

## Genetic secrets from 4,000-year-old teeth illuminate the impact of changing human diets over the centuries

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Killuragh Cave, Ireland. Credit: Sam Moore, Owner Marion Dowd.

Researchers at Trinity College Dublin have recovered remarkably preserved microbiomes from two teeth dating back 4,000 years, found in



an Irish limestone cave. Genetic analyses of these microbiomes reveal major changes in the oral microenvironment from the Bronze Age to today. The teeth both belonged to the same male individual and also provided a snapshot of his oral health.

The study, carried out in collaboration with archaeologists from the Atlantic Technological University and University of Edinburgh, is <u>published</u> in *Molecular Biology and Evolution*. The authors identified several bacteria linked to gum disease and provided the first high-quality ancient genome of Streptococcus mutans, the major culprit behind <u>tooth decay</u>.

While S. mutans is very common in modern mouths, it is exceptionally rare in the ancient genomic record. One reason for this may be the acid-producing nature of the species. This acid decays the tooth, but also destroys DNA and stops plaque from fossilizing. While most ancient oral microbiomes are retrieved from fossilized plaque, this study targeted the tooth directly.

Another reason for the scarcity of S. mutans in ancient mouths may be the lack of favorable habitats for this sugar-loving species. An uptick of dental cavities is seen in the <u>archaeological record</u> after the adoption of cereal agriculture thousands of years ago, but a far more dramatic increase has occurred only in the past few hundred years, during which sugary foods were introduced to the masses.

The sampled teeth were part of a larger skeletal assemblage excavated from Killuragh Cave, County Limerick, by the late Peter Woodman of University College Cork. While other teeth in the cave showed advanced dental decay, no cavities were visible on the sampled teeth. However, one tooth produced an unprecedented amount of S. mutans DNA, a sign of an extreme imbalance in the oral microbial community.



"We were very surprised to see such a large abundance of S. mutans in this 4,000-year-old tooth," said Dr. Lara Cassidy, an assistant professor in Trinity's School of Genetics and Microbiology, and senior author of the study. "It is a remarkably rare find and suggests this man was at a high risk of developing cavities right before his death."

The researchers also found that other streptococcal species were virtually absent from the tooth. This indicates that the natural balance of the oral biofilm had been upset—mutans had outcompeted the other streptococci, leading to the pre-disease state.



An example of a tooth prior to ancient DNA sampling. Note this was not the tooth sampled in the study. Credit: Dr. Lara Cassidy, Trinity College Dublin



The team also found evidence to support the "disappearing microbiome" hypothesis, which proposes that modern microbiomes are less diverse than those of our ancestors. This is cause for concern, as biodiversity loss can impact human health. The two Bronze Age teeth produced highly divergent strains of Tannerella forsythia, a bacteria implicated in gum disease.

"These strains from a single ancient mouth were more genetically different from one another than any pair of modern strains in our dataset, despite the modern samples deriving from Europe, Japan and the U.S.," explained Iseult Jackson, a Ph.D. candidate at Trinity, and first author of the study. "This represents a major loss in diversity and one that we need to understand better."

Very few full genomes from oral bacteria have been recovered from prior to the Medieval era. By characterizing prehistoric diversity, the authors were able to reveal dramatic changes in the oral microenvironment that have happened since.

Dr. Cassidy added, "Over the last 750 years, a single lineage of T. forsythia has become dominant worldwide. This is the telltale sign of natural selection, where one strain rises rapidly in frequency due to some genetic advantage it holds over the others. T. forsythia strains from the industrial era onwards contain many new genes that help the bacteria colonize the mouth and cause disease.

"S. mutans has also undergone recent lineage expansions and changes in gene content related to pathogenicity. These coincide with humanity's mass consumption of sugar, although we did find that modern S. mutans populations have remained more diverse, with deep splits in the S. mutans <u>evolutionary tree</u> pre-dating the Killuragh genome."

The scientists believe this is driven by differences in the evolutionary



mechanisms that shape genome diversity in these species.

"S. mutans is very adept at swapping <u>genetic material</u> between strains," said Dr. Cassidy. "This means an advantageous innovation can be spread across S. mutans lineages like a new piece of tech. This ability to easily share innovations may explain why this species retains many diverse lineages without one becoming dominant and replacing all the others."

In effect, both these disease-causing bacteria have changed dramatically from the Bronze Age to today, but it appears that very recent cultural transitions in the industrial era have had an inordinate impact.

**More information:** Iseult Jackson et al, Ancient genomes from Bronze Age remains reveal deep diversity and recent adaptive episodes for human oral pathobionts, *Molecular Biology and Evolution* (2024). DOI: <u>10.1093/molbev/msae017</u>, <u>academic.oup.com/mbe/article-1</u>... .1093/molbev/msae017

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