Ancient Biomolecules from Deep Ice Cores Reveal a Forested Southern Greenland

Eske Willerslev, *et al.*

Science **317**, 111 (2007);
DOI: 10.1126/science.1141758

Science (print ISSN 0036-8075; online ISSN 1095-9203) is published weekly, except the last week in December, by the American Association for the Advancement of Science, 1200 New York Avenue NW, Washington, DC 20005.

23. L. Mehl, B. R. Hacker, G. Hirth, P. B. Kelemen, *J. Geophys. Res.* **108**, 10.1029/2002JB002233 (2003).
24. B. R. Jicha, D. W. Scholl, B. S. Singer, G. M. Yogodzinski, S. M. Kay, *Geology* **34**, 661 (2006).
25. C.-T. Lee, X. Cheng, U. Horodyskyj, *Contrib. Min. Petrol.* **151**, 222 (2006).
26. P. B. Kelemen, K. Hanghøj, A. R. Greene, in *The Crust*, R. L. Rudnick, Ed. (Elsevier-Pergamon, Oxford, 2003), vol. 3, pp. 593–659.
27. T. V. Gerya, D. A. Yuen, *Earth Planet. Sci. Lett.* **212**, 47 (2003).
28. We thank M. Long, E. Kneller, and C. Conrad for conversations that motivated this work. Funding was provided by NSF grants EAR-9910899, EAR-0125919, and EAR-0509882.

Supporting Online Material
www.sciencemag.org/cgi/content/full/317/5834/108/DC1
SOM Text
Figs. S1 and S2
References

13 February 2007; accepted 9 May 2007
10.1126/science.1141269

Ancient Biomolecules from Deep Ice Cores Reveal a Forested Southern Greenland

Eske Willerslev,[1]* Enrico Cappellini,[2] Wouter Boomsma,[3] Rasmus Nielsen,[4] Martin B. Hebsgaard,[1] Tina B. Brand,[1] Michael Hofreiter,[5] Michael Bunce,[6,7] Hendrik N. Poinar,[7] Dorthe Dahl-Jensen,[8] Sigfus Johnsen,[8] Jørgen Peder Steffensen,[8] Ole Bennike,[9] Jean-Luc Schwenninger,[10] Roger Nathan,[10] Simon Armitage,[11] Cees-Jan de Hoog,[12] Vasily Alfimov,[13] Marcus Christl,[13] Juerg Beer,[14] Raimund Muscheler,[15] Joel Barker,[16] Martin Sharp,[16] Kirsty E. H. Penkman,[2] James Haile,[17] Pierre Taberlet,[18] M. Thomas P. Gilbert,[1] Antonella Casoli,[19] Elisa Campani,[19] Matthew J. Collins[2]

It is difficult to obtain fossil data from the 10% of Earth's terrestrial surface that is covered by thick glaciers and ice sheets, and hence, knowledge of the paleoenvironments of these regions has remained limited. We show that DNA and amino acids from buried organisms can be recovered from the basal sections of deep ice cores, enabling reconstructions of past flora and fauna. We show that high-altitude southern Greenland, currently lying below more than 2 kilometers of ice, was inhabited by a diverse array of conifer trees and insects within the past million years. The results provide direct evidence in support of a forested southern Greenland and suggest that many deep ice cores may contain genetic records of paleoenvironments in their basal sections.

The environmental histories of high-latitude regions such as Greenland and Antarctica are poorly understood because much of the fossil evidence is hidden below kilometer-thick ice sheets (*1*–*3*). We test the idea that the basal sections of deep ice cores can act as archives for ancient biomolecules.

The samples studied come from the basal impurity-rich (silty) ice sections of the 2-km-long Dye 3 core from south-central Greenland (*4*), the 3-km-long Greenland Ice Core Project (GRIP) core from the summit of the Greenland ice sheet (*5*), and the Late Holocene John Evans Glacier on Ellesmere Island, Nunavut, northern Canada (Fig. 1). The last-mentioned sample was included as a control to test for potential exotic DNA because the glacier has recently overridden a land surface with a known vegetation cover (*6*). As an additional test for long-distance atmospheric dispersal of DNA, we included five control samples of debris-free Holocene and Pleistocene ice taken just above the basal silty samples from the Dye 3 and GRIP ice cores (Fig. 1B). Finally, our analyses included sediment samples from the Kap København Formation from the northernmost part of Greenland, dated to 2.4 million years before the present (Ma yr B.P.) (*1*, *2*).

The silty ice yielded only a few pollen grains and no macrofossils (*7*). However, the Dye 3 and John Evans Glacier silty ice samples showed low levels of amino acid racemization (Fig. 1A, inset), indicating good organic matter preservation (*8*). Therefore, after previous success with permafrost and cave sediments (*9*–*11*), we attempted to amplify ancient DNA from the ice. This was done following strict criteria to secure authenticity (*12*–*14*), including covering the surface of the frozen cores with plasmid DNA to control for potential contamination that may have entered the interior of the samples through cracks or during the sampling procedure (*7*). Polymerase chain reaction (PCR) products of the plasmid DNA were obtained only from extracts of the outer ice scrapings but not from the interior, confirming that sample contamination had not penetrated the cores.

Using PCR, we could reproducibly amplify short amplicons [59 to 120 base pairs (bp)] of the chloroplast DNA (cpDNA) *rbc*L gene and *trn*L intron from ~50 g of the interior ice melts from the Dye 3 and the John Evans Glacier silty samples. From Dye 3, we also obtained 97-bp amplicons of invertebrate cytochrome oxidase subunit I (COI) mitochondrial DNA (mtDNA). Attempts to reproducibly amplify DNA from the GRIP silty ice and from the Kap København Formation sediments were not successful. These results are consistent with the amino acid racemization data demonstrating superior preservation of biomolecules in the Dye 3 and John Evans Glacier silty samples, which is likely because these samples are colder (Dye 3) or younger (John Evans Glacier) than the GRIP sample (Fig. 1A, inset). We also failed to amplify DNA from the five control samples of Holocene and Pleistocene ice taken just above the silty samples from the Dye 3 and GRIP ice cores (volumes: 100 g to 4 kg; Fig. 1B) (*7*). None of the samples studied yielded putative sequences of vertebrate mtDNA.

A previous study has shown that simple comparisons of short DNA sequences to GenBank sequences by means of the Basic Local Alignment Search Tool (BLAST) make misidentification likely (*15*). Therefore, we assigned the obtained sequences to the taxonomic levels of order, family, or genus using a new rigorous statistical approach (*7*). In brief, this Bayesian method calculates the probability that each sequence belongs to a particular clade by considering its position in a phylogenetic tree based on similar GenBank sequences. In the calculation of these probabilities, uncertainties regarding phylogeny, models of evolution, and missing data are taken into account. Sequences with >90% posterior probability of membership to a taxonomic group were assigned to that group. Additionally, a given plant taxon was only considered genuine if sequences assigned to that taxon were found to be reproducibly obtained in separate analyses (by independent laboratories for the Dye 3 sample and within the laboratory for the John Evans Glacier control sample). This strict

[1]Centre for Ancient Genetics, University of Copenhagen, Denmark. [2]BioArch, Departments of Biology and Archaeology, University of York, UK. [3]Bioinformatics Centre, University of Copenhagen, Denmark. [4]Centre for Comparative Genomics, University of Copenhagen, Denmark. [5]Max Planck Institute for Evolutionary Anthropology, Germany. [6]Murdoch University Ancient DNA Research Laboratory, Murdoch University, Australia. [7]McMaster Ancient DNA Center, McMaster University, Canada. [8]Ice and Climate, University of Copenhagen, Denmark. [9]Geological Survey of Denmark and Greenland, Denmark. [10]Research Laboratory for Archaeology and the History of Art, University of Oxford, UK. [11]Department of Geography, Royal Holloway, University of London, UK. [12]Department of Earth Sciences, University of Oxford, UK. [13]Paul Scherrer Institut (PSI)/Eidgenössische Technische Hochschule (ETH) Laboratory for Ion Beam Physics, Institute for Particle Physics, ETH Zurich, Switzerland. [14]Swiss Federal Institute of Aquatic Science and Technology (EAWAG), Switzerland. [15]GeoBiosphere Science Center, Lund University, Sweden. [16]Department of Earth and Atmospheric Sciences, University of Alberta, Canada. [17]Ancient Biomolecules Centre, Oxford University, UK. [18]Laboratoire d'Ecologie Alpine, CNRS Unité Mixte de Recherche 5553, Université Joseph Fourier, Boîte Postale 53, 38041 Grenoble Cedex 9, France. [19]Dipartimento di Chimica Generale e Inorganica, Università di Parma, Italy.

*To whom correspondence should be addressed. E-mail: ewillerslev@bi.ku.dk

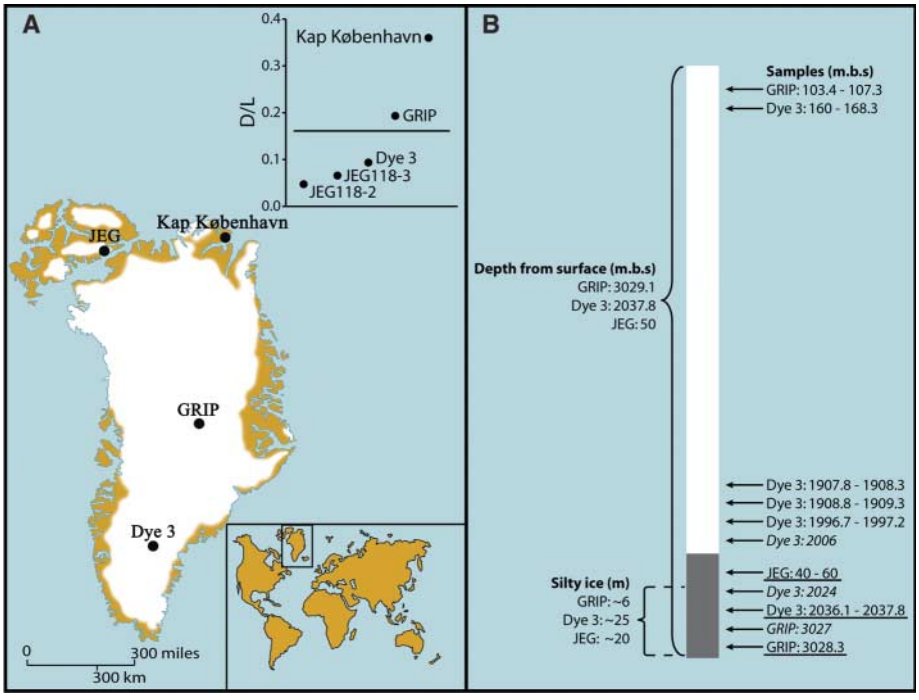

Fig. 1. Sample location and core schematics. (**A**) Map showing the locations of the Dye 3 (65°11'N, 45°50'W) and GRIP (72°34'N, 37°37'W) drilling sites and the Kap København Formation (82°22'N, W21°14'W) in Greenland as well as the John Evans Glacier (JEG) (79°49'N, 74°30'W) on Ellesmere Island (Canada). The inset shows the ratio of D– to L–aspartic acid, a measure of the extent of protein degradation; more highly degraded samples (above the line) failed to yield amplifiable DNA. (**B**) Schematic drawing of ice core/icecap cross section, with depth [recorded in meters below the surface (m.b.s.)] indicating the depth of the cores and the positions of the Dye 3, GRIP, and JEG samples analyzed for DNA, DNA/amino acid racemization/luminescence (underlined), and $^{10}Be/^{36}Cl$ (italic). The control GRIP samples are not shown. The lengths (in meters) of the silty sections are also shown.

criterion of authenticity obviously dismisses many putative taxa that are present at low abundance or have heterogeneous distributions, as is typical of environmental samples (*16*), but efficiently minimizes the influence of possible low-level contamination and misidentifications due to DNA damage (*17*).

Approximately 31% of the sequences from the John Evans Glacier silty sample were assigned to plant taxa that passed the authentication and identification criteria. These belong to the order Rosales, the family Salicaceae, and the genus *Saxifraga* (Table 1). This result is consistent with the John Evans Glacier forming no more than a few thousand years ago in a high Arctic environment (*18*), characterized by low plant diversity and sparse vegetation cover similar to that currently surrounding the glacier, which consists mainly of Arctic willow (Salicaceae), purple saxifrage (*Saxifraga*), *Dryas* (Rosales), and Arctic poppy (*Papaver*) (*19*). Thus, by confirming the expected result, the John Evans Glacier study can be regarded as a positive control, showing that DNA data from silty ice reliably record the local ecology.

In contrast to the John Evans Glacier silty sample, the 45% of the Dye 3 DNA sequences that could be assigned to taxa reveal a community very different from that of Greenland today. The taxa identified include trees such as alder (*Alnus*), spruce (*Picea*), pine (*Pinus*), and members of the yew family (Taxaceae) (Table 1). Their presence indicates a northern boreal forest ecosystem rather than today's Arctic environment. The other groups identified, including Asteraceae, Fabaceae, and Poaceae, are mainly

Table 1. Plant and insect taxa obtained from the JEG and Dye 3 silty ice samples. For each taxon (assigned to order, family, or genus level), the genetic markers (*rbc*L, *trn*L, or COI), the number of clone sequences supporting the identification, and the probability support (in percentage) are shown. Sequences have been deposited in GenBank under accession numbers EF588917 to EF588969, except for seven sequences less than 50 bp in size that are shown below. Their taxon identifications are indicated by symbols.

Order	Marker	Clones	Support (%)	Family	Marker	Clones	Support (%)	Genus	Marker	Clones	Support (%)
JEG sample											
Rosales	*rbc*L	3	90–99								
Malpighiales	*rbc*L	2	99–100	Salicaceae	*rbc*L	2	99–100				
	*trn*L	5	99–100		*trn*L	4	100				
Saxifragales	*rbc*L	3	92–94	Saxifragaceae	*rbc*L	2	92	*Saxifraga*	*rbc*L	2	91
Dye 3 sample											
Coniferales	*rbc*L	44	97–100	Pinaceae*	*rbc*L	20	100	*Picea*	*rbc*L	20	99–100
	*trn*L	27	100		*trn*L	25	100	*Pinus*†	*trn*L	17	90–99
				Taxaceae‡	*rbc*L	23	91–98				
					*trn*L	2	100				
Poales§	*rbc*L	67	99–100	Poaceae§	*rbc*L	67	99–100				
	*trn*L	17	97–100		*trn*L	13	100				
Asterales	*rbc*L	18	90–100	Asteraceae	*rbc*L	2	91				
	*trn*L	27	100		*trn*L	27	100				
Fabales	*rbc*L	10	99–100	Fabaceae	*rbc*L	10	99–100				
	*trn*L	3	99		*trn*L	3	99				
Fagales	*rbc*L	10	95–99	Betulaceae	*rbc*L	8	93–97	*Alnus*	*rbc*L	7	91–95
	*trn*L	12	100		*trn*L	11	98–100		*trn*L	9	98–100
Lepidoptera	COI	12	97–99								

*Env_2, *trn*L ATCCGGTTCATGAAGACAATGTTTCTTCTCCTAAGATAGGAAGGG. Env_3, *trn*L ATCCGGTTCATGAAGACAATGTTTCTTCTCCTAATATAGGAAGGG. Env_4, *trn*L ATCCGGTTCATGAGGACAATGTTTCTTCTCCTAATATAGGAAGGG. †Env_5, *trn*L CCCTTCCTATCTTAGGAGAAGAAACATTGTCTTCATGAACCGGAT. Env_6, *trn*L TTTCCTATCTTAGGAGAAGAAACATTGTCTTCATGAACCGGAT. ‡Env_1, *trn*L ATCCGTATTATAGGAACAATAATTTTATTTTCTAGAAAAGG. §Env_7, *trn*L CTTTTCCTTTGTATTCTAGTTCGAGAATCCCTTCTCAAAACACGGAT.

herbaceous plants and are represented by many species found in northern regions at present (Table 1). The presence of these herb-dominated families suggests an open forest where heliophytes thrived. Additionally, we recorded taxa that are common in present-day Arctic and/or boreal regions but lacked identity between independent laboratories. These are yarrow (*Achillea*), birch (*Betula*), chickweed (*Cerastium*), fescue (*Festuca*), rush (*Luzula*), plantain (*Plantago*), bluegrass (*Poa*), saxifrage (*Saxifraga*), snowberry (*Symphoricarpos*), and aspen (*Populus*). Although not independently authenticated at the sequence level, the presence of these taxa adds further support to the former existence of a northern boreal forest ecosystem at Dye 3.

To date, the youngest well-dated fossil evidence of native forest in Greenland is from macrofossils in the deposits of the Kap København Formation from the northernmost part of Greenland and dates back to around 2.4 Ma (*1*, *2*). Other less well dated traces of forests in Greenland include wood at two other late Cenozoic sites in northern Greenland (*20*), pollen spectra of unknown age in marl concretions found in a late glacial moraine, and wood and spruce seeds in eastern Greenland (*21*). The core from Dye 3, located almost exactly 2000 km to the southwest of the Kap København Formation (Fig. 1A), therefore, provides direct evidence of a forested southern-central Greenland.

The invertebrate sequences obtained from the Dye 3 silty ice are related to beetles (Coleoptera), flies (Diptera), spiders (Arachnida), brushfoots (Nymphalidae), and other butterflies and moths (Lepidoptera) (taxonomic identification probability support between 50 and 99%). However, only sequences of the Lepidoptera are supported by more than 90% significance (Table 1). Thus, although detailed identifications of the COI sequences are in general not strongly supported, the results show that DNA from a variety of invertebrates can be obtained from sediments even in the absence of macrofossils, as was previously shown for plants, mammals, and birds (*9*–*11*).

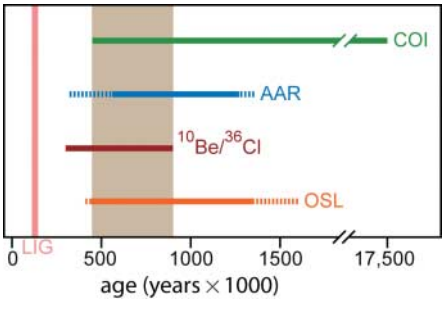

Fig. 2. Summary of dating results for the silty ice from Dye 3. From top to bottom, the bars indicate: maximum likelihood estimates for the branch length of the invertebrate COI sequences (COI); amino acid racemization results with the use of alternative activation energies, models of racemization behavior, and basal temperature histories (AAR); age estimate from ^{10}Be/^{36}Cl measurements in silty ice; and minimum ages based on single-grain luminescence results (optically stimulated luminescence or OSL). The time span covered by all dating methods (450 to 800 ka) is marked in gray. Stippled lines represent the results of less likely models. The maximum age estimate for the invertebrate COI sequences is based on an unlikely slow substitution rate [for details, see text and (*6*)].

Several observations suggest that the DNA sequences we obtained from the Dye 3 ice are of local origin and not the result of long-distance dispersal. The reproducible retrieval of diverse DNA from the silty basal ice but not from similar or larger volumes of the overlying "clean" ice largely precludes long-distance atmospheric dispersal of microfossils as a source of the DNA.

Although pollen grains are found in the Greenland ice sheet, including the Dye 3 silty ice (*7*), the concentrations are in general too low [~15 grains per liter (*22, 23*)] for them to be present in the sample volumes studied. Furthermore, long-term survival of DNA in pollen has proved fairly poor (*24*), and the vast majority of angiosperm pollen does not contain cpDNA (*25*). These factors effectively exclude pollen as the general source of plant DNA from the silty ice. Moreover, the Dye 3 silty ice appears to have originated as solid precipitation without going through stages of superimposed ice and most likely formed by mixing in the absence of free water (i.e., ice that has never melted) (*26*), effectively excluding subsurface transportation. As explained in (*27*), the ice is believed to be predominantly of local origin, having been shielded from participating in the large-scale glacier flow by a bedrock trough, in agreement with the solid ice–mixing hypothesis (*26*). Thus, being of local origin, the DNA sequences from the Dye 3 silty ice must be derived from the plants and animals that inhabited this region the last time that it was ice-free, because possible older DNA records from previous ice-free periods will vanish with the establishment of a new ecosystem, or at least be out-competed during PCR by DNA from the most recent record. The plant taxa suggest that this period had average July temperatures that exceeded 10°C and winter temperatures not colder than –17°C, which are the limits for northern boreal forest and *Taxus*, respectively (*1*). Allowing for full recovery of the isostatic depression that is produced by 2 km of ice, Dye 3 would have been ~1 km above sea level. In combination, these factors suggest that a high-altitude boreal forest at Dye 3 may date back to a period considerably warmer than present.

There are no established methods for dating basal ice, and it remains uncertain whether the overlying clean ice of Dye 3 is temporally contiguous with the lower silty section. Therefore, to obtain a tentative age estimate for the Dye 3 silty ice and its forest community, we applied a series of dating techniques: ^{10}Be/^{36}Cl isotope ratios, single-grain luminescence measurements, amino acid racemization coupled with modeling of the basal ice temperature histories of GRIP and Dye 3, and maximum likelihood estimates for the branch length of the invertebrate COI sequences (*7*). All four dating methods suggest that the Dye 3 silty ice and its forest community predate the Last Interglacial (LIG) [~130 to 116 thousand years ago (ka)] (Fig. 2), which contrasts with the results of recent models suggesting that Dye 3 was ice-free during this period (*28*, *29*). Indeed, all four dating methods give overlapping dates for the silty ice between 450 and 800 ka (Fig. 2), exceeding the current record of long-term DNA survival from Siberian permafrost of 300 to 400 ka (*9*). However, because of the many assumptions and uncertainties connected with the interpretation of the age estimates (*7*), we cannot rule out the possibility of a LIG age for the Dye 3 basal ice.

Our results reveal that ancient biomolecules from basal ice offer a means for environmental reconstruction from ice-covered areas and can yield insights into the climate and the ecology of communities from the distant past. Because many deep ice cores exist from both hemispheres and further drillings are planned, this approach may be used on a larger scale. Basal ice at even lower temperatures than Dye 3 might contain an archive of genetic data of even greater antiquity.

References and Notes

1. O. Bennike, *Medd. Groenl. Geosci.* **23**, 85 (1990).
2. S. Funder *et al.*, *Bull. Geol. Soc. Den.* **48**, 117 (2001).
3. J. E. Francis, R. S. Hill, *Palaios* **11**, 389 (1996).
4. W. Dansgaard *et al.*, *Science* **218**, 1273 (1982).
5. W. Dansgaard *et al.*, *Nature* **364**, 218 (1993).
6. L. Copland, M. Sharp, *J. Glaciol.* **47**, 232 (2001).
7. Materials and methods are available as supporting material on *Science* Online.
8. M. J. Collins, M. Riley, in *Perspectives in Amino Acid and Protein Geochemistry*, G. A. Goodfriend *et al.*, Eds. (Oxford Univ. Press, New York, 2000), p. 142.
9. E. Willerslev *et al.*, *Science* **300**, 791 (2003).
10. M. Hofreiter *et al.*, *Curr. Biol.* **13**, R693 (2003).
11. J. Haile *et al.*, *Mol. Biol. Evol.* **24**, 982 (2007).
12. E. Willerslev *et al.*, *Trends Ecol. Evol.* **19**, 141 (2004).
13. M. B. Hebsgaard *et al.*, *Trends Microbiol.* **13**, 212 (2005).
14. E. Willerslev, A. Cooper, *Proc. R. Soc. London Ser. B* **272**, 3 (2005).
15. M. Hofreiter *et al.*, *Mol. Ecol.* **9**, 1975 (2000).
16. F. Von Wintzingerode *et al.*, *FEMS Microbiol. Rev.* **21**, 213 (1997).
17. J. Binladen *et al.*, *Genetics* **172**, 733 (2006).
18. W. Blake Jr., *Radiocarbon* **31**, 570 (1989).
19. B. A. Bergsma *et al.*, *Arctic* **37**, 49 (1984).
20. O. Bennike, *Geol. Greenland Surv. Bull.* **186**, 29 (2000).
21. O. Bennike *et al.*, *Palaeogeogr. Palaeoclimatol. Palaeoecol.* **186**, 1 (2002).
22. J. C. Bourgeois *et al.*, *J. Geophys. Res.* **106**, 5255 (2001).
23. J. C. Bourgeois, *J. Glaciol.* **36**, 340 (1990).
24. L. Parducci *et al.*, *Mol. Ecol.* **14**, 2873 (2005).
25. H. Shi-yi, *Acta Bot. Sin.* **39**, 363 (1997).
26. R. A. Souchez *et al.*, *Geophys. Res. Lett.* **25**, 1943 (1998).
27. G. H. Gudmundsson, *J. Glaciol.* **43**, 80 (1997).
28. J. T. Overpeck *et al.*, *Science* **311**, 1747 (2006).
29. B. L. Otto-Bliesner *et al.*, *Science* **311**, 1751 (2006).

30. We thank S. Funder, P. Hartvig, J. C. Bourgeois, O. Seberg, J. J. Böcher, K. Høegh, J. W. Leverenz, and S. Y. W. Ho for helpful discussions and R. Bailey, N. Belshaw, N. Charnley, C. Doherty, and D. Peat for technical assistance and advice. E.W., T.B.B., and M.B.H. were supported by the Carlsberg Foundation, Denmark, and NSF. E.W. and K.E.H.P. were both supported by Wellcome Trust Bioarchaeology Fellowships. The Natural Environment Research Council supported K.E.H.P. and M.J.C. E.C. received a Marie Curie Intra European Fellowship (grant number 501340). E.W. and M.C. acknowledge support from the European Union (MEST-CT-2004-007909). M.B. and H.N.P. were supported by the Natural Sciences and Engineering Research Council of Canada (NSERC) (grant number 299103-2004) and McMaster University. M.S. and J.B. were supported by NSERC and the Polar Continental Shelf Project. M.H. was supported by the Max Planck Society. J.B. was supported by the Swiss National Science Foundation.

Supporting Online Material
www.sciencemag.org/cgi/content/full/317/5834/111/DC1
Materials and Methods
Figs. S1 to S8
Tables S1 to S8
References

26 February 2007; accepted 11 May 2007
10.1126/science.1141758

Middle Paleolithic Assemblages from the Indian Subcontinent Before and After the Toba Super-Eruption

Michael Petraglia,[1,2]* Ravi Korisettar,[3] Nicole Boivin,[1] Christopher Clarkson,[4] Peter Ditchfield,[5] Sacha Jones,[1] Jinu Koshy,[3] Marta Mirazón Lahr,[1] Clive Oppenheimer,[6] David Pyle,[7] Richard Roberts,[8] Jean-Luc Schwenninger,[5] Lee Arnold,[8] Kevin White[9]

The Youngest Toba Tuff (YTT) eruption, which occurred in Indonesia 74,000 years ago, is one of Earth's largest known volcanic events. The effect of the YTT eruption on existing populations of humans, and accordingly on the course of human evolution, is debated. Here we associate the YTT with archaeological assemblages at Jwalapuram, in the Jurreru River valley of southern India. Broad continuity of Middle Paleolithic technology across the YTT event suggests that hominins persisted regionally across this major eruptive event.

The Youngest Toba Tuff (YTT) eruption of 74,000 years ago (74 ka) was Earth's largest volcanic event in the past two million years (*1*–*3*). It was two orders of magnitude larger (in erupted mass) than the largest known historic eruption, that of Tambora, also in Indonesia (*4*). The YTT involved the eruption of a minimum of 2800 km^3 (7×10^{15} kg) of magma, of which at least ~800 km^3 was transported in atmospheric ash plumes that blanketed an area from the South China Sea to the Arabian Sea (*2*, *3*). Its impact on Earth's atmosphere and climate (*5*–*7*) and on local animal and plant populations remains a matter of contention (*5*, *7*–*12*).

The Indian subcontinent contains extensive YTT deposits (*13*–*15*). Here we describe an archaeological sequence from south India that includes a substantial YTT layer and sheds light on the eruption's impact on climate, environments, and hominin populations. In the Kurnool District of Andhra Pradesh in southern India, stratified archaeological sites in the Jurreru River valley contain stone artifacts in association with faunal remains in caves, rockshelters, and open-air localities (*16*, *17*) (Fig. 1). The archaeological record spans all periods of the Paleolithic. In addition, current mining activities have exposed tephra deposits over an area of 64 ha. Ash is, however, certainly buried over a wider area within the valley (fig. S1), and we estimate its total volume at $7 \pm 0.7 \times 10^5$ m^3, based on the interpolation of 225 depth observations made at mining exposures.

We conducted electron probe microanalysis (EPMA) of volcanic glass shards from the Jwalapuram tephra to compare their geochemical signatures with those of the Older Toba Tuff (OTT, dated to ~840 ka) and the Middle Toba Tuff (MTT, dated to ~500 ka) (*4*). The results show that the Jwalapuram ash is a distal deposit of the YTT (figs. S3 and S4), based on its close similarities with proximal deposits of YTT in Sumatra and with previously characterized distal occurrences in India (*13*, *14*, *18*).

Jwalapuram locality 3 preserves more than 7.5 m of sedimentary deposits, including a 2.55-m-thick deposit of ash, and a sequence of lithic artifacts that straddle the ash layer (fig. S2). Soft sediment deformation structures suggest that the tephra initially accumulated on a wet clay substrate, probably in a lacustrine environment. The abrupt transition from light gray ash to an orange (but still ash-rich) silt horizon immediately above the ash sequence represents a major change in depositional regime. We interpret this as evidence that the lake dried up soon after the ash fall, possibly during the onset of glacial conditions in oxygen isotope stage 4.

The stone tool assemblages were found in trenches placed across the landscape (that is, at Jwalapuram localities 3, 17, and 21). At Jwalapuram locality 3, we used optical dating to obtain burial ages for sediment samples from archaeological layers above (JLP-380) and below (JLP3A-200) the ash. Ages of 77 ± 6 and 74 ±

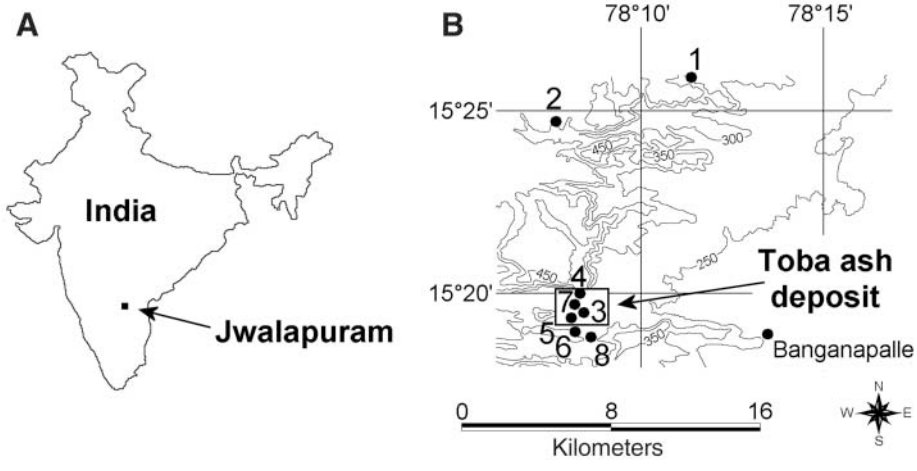

Fig. 1. Location of Jwalapuram, archaeological sites, and tephra deposits. (**A**) Location of the Jwalapuram study area. (**B**) Key archaeological localities in the Kurnool District include the Upper Paleolithic caves of Billasurgum (1) (*17*) and Muchchatla Chintamanu Gavi (2) (*16*). Jwalapuram localities include 17 (3, Middle Paleolithic), 9 (4, Microlithic), 3 (5, Middle Paleolithic), 20 (6, Middle Paleolithic), 21 (7, Middle Paleolithic), and Tank (8, Acheulean).

[1]Leverhulme Centre for Human Evolutionary Studies, University of Cambridge, Cambridge CB2 1QH, UK. [2]Human Origins Program, National Museum of Natural History, Smithsonian Institution, Washington, DC 20560, USA. [3]Department of History and Archaeology, Karnatak University, Dharwad 580 003, India. [4]School of Social Science, University of Queensland, St. Lucia, Queensland 4072, Australia. [5]Research Laboratory for Archaeology and the History of Art, University of Oxford, Oxford OX1 3QY, UK. [6]Department of Geography, University of Cambridge, Downing Place, Cambridge CB2 3EN, UK. [7]Department of Earth Sciences, University of Oxford, Parks Road, Oxford OX1 3PR, UK. [8]GeoQuEST Research Centre, School of Earth and Environmental Sciences, University of Wollongong, Wollongong, New South Wales 2522, Australia. [9]Department of Geography, University of Reading, Whiteknights, Reading RG6 6AB, UK.

*To whom correspondence should be addressed. E-mail: m.petraglia@human-evol.cam.ac.uk