

(SS13) Bridging the Gap between Palynology and Phylogeography: From Ice Core Genetics to Lakebed Sediments

Date: August 29

Place: Room 5336 (oral)

Organizer: Lynn Anderson-Carpenter

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Purpose: The merging of disparate scientific fields can provide the most exciting and challenging aspects of research. The combination can often give new results and solve long-standing questions that neither field could address alone. For example, the combination of ancient DNA and palynology can provide taxonomic identification for morphologically similar species. Differentiating these species can often give a finer-scale resolution of shifting geographic ranges. Additional investigation can elucidate genetic change over time and give insight to implications for organismal response to shifting climates. This research is not without its challenges, and the technical implications must be addressed in parallel with this new data. For example contamination is a constant problem, particularly as DNA concentration tends to decrease with increasing sample age.

The purpose of this session is to bring together ancient DNA with the fields of palynology, paleoecology and phylogenetics. We will address a wide variety of topics including the combination of contemporary genetic patterns with paleoecological data, a DNA analysis of pollen and sediments, ice core genetics, and the combination of paleobotanical models with contemporary genetic data. Such mergers are well suited to tackle a variety of questions that are certain to provide an entertaining and informative discussion.

Oral Presentation

Aug. 29 [AM1] Room: 5336

Chair: Lynn Anderson-Carpenter

[Introduction] SS13-O01

Lynn Anderson-Carpenter

9:00-9:20 **Glacial survival of boreal trees in northern Scandinavia** [SS13-O10](#)

Laura Parducci, [Tina Jørgensen](#), Mari Mette Tollefsrud, Ellen Elverland, Torbjørn Alm, Sonia L. Fontana, K. D. Bennett, James Haile, Irina Matetovici, Yoshihisa Suyama, Mary E. Edwards, Kenneth Andersen, Morten Rasmussen, Sanne Boessenkool, Eric Coissac, Christian Brochmann, Pierre Taberlet, Michael Houmark-Nielsen, Nicolaj Krog Larsen, Ludovic Orlando, M. Thomas P. Gilbert, Kurt H. Kjær, Inger Greve Alsos, Eske Willerslev

9:20-9:40 **Insights from combining pollen based postglacial range expansions with spatial distributions of genetic markers** [SS13-O02 \(154\)](#)

[Thomas Giesecke](#), Simon Brewer

9:40-10:00 **Impact of past environmental changes on the genetic diversity of the remnant natural populations of Cilician fir in Lebanon** [SS13-O03 \(15\)](#)

[Lara Awad](#), Bruno Fady, Carla Khater, Rachid Cheddadi

10:00-10:20 **Integrating fossil data, vegetation modelling and present populations genetic analyses to reconstruct *Larix* history in Europe: first results** [SS13-O04 \(550\)](#)

[Stefanie Wagner](#), Sophie Gerber, Rémy Petit, Thomas Litt

Aug. 29 [AM2] Room: 5336

Chair: Lynn Anderson-Carpenter

10:50-11:10 **Ancient DNA from lake sediments: Bridging the gap between paleoecology and genetics** [SS13-O05 \(9\)](#)

Lynn L Anderson-Carpenter, Jason S McLachlan, Stephen T Jackson, Melanie Kuch, Candice Y Lumibao, Hendrik N Poinar

Chair: Candice Y. Lumibao

11:10-11:30 **Genetic tool for understanding long-term forest changes: analysis of ancient DNA from lakebed sediments** [SS13-O06 \(299\)](#)

Candice Y. Lumibao, Jason McLachlan, Melanie Kuch, Lynn Anderson-Carpenter, Hendrik Poinar

11:30-11:50 **DNA analysis for identification of a *Pinus* pollen grain at subsection level from the Belukha glacier, Altai Mountains, Russia** [SS13-O07 \(363\)](#)

Fumio Nakazawa, Yoshihisa Suyama, Nozomu Takeuchi, Koji Fujita, Satoshi Imura, Hiroshi Kanda, Hideaki Motoyama

11:50-12:10 **A forest-meadow palaeo-ecosystem in northwestern Greenland recovered by ancient DNA of the Camp Century Ice Core** [SS13-O08 \(456\)](#)

Astrid M. Z. Schmidt, Raimund Muscheler, Ann-Marie Berggren, Christine Thiel, Morten Rasmussen, Aurelién Ginolhac, Ludovic Orlando, Sarah Aciego, Jan Pieter Buylaert, Andrew Murray, Jørgen Peder Steffensen, Eske Willerslev

Aug. 29 [PM2] Room: 5336

Chair: Candice Y. Lumibao

14:30-15:10 **[Keynote] A multidisciplinary characterization of a Pleistocene refugium in the interior Pacific Northwest of North America** [SS13-O09 \(148\)](#)

Daniel G. Gavin, Erin M. Herring, Katy Heath, Solomon Dobrowski, Matias Fernandez, Feng Sheng Hu

SS13-O10

Glacial survival of boreal trees in northern Scandinavia

Laura Parducci^{1,2*}, Tina Jørgensen^{2*}, Mari Mette Tollefsrud^{3*}, Ellen Elverland^{4*}, Torbjørn Alm⁴, Sonia L. Fontana^{1,5,6}, K. D. Bennett^{7,8}, James Haile^{2,9}, Irina Matetovici^{1,10}, Yoshihisa Suyama¹¹, Mary E. Edwards¹², Kenneth Andersen², Morten Rasmussen^{2,13}, Sanne Boessenkool¹⁴, Eric Coissac¹⁵, Christian Brochmann¹⁴, Pierre Taberlet¹⁵, Michael Houmark-Nielsen², Nicolaj Krog Larsen¹⁶, Ludovic Orlando², M. Thomas P. Gilbert², Kurt H. Kjær², Inger Greve Alsos⁴, Eske Willerslev²

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*These authors contributed equally to this work.

It is commonly believed that trees were absent in Scandinavia during the last glaciation and first recolonized the Scandinavian Peninsula with the retreat of its ice sheet some 9000 years ago. Here, we show the presence of a new and rare mitochondrial DNA haplotype of spruce that appears unique to Scandinavia and with its highest frequency to the west—an area believed to sustain ice-free refugia during most of the last ice age. We further show the survival of DNA from this haplotype in lake sediments and pollen of Trøndelag in central Norway dating back ~10,300 years and chloroplast DNA of pine and spruce in lake sediments adjacent to the ice-free Andøya refugium in northwestern Norway as early as ~22,000 and 17,700 years ago, respectively. Our findings imply that conifer trees survived in ice-free refugia of Scandinavia during the last glaciation, challenging current views on survival and spread of trees as a response to climate changes.

SS13-O02 (154)

Insights from combining pollen based postglacial range expansions with spatial distributions of genetic markers

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Until recently our knowledge of vegetation history was entirely based on fossil plant remains with associated age information based on direct or indirect dating methods. Combinations of several hundreds of these investigations yield maps of past changes in distribution and abundance. The inferred changes in distribution have been used to estimate rates of postglacial spread of common tree and shrub species. More recently, analysis of genetic markers of living populations has revealed ancestries between populations. Often, the spatial patterns of ancestry contradict the interpretations

based on fossil data, indicating different directions of spread but also that some species must have occurred in regions long before their presence was documented by pollen or macrofossil remains. In consequence, spreading rates deduced from pollen data are potentially erroneous and misleading. However, these fossil records contain valuable information on population size and dynamics. Moreover, where low-abundance spread and survival of small populations is unlikely, minimum estimates of rates of spread may still be obtained from pollen datasets. We present a new set of maps depicting the past distribution and abundance of common trees, based on pollen diagrams collected in the European pollen database. Exploring these maps in several dimensions, we will examine their biases in depicting species distributions, while stressing their usefulness in showing abundance changes in time and space. For selected trees we will compare the maps to published data on the ancestries between populations and discuss the implications for our understanding of postglacial vegetation change. We will evaluate published rates of postglacial tree spread against carefully chosen examples.

Keywords: fossil pollen maps, plant DNA, phylogeography, postglacial range expansion.

SS13-O03 (15)

Impact of past environmental changes on the genetic diversity of the remnant natural populations of Cilician fir in Lebanon

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Abies cilicica (Antoine & Kotschy) Carrière is a conifer species belonging to the high mountain ecosystems of Southern Turkey, Northwestern Syria and Northern Lebanon. In Lebanon, the Cilician fir forests reach their southernmost distribution and are being degraded by a variety of threats from grazing to intensive logging. The aim of this study is to highlight the genetic diversity status of these forests for future conservation strategies. Hence, we have extensively sampled 15 populations and performed genetic analyses using 10 nuclear microsatellite markers. Concomitantly, we are currently analyzing the pollen content of two new fossil records collected in the vicinity of fir populations. The founder effect that promoted the existence of these forests partly dictated the genetic diversity of their populations. A low level of genetic differentiation and no isolation by distance were found between populations suggesting the existence of a significant historical gene flow between populations, therefore, a rather recent fragmentation of these populations is mostly probable. A pollen record spanning the last 30 kyr BP is under study. This will allow us to confirm whether or not *Abies cilicica* was continuously present in the area.

Keywords: fossil pollen, microsatellite, *Abies cilicica*, Eastern Mediterranean, Quaternary.

SS13-O04 (550)

Integrating fossil data, vegetation modelling and present populations genetic analyses to reconstruct *Larix* history in Europe: first results

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Our study aims at elucidating consequences of past climate changes for European *Larix* populations by integrating palaeoecological, genetic data and spatial modelling. Such retrospective approaches can help to evaluate and calibrate models predicting climate change-induced modifications of tree distribution. A limitation of current studies dealing with forest trees is that they do not truly integrate the different types of data, which are usually displayed and analyzed separately. In this study, we use statistical approaches based on coalescent modelling to test hypotheses for the past demography and evolutionary history of tree populations. The coalescent modelling relies on multilocus nuclear microsatellite information of modern DNA, with palaeoecological data and single-locus genetic data (from chloroplast and mitochondria) representing extrinsic information that help frame *a priori* hypotheses. Trees present several advantages for such studies. They have left an abundant fossil record (both macrofossils and pollen) that can be used to reconstruct their past distribution with great precision. Among trees, *Larix decidua* is of special interest as, during several climatic episodes of the last glacial, it played an important role as a dominant constituent within taiga, forest-tundra and forest-steppe communities in the north-central European and Alpine lowlands. Due to its low competitive abilities, it became restricted to the European mountain ranges in the course of the Holocene, when other tree species were starting to recolonize European plains. The planned palaeoecological and molecular genetic analyses will enable detailed inferences on the demography and evolution of European larch populations in a long-term perspective. We will present our first results concerning fossil data compilation in form of chronological maps depicting remarkable range changes under changing climate, their translation into coalescent scenarios, as well as basic population genetic statistics and genetic cluster analysis based on nuclear microsatellite data, the latter uncovering a striking phylogeographic structure. We want to emphasise that such basic analyses are an essential intermediate step before continuing with more complex coalescent models.

Keywords: *Larix decidua*, palaeoecology, modern genetics, phylogeography.

SS13-O05 (9)

Ancient DNA from lake sediments: Bridging the gap between paleoecology and genetics

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Quaternary plant ecology in much of the world has historically relied on morphological identification of macro- and microfossils from sediments of small freshwater lakes. Here, we report new protocols that reliably yield DNA sequence data from Holocene plant macrofossils and bulk lake sediment used to infer ecological change. This will allow changes in census populations,

estimated from fossils and associated sediment, to be directly associated with population genetic changes. We successfully sequenced DNA from 64 samples (out of 126) comprised of bulk sediment and seeds, leaf fragments, bud scales, and samaras extracted from Holocene lake sediments in the western Great Lakes region of North America. Overall, DNA yields were low. However, we were able to reliably amplify samples with as few as 10 copies of a short cpDNA fragment with little detectable PCR inhibition. Our success rate was highest for sediments < 2000 years old, but we were able to successfully amplify DNA from samples up to 4600 years old. DNA sequences matched the taxonomic identity of the macrofossil from which they were extracted 79% of the time. Exceptions suggest that DNA molecules from surrounding nearby sediments may permeate or adhere to macrofossils in sediments. An ability to extract ancient DNA from Holocene sediments potentially allows exciting new insights into the genetic consequences of long-term environmental change. The low DNA copy numbers we found in fossil material and the discovery of multiple sequence variants from single macrofossil extractions highlight the need for careful experimental and laboratory protocols. Further application of these protocols should lead to better understanding of the ecological and evolutionary consequences of environmental change.

Keywords: ancient DNA, paleoecology, macrofossils, bulk sediment, plant.

SS13-O06 (299)

Genetic tool for understanding long-term forest changes: analysis of ancient DNA from lakebed sediments

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Paleoecological reconstructions based on fossil pollen records have given us much insight on how forest community compositions shift over time in response to environmental perturbations. Climate had been shown as the main driver of forest change in the past in North America. Such long-term records are necessary to make better prediction on how forest ecosystems will respond to future climatic and environmental changes. However, the long-distance dispersal of pollen and similarities in pollen morphotypes in some species (e.g. different oak species) can potentially skew or limit the inferences we can make about these long-term forest changes. Here, we present a genetic tool that can potentially address these limitations: chloroplast DNA from trees that are preserved in lake sediments for thousands of years. We extracted, amplified and sequenced chloroplast DNA (cpDNA) from lake sediment core obtained from Little Pond-Royalston in Massachusetts, USA. As chloroplast DNA can only be found in somatic tissue rather than pollen, and that cpDNA sequence is unique to individuals and species, these address both issues. To examine different tree species composition across different time periods, species-specific chloroplast markers were first developed from 16 extant tree species present in Massachusetts to generate species sequence database. These markers were then used to develop a small (~40-bp) cpDNA probes. Preliminary results suggest we can reliably extract, amplify and sequence these probes from sediments up to ~ 6000 year-old and reliably distinguish different tree species. As one of the persistent problems in ancient DNA is low DNA yield, current work involves enriching for the target cpDNA by creating libraries of DNA fragments and subsequently amplifying them. Preliminary analysis indicate this boosted up DNA yield (up to 10 E⁷ copies per uL). These results suggest the potential of ancient DNA analysis for elucidating long-term forest changes. Continued work will allow for reconstruction of shifts in forest

community composition in response to known significant events. For instance, we can examine in finer resolution the impact of the mid-Holocene hemlock decline in the region.

Keywords: paleocology, chloroplast ancient DNA, forest dynamics, Massachusetts.

SS13-O07 (363)

DNA analysis for identification of a *Pinus* pollen grain at subsection level from the Belukha glacier, Altai Mountains, Russia

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Pollen taxon in sediment samples can be identified by analyzing pollen morphology. Identification of related species based on pollen morphology is difficult and is limited primarily to genus or family. Because many pollen grains in mid- and low-latitude glaciers contain protoplasm, genetic information of pollen grains should enable identification of plant taxa below the genus level. Such capability would be extremely useful for reconstructing information on past vegetation, climate, and environments in ice core studies. However, no studies have attempted detailed identification using DNA sequences obtained from pollen found in glaciers. As a preliminary step, the present study attempted to analyze the DNA of *Pinus* pollen grains extracted from surface snow collected from the Belukha glacier in the Altai Mountains of Russia in the summer of 2003. Fragments of 134-147 bp from five loci of the chloroplast genome in each *Pinus* pollen grain were amplified by multiplex polymerase chain reaction, and the DNA products were sequenced in order to identify them at subsection level. As a preliminary result, the success rate for sequence amplification in the present study was 35%. *Pinus* is a taxon with approximately 111 recognized species in two subgenera, four sections and 11 subsections. From the sequences obtained for the six grains, four pollen grains were identified as belonging to subsection *Pinus*. Trees of *Pinus sylvestris*, in subsection *Pinus*, are currently found surrounding the glacier. The consistency of results for this subsection suggested that these pollen grains originated from the same *Pinus* trees found in the surroundings, which spread also as far as Europe. Interestingly, other two grains were identified as subsection *Australes* that is found in North America, Mexico, Central America and Caribbean.

Keywords: glacier, pollen analysis, PCR, ice core.

SS13-O08 (456)

A forest-meadow palaeo-ecosystem in northwestern Greenland recovered by ancient DNA of the Camp Century Ice Core

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Ancient DNA (aDNA) has previously been detected in the basal ice from the Dye 3 deep ice core from southern Greenland and used to reconstruct a rich boreal ecosystem based on a limited number of cloned DNA sequences from plants and insects. Here we present an extended study of deep sequenced ancient DNA amplified from basal ice in the Camp Century ice core from northwestern Greenland. Camp Century was drilled on the margin of the supposedly reduced Greenland Ice Sheet during the last interglacial period. Our results show that the area remained covered by ice during the previous interglacial, the Eemian, 130-116KY. In addition, we find that the area was last free of ice in a period that likely extends further back in time than MIS 11. The aDNA assigns to a relatively diverse biota with boreal to northern temperate taxa, suggesting an ecosystem with forest-meadow landscape with mountains. To be able to provide a climate warm enough to support such an ecosystem we believe this time period coincided with the Dye 3 location and possibly extends back to approximately one million years ago. The biodiversity from Camp Century share plant taxa with both the fossil findings from the Kap København formation in northern Greenland and with taxa from the southern Greenland Dye 3 site, like e.g. *Taxus*.

SS13-O09 (148)

A multidisciplinary characterization of a Pleistocene refugium in the interior Pacific Northwest of North America

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The Pacific Northwest, USA and Canada, supports a mesic conifer-dominated forest that presents an excellent opportunity to synthesize paleoecological and phylogeographic studies that currently show conflicting evidence of glacial refugia. Over 100 plant taxa with a mainly coastal distribution (west of the Cascade and Coast ranges) reappear in the “interior wetbelt” disjunction extending from northern Idaho to central British Columbia. Modern plant distributions are strongly suggestive of a glacial refugium in the Clearwater River system of northern Idaho, within 200 km of the maximum Pleistocene ice extent. The river canyons and mountains of unglaciated areas of the Clearwater support narrowly distributed disjuncts and endemics. In addition, several phylogeographic studies support the long-term existence of mesic refugia. For example, several species show deep genetic divergence among river canyons, suggesting these species existed *in situ* through multiple glacial cycles. In contrast, the paleoecological evidence and paleoclimate simulations suggest the regional climate through the glacial maximum was quite inhospitable to mesophytes. The few pollen records from the broader region show the mesophytic tree species (western hemlock and western redcedar) increased to detectable levels only within the last 4000 years after the onset of neoglacial climate and expanded in a north-to-south pattern, and our preliminary results from a core spanning >40,000 years within the refugium indicate a pine and spruce parkland during the late Pleistocene. Climate

models simulate regional summer temperatures that were 8°C colder during the last glacial maximum. We present an approach for merging three complementary lines of evidence regarding the existence of small glacial refugia: paleoecological records, phylogeographic analysis, and fine-resolution species distribution modeling. A combination of the most robust inferences from each approach will permit detection of the existence of refugia as well as the spatial patterns and ecological context of refugial populations. Merging these three lines of evidence presents new challenges in the synthesis of historical and ecological biogeography, but may eventually reveal the individual histories of species that, in sum, comprise the modern biodiversity in mountainous landscapes.

Keywords: palynology, refugia, phylogeography, microclimate, paleoclimate.