S2 Variability as a function of standardized sample size for samples from before (black) and after (grey) 12 ka. (a) The mean number of distinct alleles per locus (allelic richness). (b) The mean number of private alleles per locus. Panels (c) and (d) correspond to (a) and (b) respectively, but show the analyses where only individuals with complete genotypes (n = 44) were included.

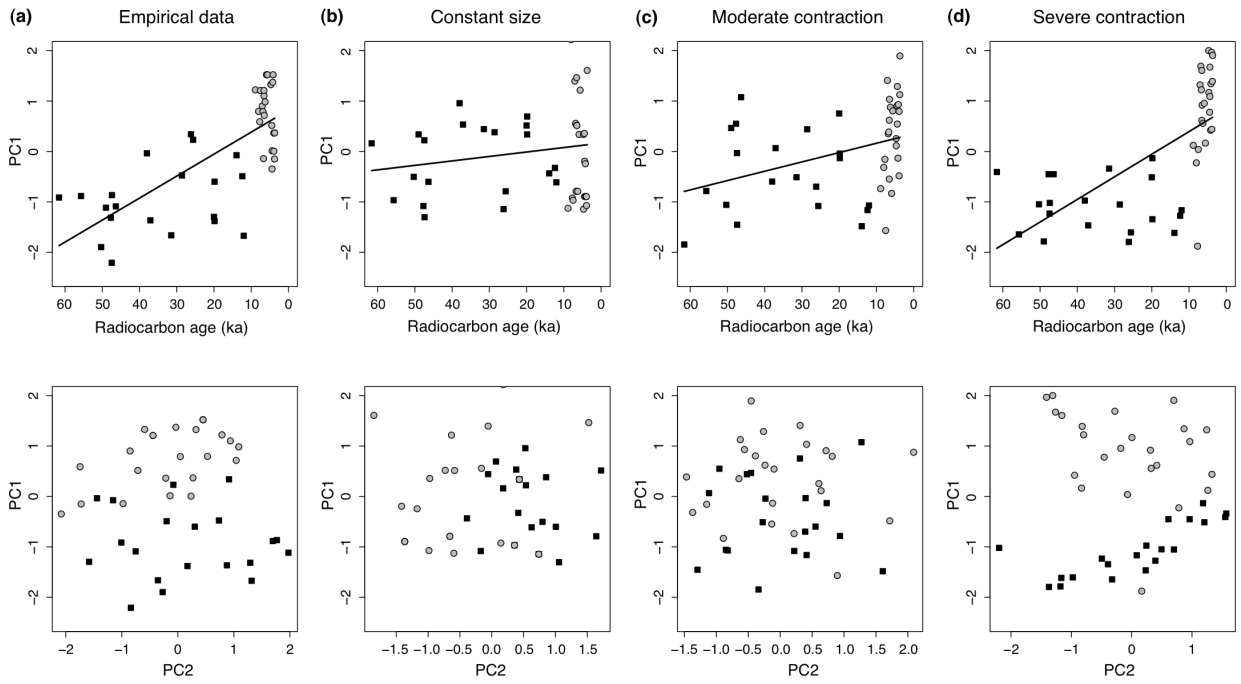

Fig. S3 Posterior predictive simulations recapitulating the correlation between PC1 and time when the effective population size after 12 ka is severely contracted. Samples >12 ka are shown as black squares and samples <9 ka are shown as grey circles. PC1 plotted against time is shown on top and PC1 vs. PC2 on bottom. (a) Empirical data. (b) A model with a constant low size of N_e = 2,000. (c) A model with a demographic event at 12 ka where N_e changed from 20,000 to 2,000. (d) A model where N_e changed from 20,000 to 700.

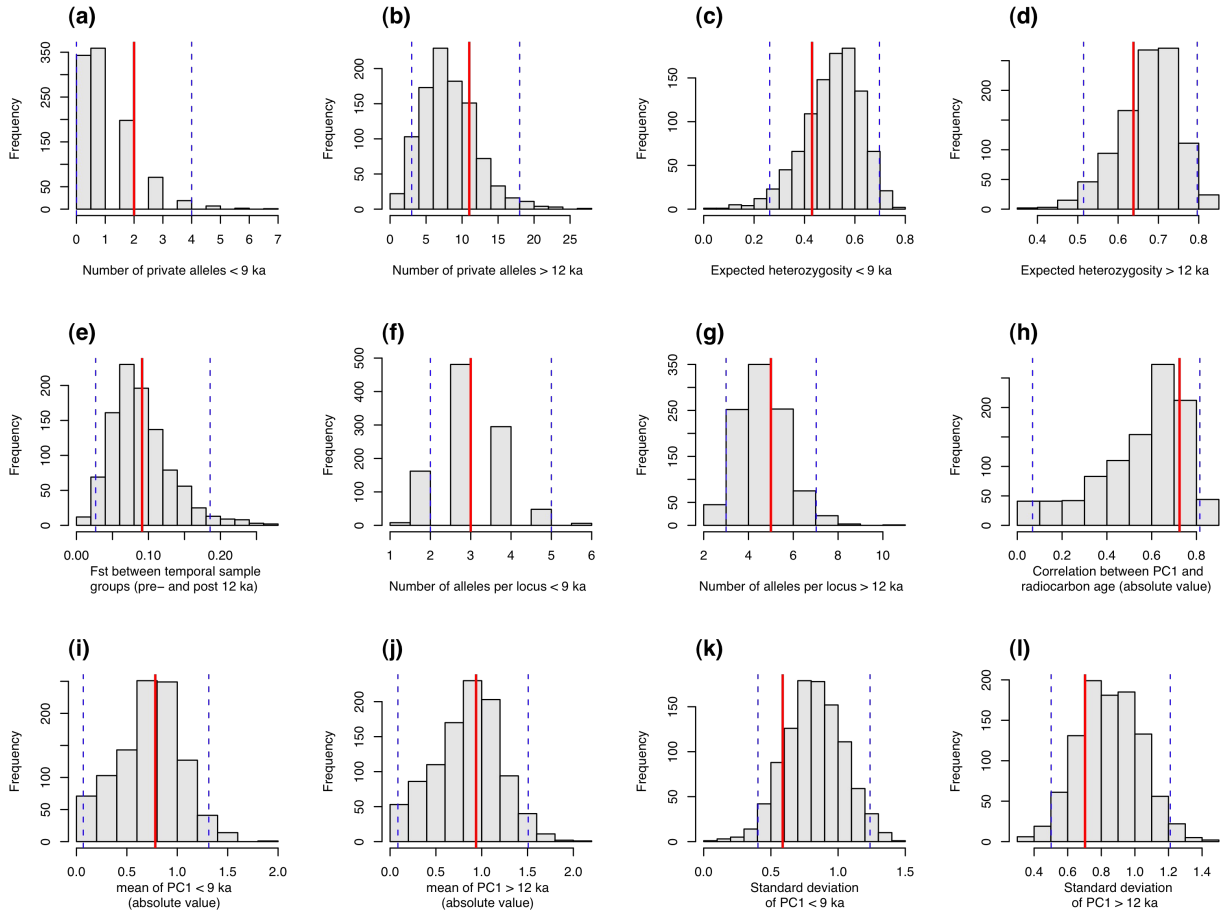

Fig. S4 Marginal posterior predictive distribution for each of 12 different summary statistics based on posterior parameter distributions obtained for the 2-epoch model. The empirical estimate of each summary statistic based on mammoth microsatellite data is indicated with a solid red line. The central 95% interval is indicated with blue dashed lines.

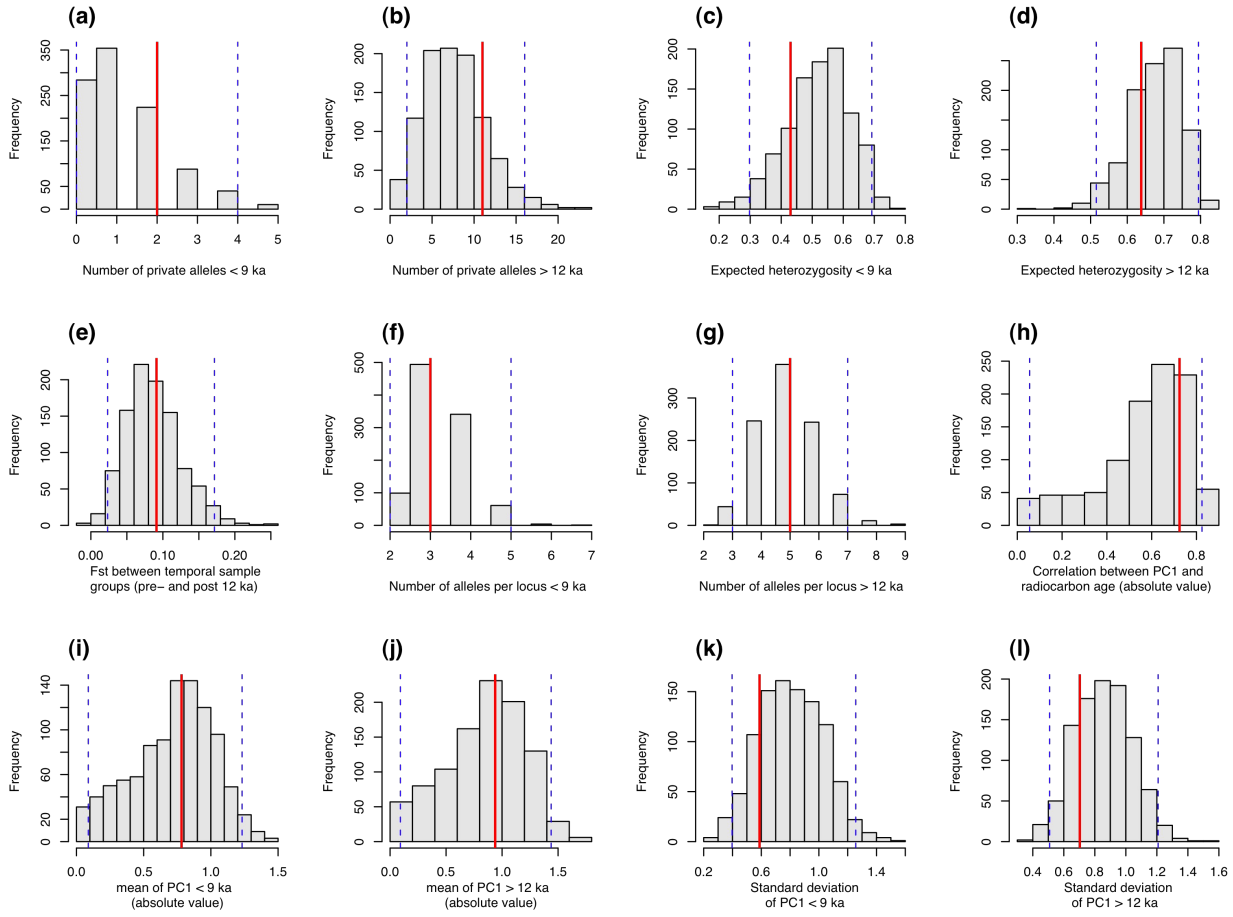

Fig. S5 Marginal posterior predictive distribution for each of 12 different summary statistics based on posterior parameter distributions obtained for the 3-epoch model. The empirical estimate of each summary statistic based on mammoth microsatellite data is indicated with a solid red line. The central 95% interval is indicated with blue dashed lines.

Author contributions

V.N. designed the project, performed DNA analyses, computed population-genetic statistics, carried out Bayesian clustering analyses and co-wrote the paper; J.H. optimized the genotyping and performed DNA analyses; P.S. designed, performed and wrote the text on the PC analysis and the approximate Bayesian demographic analysis under supervision from M.J., who also computed population-genetic summary statistics; N.J.M. helped with the genotyping; S.V. contributed with material and data; P.W.S., K.L., I.B., A.A. and A.L. contributed with resources and information, and helped interpret the data; L.D. conceived and designed the project, performed DNA analyses, and co-wrote the paper. All authors discussed the results and contributed to the preparation of the manuscript.

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