Cultures of dairying: gene-culture-microbiome evolution and the ancient invention of dairy foods



# Cultures of dairying: gene-culturemicrobiome evolution and the ancient invention of dairy foods

## Sprawozdania

Informacje na temat projektu Finansowanie w ramach DAIRYCULTURES **EXCELLENT SCIENCE - European Research** Council (ERC) Identyfikator umowy o grant: 804884 Koszt całkowity Strona internetowa projektu 🗹 € 1 499 988.00 DOI Wkład UE 10.3030/804884 🔼 € 1 499 988,00 Koordynowany przez Projekt został zamknięty MAX-PLANCK-GESELLSCHAFT ZUR FORDERUNG DER Data podpisania przez KE WISSENSCHAFTEN EV 29 Października 2018 Germany Data rozpoczęcia Data zakończenia 1 Listopada 2018 31 Października 2023

### Periodic Reporting for period 4 - DAIRYCULTURES (Cultures of dairying: gene-culture-microbiome evolution and the ancient invention of dairy foods)

Okres sprawozdawczy: 2023-05-01 do 2023-10-31

Podsumowanie kontekstu i ogólnych celów projektu

Dairy products are nutritional resources of global economic importance, and their emergence in prehistory marks a major shift in human dietary ecology. However, basic questions regarding the origins and role of dairying in early human societies remain poorly understood. It is now known that adult hypolactasia (the inability to produce lactase and digest milk sugar) is an ancestral human trait, and that relatively few human populations have genetic variants that allow continued milk digestion into adulthood, a trait known as lactase persistence (LP). The rise of LP has been regarded as a classic example of gene-culture evolution; however, the association between LP and lactose intolerance phenotypes is variable, and LP genotypes do not consistently appear in the archaeological record until more than 5,000 years after the origins of dairying. This has left archaeologists with a puzzling problem, a "milk paradox" regarding how and why ancient peoples developed milk into a dietary resource, how the Bronze Age steppe migrations contributed to the spread of dairying across Eurasia, and what other factors besides LP may have been involved this process. There is now a growing body of evidence that microbes have played important, yet overlooked, roles in the successful establishment of prehistoric dairying economies, and that they may be the key to understanding and overcoming lactose intolerance today.

Prior to this study, there were many outstanding problems regarding our understanding of animal milkbased subsistence in traditional dairying societies like Mongolia. For example, little was known about the earliest phases of dairying in Mongolia, now how Mongolian populations changed through time as nomadic pastoralism became the dominant lifeway on the Eurasian steppe. The dispersal and spread dairy domesticated livestock and the time depth of dairying was also poorly understood, and the methods for the detection of milk proteins from dental calculus and archaeological food containers were underdeveloped, making the identification of archaeological dairying challenging. Finally, relatively little was known about the relationship between the gut microbiome and dairy digestion in traditional dairying societies.

The DAIRYCULTURES project set out to address these problems and to answer fundamental questions about the prehistory of dairying by focusing on Mongolia, a country where the rural diet is primarily made up of dairy products, and where dairying has been practiced since prehistory. The project applied new and emergent genomics and proteomics techniques to: (1) reconstruct the population history of Mongolia from the Eneolithic period to the present and to examine the dynamics of genetic lactase persistence in Mongolian populations over the past 6,000 years; (2) reconstruct the origins and dispersal of domesticated dairy livestock in Asia, with a focus on the establishment of dairy pastoralism in Mongolia; (3) improve methods for detecting milk proteins in archaeological dental calculus and reconstruct the development and spread of dairy technologies into Mongolia; (4) ethnographically document the lives of Mongolian dairy pastoralists today and characterize the dairy products they produce; (5) explore the relationship between dairy consumption, lactase persistence, lactose intolerance, and gut microbiome composition and function in dairy-consuming societies by conducting a metagenomics study of the gut microbiomes of nomadic pastoralists and urban populations in Mongolia today.

Prace wykonane od początku projektu do końca okresu sprawozdawczego oraz najważniejsze dotychczasowe rezultaty Our research began with obtaining permissions for archaeological, human, and animal studies, aligning with Nagoya Protocol principles. Exchanges, workshops, and conferences, including major events in Central Asia and Portugal, facilitated dissemination. Over 50 presentations, a documentary, and collaborations with museums led to public lectures, a museum exhibit, and full Mongolian translations of key articles. The 2023 AAA's Exemplary Cross-Fields Award recognized our work on human-microbial relationships in the DAIRYCULTURES project.

Bench protocols for ancient DNA and proteins were published. Genetic and proteomic data were deposited in ENA and PRIDE. Ethnographic materials were shared online, complemented by a companion article in Current Anthropology.

We reconstructed 214 ancient Mongolian genomes, unveiling population dynamics over 6,000 years. LP alleles' Bronze Age introduction in Mongolia and their non-selection were highlighted. Additionally, 18 ancient genomes from the Tarim Basin revealed a Bronze Age dairy pastoralist population lacking genetic lactase persistence.

Dental calculus proteomes analysis identified the Afanasievo as the earliest dairy pastoralists in Mongolia at 3000 BCE. Dairying evolution revealed sheep focus initially, expanding to include goats and cattle, and later horse and camel milking in subsequent periods. Afanasievo migration to the Dzungarian basin influenced local adoption of cattle milking. The genetic origin of Afanasievo in the North Caucasus steppe led to a high-resolution reconstruction of the spread of dairying from the Near East/Anatolia through the Eurasian steppe to Mongolia.

Bronze Age livestock and dairy technology's cultural exchange spread further than human migrations alone. Genetic screening of 1,065 livestock bones across the Eurasian steppe highlighted the importance of cultural exchange in early dairying. Prescreening via ZooMS methods improved genetic sequence efforts. Modern livestock genomes in Mongolia were sequenced, surpassing initial plans in both sample size and sequencing effort.

Microbiome sampling, LP testing, and breath hydrogen tests offered insights. Distinct microbial communities in nomadic pastoralist and urban gut microbiomes were observed. Nomadic pastoralists maintained high gut microbiome diversity, Prevotella abundance, Bifidobacteria variability, and elevated levels of dairy-associated lactic acid bacteria. De novo metagenomic assembly identified novel bacterial species, shaping ongoing peer-reviewed publications.

Pandemic-related disruptions redirected funds to livestock genomic sequencing, delaying microbiome data completion. Despite challenges, this project yielded 28 publications, 59 conference presentations, 2 conferences, 4 workshops, 1 museum exhibit, 1 documentary film, and diverse outreach resources. The extensive impact demonstrates the project's significance in anthropology and genetic research.

### Innowacyjność oraz oczekiwany potencjalny wpływ (w tym dotychczasowe znaczenie społeczno-gospodarcze i szersze implikacje społeczne projektu)

During the project, we took advantage of major technological advances in the field, and this allowed us to push the boundaries of the science. Our study of ancient human genomes in Mongolia was the largest and most ambitious such study in Asia at the time of its publication. After refining and optimizing ancient protein extraction, and we recovered the largest number of milk protein PSMs of any ancient proteomics study to date. For our gut microbiome study, we added a de novo metagenomic assembly component, a method that was not yet available at the time of our initial proposal. And for all aspects of the study, we were able to investigate more samples and generate more genetic sequencing data than initially planned because we took advantage of new technologies and efficiencies that emerged during the course of the study.



Traditional yak milking in northern Mongolia

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