Prehistoric human remains from South-America help to uncover the origins of treponemal diseases

One of the biggest mysteries in historical epidemics is whether syphilis was introduced in Europe by Columbus’ first voyage to the Americas. A new international study, led by the University of Basel, reconstructed four *Treponema pallidum* genomes from nearly 2000-year-old remains from Brazil, that were most closely related to bejel-causing bacteria. These findings support the existence of treponemal infections in pre-Columbian times with treponemal diseases already present in the New World at least 1000 years before Columbus set sail.

Treponematoses are infectious diseases caused by the *Treponema pallidum* bacteria. Endemic treponematoses, yaws and bejel, are transmitted mainly via skin contact, in contrast to syphilis, which is generally known as a sexually transmitted disease. Recently, all treponemal diseases are re-emerging and have shown signs of antibiotic resistance, causing concern and considerable public-health burden. The endemic treponemes are currently relatively rare outside developing countries, while syphilis continues to infect globally.

**Bejel on the ancient American continent**

The new international study by researchers from Switzerland (University of Basel, University of Zurich, ETH Zurich), Austria (NHM Vienna, University of Vienna), Spain (University of Valencia) and Brazil (University of Sao Paulo) is the first to reconstruct and analyse genomes of *T. pallidum* bacteria from an ancient American context, namely the coastal region of Santa Catarina in Brazil. Altogether four genomes were recovered from nearly 2000-year-old human remains found buried in a shell mound. All four genomes belong to the bejel-causing strain, *T. pallidum endemicum*, and show such close similarity to modern strains, that this subspecies seems to have survived time nearly unaltered. Today, bejel is most commonly found in the hot and arid regions of the world, whereas Jabuticabeira II, the ancient burial site where the samples in this study were excavated, is situated in a tropical coastal location.

The existence of a treponemal disease in prehistoric South America suggests that the bacterial family had already spread globally in pre-contact times, or emerged on the American continent. “However, the fact that the findings represent an endemic type of treponeme leaves the origin of the sexually transmitted syphilis still unsettled”, the last author of the study, Prof. Verena Schuenemann from the University of Basel reminds.

**Insights into the archaeological sites on coastal Brazil**

The bones recovered from archaeological sites on the coast of Brazil were first analysed in systematic studies by Dr. José Filippini, who states that „there is morphological evidence of treponematoses in many of the 45 sites I analysed“. These early coastal populations of Brazil constructed mountain-like shell mounds, or Sambaquis. His colleague at University of Sao Paulo, Dr. Luis Pezo-Lanfranco, who also studies the diet and oral health of these Sambaqui populations, reminds us that „sambaquis are made of shells, sand, and black soil, in which these sedentary fisher-hunter-gatherers also buried their dead in complex rituals. Stone mortars, monkey tooth necklaces, whole fish, fire pits, and sometimes stone animal statues were given to them as burial offerings“. The Sambaqui culture survived for about 8000 years, until about 1000 years ago, making it one of the longest-lasting cultures ever. „The necessary contact between people to maintain the Sambaqui culture across thousands of square kilometres and thousands of years could be one of the explanations for the spread of contagious infectious diseases, such as the skin treponematosis bejel“, argues Prof. Sabine Eggers from the Natural History Museum in Vienna who led the anthropological investigations of Jabuticabeira II.

 **Recombination the key to the treponemal evolution?**

Many bacterial species acquire useful new traits through horizontal gene transfer or recombination. In treponemes, several recombination events involving both modern and ancient genomes were detected requiring already distinct subspecies co-infecting the same host at some point in the past. “Although we cannot pinpoint the exact timing of these events, our analyses show that the recombining genes affect the structure of the treponemes’ phylogenetic tree, and suggest that recombination might be one of the driving mechanisms in the divergence between the subspecies that cause different treponemal infections”, says Dr. Marta Pla-Díaz, one of the first authors of the study and a postdoctoral researcher at the University of Basel. She developed methods to analyze evolutionary processes such as selection and horizontal gene flow in ancient and modern bacterial genomes during her PhD thesis at the University of Valencia, which were included in this study. Prof. González-Candelas, mentor of Dr. Pla-Díaz and coauthor of this study, said that „including ancient genomes is essential to understand which evolutionary processes and factors acted in the past and, in the case of *Treponema pallidum*, how, when, and hopefully where, they brought a new pathogen to cause such a serious pandemic as that caused by syphilis over the past five centuries.”

**Archaeogenetic aides to old enigmas**

The distinction of bacterial species causing historical epidemics has previously relied mainly on osteological evidence visible on skeletal materials. In the modern era, archaeogenetic methods can be used for solving some persistent enigmas surrounding these diseases. “In this study, we surprisingly found treponemal DNA in both clearly infected specimens, and outwardly clean-looking bone samples, demonstrating the importance of genetic evidence in confirming paleopathological diagnoses” says Dr. Kerttu Majander, postdoctoral researcher at the University of Basel, and one of the first authors of the study. With the recent advances in ancient DNA methodology, it has not only been proven possible to reconstruct ancient treponemal genomes from archaeological remains, but to pinpoint the agent in individual disease cases down to the subspecies level. Unexpected findings, like recovering a prehistoric agent of bejel in a coastal context on the American continent, showcase the potential of ancient DNA beyond inferences drawn from modern pathogen genomes, or archaeological interpretations alone.

With the help of molecular clock dating, the evolution of a species can be illuminated by even a single high-quality genome: the best performing specimen in this study pushed the divergence of the whole *T. pallidum* family back in time to about 12,000-550 BCE, and in the future, older genomes might still appear with a similar effect. With each improved estimate we are one step closer to the goal of uncovering the origins of this age-old foe. “The emergence of syphilis might still leave space for the imagination, but at least we now know beyond doubt that treponemal infections were no strangers to the American inhabitants who lived and died centuries before the continent was explored by Europeans,” concludes Prof. Verena Schuenemann.

Further analyses on both ancient and modern treponemal genomes around the globe will be necessary to gain a comprehensive understanding of the history and interaction among the different members of the *T. pallidum* family and their potential. A focus on locations where bejel and syphilis coexist is expected to provide valuable insights into the dynamics of recombination and the adaptability of this enduring human bacterial pathogen that has yet to be eradicated.