Modern lab reaches across the ages to resolve plague DNA debate

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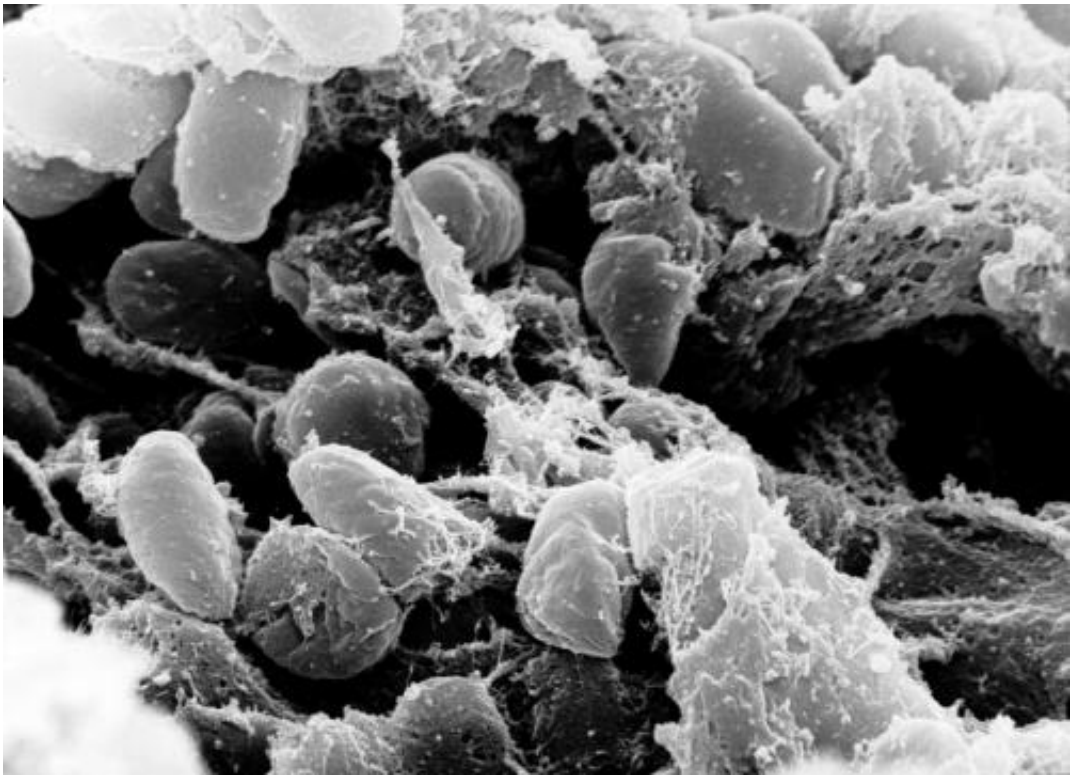

An electron micrograph depicting a mass of Yersinia pestis bacteria (the cause of bubonic plague) in the foregut of the flea vector. Photo courtesy of the National Institutes of Health.

(Phys.org) —From within an ancient German gravesite to laboratories under the harshest extremes of scientific scrutiny, traces of DNA from a deadly disease illuminate the cold pages of history with modern insight.

How a plague from 1,500 years ago—long a reputed ancestor to the notorious Black Death—found its way back to relevance is a story of high science intersecting with a distinctly human touch, and parts of it played out at the Microbial Genetics and Genomics laboratory at Northern Arizona University.

"To be able to go back and answer one of the great unanswered questions in history is extremely satisfying," said MGGen Associate Director Dave Wagner, who was co-author of a study published in the prestigious journal *PLOS Pathogens*. "It's some of the most fun I've had in academics."

The project's outcome resolves what had been an ongoing academic debate: The Justinianic Plague that started in A.D. 541 can indeed be placed on the family tree of the disease, linking it to the well-known pandemic of the 14th–17th centuries, and finally to a third pandemic that began around 1850.

The implications resonate both with history and with civilization today. Not only is the first pandemic tied to the other two, but it almost certainly began in Asia—not in Africa, as previously thought. And any deeper understanding of the disease (known technically as Yersinia pestis), which has potential as a bioterror weapon, makes our world a little safer.

Wagner recounts the two-year-long sequence of events with the ease of a narrator who played a pivotal role in the outcome. His decade of experience working with plague activity in northern Arizona, initially among prairie dogs, helped establish the reputation of MGGen as a world-class center of expertise on the topic.

That led to an ongoing collaboration between Wagner and colleagues at the Bundeswehr Institute of Microbiology in Munich, who, together with

individuals from State Collection for Anthropology and Palaeoanatomy in Munich, led the overall study. Much of their joint work is funded by the U.S. Department of Homeland Security, which Wagner said is interested in techniques that could be used following a bioterrorism event to identify the agent and determine its origins.

Paul Keim, director of MGGen and a co-author, said "international plague collaborations are increasingly difficult due to government regulations restricting material exchanges. This work succeeded through team perseverance and the electronic exchange of data."

After the discovery of an ancient burial site near Munich, Wagner and MGGen were brought in by the Germans as experts on plague DNA genotyping. The age of the remains presented an opportunity to answer the historical debate by extracting some of the teeth, grinding them up and searching for traces of Y. pestis DNA.

But there the story adds a layer of complexity.

"We realized that for the results to be accepted by the scientific community, we couldn't do the work in this laboratory," Wagner said.

Ironically, MGGen's status and reputation in plague DNA research meant it would not withstand the scrutiny of experts in ancient DNA, a field that demands its own set of protocols and protections against contamination by modern DNA. The highly critical community had rejected an earlier study about the Justinianic Plague because, in part, it did not meet their extraordinary standards.

Yet in the end, MGGen did contribute because two research associates under Wagner's guidance combined to form a team that blended thoughtful creativity with meticulous technical skills.

Dawn Birdsell and Katy Parise rose to the challenge of working with ancient DNA, and while playing their part in a centuries-long chronicle of science and history, they made a little of their own.

To advise the German scientists by generating their own results, the pair needed to rethink MGGen's procedures and capabilities to address two daunting obstacles: the fragility of the ancient DNA samples and the uncompromising protocols of analyzing them.

During long nights of contemplation, Birdsell, with a Ph.D. in physiology and a noted talent for genotyping, imagined each step of an elaborate, systematic approach to assaying ancient DNA in a lab designed for the modern stuff. She knew that even the most minor details would face severe skepticism.

"These samples are so ancient that they're in the range where our technology has certain limitations," Birdsell said. She redesigned the lab's protocol, then engineered a process to assure the process was working properly while avoiding the risk of contamination. The resulting strategy was her own, and had never been published.

"This is a profession that really is 50 percent creativity and 50 percent linear and logical," Birdsell said, which is precisely why she recruited Parise, who was finishing her MS in biology at NAU while working on the project. "It was my observation that she was a meticulous, careful technician," Birdsell said.

As Parise put it, "I think it just makes sense to do the absolute best you can the first time, so I take my time, I think about things and I don't rush. We didn't have the time to make any mistakes and the samples were way too precious."

Ultimately, after Parise's efforts to confirm all the protocols through lab

experiments, Birdsell handled the ancient samples.

"I was always very clear that I would process the final sample because I wanted the responsibility on me," Birdsell said. "It also motivated me. You just execute and stick with the planning and don't deviate."

When the analysis turned up ancient plague DNA, the two knew their work had paid off. When their collaborators' two labs in Germany—labs specifically designed for the work—confirmed the results, "we were pretty thrilled," Birdsell said. "If anything, it's a story of what a successful team can produce."

Provided by Northern Arizona University