



DR ARTUR ROŻAŃSKI

SECRETS HIDDEN IN TEETH

To retain our cultural identity in the modern world and sensibly think about the future, we need to thoroughly study the past," says **Prof. Marek Figlerowicz** from the PAS Institute of Bioorganic Chemistry, who leads the project "The Dynasty and Society of Piast-Era Poland in the Light of Integrated Historical, Anthropological, and Genomic Research."



Human remains resting in the subterranean vaults of the Gothic basilica forming part of the Cistercian monastery in Lubiąż. Research to date has indicated that the basilica was the burial location of the Silesian Piasts, including Bolesław I the Tall.

ACADEMIA: Do the new techniques for isolating, sequencing, and analyzing ancient DNA, or aDNA, offer a way to glean new historical knowledge?

MAREK FIGLEROWICZ: We applied for funding from Poland's National Science Centre for such a project as a direct consequence of the extraordinary progress that has recently been made in the field of DNA sequencing. Working out the first full human genome sequence, published in 2003, took 10 years of intensive study. Now it takes just a few days to do the same thing, and the cost is relatively low: with current hardware, from one to two thousand dollars per genome. New techniques have also been developed to make it possible to isolate DNA not only from modern humans, but also from ancient animal, human, and plant remains dating back thousands of years. The Neanderthal genome has already been sequenced, and we have sequenced the genomes of Neolithic people. These two technological advancements have enabled us to think in real terms about using biological methods in historical or archaeological research. Discovering history by studying the organisms that were then alive.

You are a precursor of this method in Poland.

One could say that about the team we have put together: the Poznań Center for Archaeogenomics. We want to

study not small fragments of genomes, for instance by genotyping just mitochondrial DNA or Y chromosomes, but rather whole genomes. To sequence the longest possible portions thereof, so as to draw conclusions on that basis.

However, studies of this kind are controversial.

On the one hand, historians and archaeologists have up to now considered themselves experts on the past. On the other hand, we have anthropologists studying human remains. Based on bone structure and simple genetic tests, they can also say something about history. Our genomic approach is quite simply completely new. But when we talk about history, we have to remember that in genomic research we are mainly referring to biological history. Our studies do not contribute decisive new information to cultural history. For instance, we can imagine that a person of fully Germanic descent moved as a child into areas inhabited by Slavs and was raised there in the Slavic spirit, and so culturally he would have considered himself a representative of the Slavic tribes. But when we look at his remains from the genetic standpoint, we will say: this is a genome typical for a certain period in the history of Germanic tribes. For this same reason, such research will only make sense if it is interdisciplinary. We have to consider

the history that is inscribed in genomes in conjunction with our historical and archaeological knowledge. Only such a combination can give us sensible answers to numerous questions that are now arising.

At this stage of the project, can you already say something about the origins of the population inhabiting the region between the Oder and Vistula Rivers in the times when the first Polish state, ruled by the Piast dynasty, was forming?

We have certain hypotheses, we have obtained preliminary data, but I would not like to talk about it because it still requires very careful verification. I can talk about how our research will proceed and what assumptions we started with. To begin with, note that we have very limited knowledge about what happened in the territories now called Poland back in the first millennium. There are many hypotheses in this regard, including two main ones.

The first maintains that at the start of the first millennium, there were mainly Germanic tribes living to the West of the Vistula. That changed radically around the fifth century: the Germans withdrew to the west and Slavic tribes moved in, occupying territories all the way to the Labe, beyond Berlin. The border we currently know was formed at the end of the first millennium, in other words when the Piast state emerged. The second hypothesis is that the territories were actually inhabited by the same population the whole time, only ruled by different rulers. There were political divisions, but not biological ones. To try to verify those theories, we decided to collect DNA from people living between the Oder and Vistula at the start of the first millennium, in other words in Roman times, and compare it to DNA from people living here around the year 1000, around when the Piast state was emerging. However, the issue is far from simple, as there is no such thing as a Slavic or Germanic gene.

We will be scrutinizing the genetic diversity within the given population – people from the start of the millennium, people from the end of the millennium, and both groups combined. And if the variability remains on the same level, that means that the same population lived here both at the beginning of the first millennium and at the end. If it is diametrically different, we will have clear biological evidence that a one set of people lived here earlier, then moved away to be replaced by another. We already have around 100 genomes sequenced from the Roman period, and now we are trying to obtain the same from the Piast era. We already have the first results, but they are very rudimentary.

Inactively I would lean towards the first hypothesis. And I am more convinced about the second. Here we need to consider how tribal population migrations worked.

When people became crowded in a certain portion of Europe or Asia Minor in ancient times, a small group would head off to look for new territory, taking their civilization with them. If it was more developed than the local civilization they encountered in the new lands, it began to take predominance over the latter. Genetically, however, the predominant share of the people in this territory will remain local, rather than being of outside origin. I would suspect, given that Europe in those days was mainly populated in small settlements scattered across the forests (especially in our Polish lands), the local population remained the same the whole time. Only the “administrators” changed.

So you hypothesize that the elite in our lands came from elsewhere, bringing civilization with them?

There are at least two possibilities. It may have been the case that a very strong group came in, founded a dynasty, and then built up an elite from the local population, ultimately a state. Or that people came in from elsewhere, bringing new ideas, but they were not strong enough to seize power. They therefore began to advise the local authorities and became the elite, whereas the dynasty remained local. Of course, it may have been the case that both the dynasty and the elite were local.

A strong group moves in and rules – that sounds like an occupation.

All the signs are that is precisely how Russia emerged. The Vikings sailed down the rivers from the north, and Russia’s first dynasty, the Rurikids, were of Viking origin, as genetic testing has confirmed.

Rurik was a Viking?

Yes. And another issue – people often think that we can answer complex questions by isolating and studying a single genome. But if such an analysis is to be done responsibly, it must be done in a broad context. Within a given area, we need to study the so-called non-elite and the elite, and also the ruling dynasty. Only by comparing all three groups can we say something more. Moreover, the results of my research should be combined with similar studies being done around us, in Germany, Russia, the Czech Republic. We recently visited the latter. There is a hypothesis that the foundations of the Piast state were laid by fugitives from the Great Moravian state.

You mean from Nitra?

Yes. Even though Great Moravia adopted Christianity, like the Piast state later did, so as to become part of the broader European community and thereby gain protection, it did not manage to survive and was conquered. One of the hypotheses is that fugitives from Nitra migrated north, where they created a new state. Unfortunately, there is no way to verify that hypothesis at present, because there are practically no documents describing the emergence of the Piast state.

GENOMICS AND THE EARLY POLISH STATE

Apart from just the few lines in the *Dagome iudex*.

Yes, but that document did not survive to our times. The oldest known record of it, a summary, dates from the end of the tenth century. And so we do not even have any document we could refer to directly. This is a question of civilization. If we look at the expansion of the Roman state, we see that we had the misfortune of having it pass us by. The Romans halted in the Carpathians. Their influence extended into parts of today's Ukraine, but not into Poland. Roman documents are a source of historical records. When we talk about our project to scholars from Western Europe, they are very surprised because they have shelves full of books on the analogous topic. We have practically nothing. Only the chronicles of Wincenty Kadłubek and Gallus Anonymus. But as far as the early Piast state is concerned, the chronicles written after Gallus Anonymus are generally fantasies written so that we Poles could feel better about ourselves, among other nations.



JAKUB OSTALOWSKI

Why were the Romans uninterested in us?

An answer to that is provided by modern times. Look at how refugees are moving from south to north. They avoid Poland not just because we are not as economically developed as Germany, but also for geographical reasons, due to the lay of the land. People tend to move along rivers, because they have to have access to water, and avoid crossing mountains. The same pathways that are in use today were used a thousand, two thousand, and five thousand years ago.

Can genomic research determine the origins of the Piasts?

Of the groups we want to study, the dynasty is the hardest. Not many remains of the first Piasts have been preserved. In Poznań there is a chapel that supposedly holds the grave of Mieszko I and Bolesław I the Brave. Unfortunately, there is nothing in it. Several hundred years ago, there was a person in Poland greatly interested in preserving the remains of well-known people. He received consent from the bishop of Poznań to take part of the skull of Bolesław the Brave from that grave and put it in his collection. Soon after a fire in the cathedral, the tower of the chapel collapsed. Some bones were found afterwards, but it is not clear whose. They were put into a single box. The collector gave the skull fragment to the Czartoryski family, and it is today at the Czartoryski Museum in Kraków. However, we have no proof that this is an authentic fragment of Bolesław I's skull.

We have researched how many known Piast burial places there are in Poland, so as to select the ones that are most certain and take samples from them to study. More than 500 such locations have been described, more than 300 of them in detail. But when we began visiting them, the sarcophaguses usually turned out to be empty. Now we know that there are about 20 places containing nearly certain Piast remains. We are

considering the main Piast line, as well as the Silesia and Mazovia lines.

To identify the origins of the dynasty, we have to study the Y chromosomes. The pace of mutation is so slow that if a male-line descendant of Mieszko I is alive today, he should have an identical Y chromosome. We will have to check whether the Y chromosome is the same in all the Piasts we take samples from. It will be awful if it the chromosomes turn out to be different. This would mean that the technique will not teach us anything about the origin of the Piasts, and it will attest to the infidelity of their wives. If the chromosome is the same in many samples, we will obtain a benchmark to use in evaluating the uncertain remains. Perhaps then we will manage to re-confirm some of the remains.

Have you already managed to study the hard-to-access remains of Władysław I the Elbow-High and Kazimierz the Great, resting in the crypt of Wawel Castle in Kraków?

Not yet, but I believe that we will. In fact, all the Piast remains should be hard to access. Those that are easily accessible will have been taken out, mixed with other remains, or moved elsewhere. That is a widespread phenomenon. In France, during the French Revolution, the remains of kings were thrown into the Seine. And so, the idea has arisen to gather together several groups of European researchers and together try to reconstruct the history of the important families, because science now gives us such a chance. We should not waste it.

What kind of remains are most useful for DNA research?

DNA stands the chance of surviving the longest in the best condition in bones of the highest density – i.e. parts

Prof. Marek Figlerowicz

is director of the PAS Institute of Bioorganic Chemistry. He studies the molecular mechanisms involved in processes generating genetic variability and the involvement of RNA in regulating gene expression.

Marek Figlerowicz
@ibch.poznan.pl

of the skull and tibia bone. But the teeth are the most certain, in view of how they are constructed. They are well enervated inside, covered outside in enamel and well protected. They need to be cleansed of bacteria, fungi, and other microorganisms. We can drill into them, and DNA can be extracted from the powder so obtained.

Can the confirmed Piast remains be used to infer something about their lifestyle or type of diet?

There is such a possibility, but not by means of DNA research, as genes do not predestine us for any particular diet. At present, isotope research techniques are being intensively developed. Chemical elements frequently occur in as several different isotopes in nature, variants differing in terms of the number of neutrons, and therefore atomic mass. The relative ratio of certain isotopes varies from place to place on the surface of the Earth. One isotope frequently used in this way is strontium. Researchers have developed maps showing how different strontium isotopes are distributed across various regions of Europe. Teeth can be studied using mass spectroscopy to identify the quantities of specific isotopes and therefore ascertain where the given individual must be from. But this only applies to the period between the appearance of adult teeth and the 19th year of life. Teeth, like tree trunks, have rings. If we study the isotope composition of the individual layers, we can trace where the person spent their time in Europe. Isotope analysis can also tell us whether a person had more of a meat-based or vegetable-based diet.

We have to consider the history that is inscribed in genomes in conjunction with our historical and archaeological knowledge.
Only such a combination of information can yield sensible answers.

The bishop of Płock initially did not allow the Piast graves under the cathedral there to be opened. But you did ultimately manage to access them?

When we contacted the bishop, it turned out that someone else had previously asked for consent to open the sarcophaguses. More and more people want to carry out analogous studies. But generally only individual ones, which do not make any greater sense. We need to consider establishing a body to consider this issue in the ethical sense. We have proposed for a commission to be set up as part of the project, tasked with working out certain operating principles. That commission would evaluate whether a particular study could contribute enough that it justifies disturbing the remains.

The plans call for the project to compare the genomes of 800 samples. How many have you managed to collect so far?

Ancient DNA (aDNA), usually from a sample isolated from a tooth, constitutes less than 5%. But that is a very good result. The remainder of the DNA is from fungi, bacteria, and other organisms growing in the studied bone. We planned to collect 800 samples and use the sequencing method to carry out a preliminary screening. Today these methods are so efficient that one sequencing reaction can provide 400 million readings – short DNA sequences. This enables the DNA samples to be pooled. First we mark them with a special code, kind of like a barcode. We can mix them together and in one sequencing reaction analyze around 200 samples. Then for each of them we will have several million readings. On this basis we can identify the quantity of aDNA in the sample. Of the 800 samples, we will choose only those that contain the proper quantity of aDNA of the necessary quality. And so the 800 samples will be reduced to around 200. We will have 50 samples for each of four groups: the Roman period, Piast non-elite, Piast elite, and Piast dynasty. We can already see that in the case of the latter group we will not manage to acquire fifty 100%-certain samples. But perhaps we will manage to find several of them, and on that basis establish a benchmark Piast Y chromosome. The project is meant to last five years. So far we already have more or less as many sequenced samples as we need for the Roman period, around 50, and they are quite decent. From the Piast era we have around 100. We would like them to have both good DNA and good historical-archaeological description. In parallel we want to find out what kind of bacteria these people lived with. Aside from soil bacteria, we are finding that teeth contain traces of the DNA of bacteria typical of the oral cavity. Recently we found the DNA of the whooping cough bacteria from a thousand years ago. On this basis we can conclude many interesting things, such as the incidence of a given disease in a given territory.

Why is such precise study of the past so important?

I think that we have a big problem with our history in Poland. Because it is so poorly known, our understanding of the world is not fully appropriate. We concoct various legends about our own history. The chronicler Wincenty Kadłubek, for instance, reports us as deriving from Noah's sons. The novelist Henryk Sienkiewicz wrote historical tales to raise people's spirits, but we often take them true. But in order to retain our cultural identity in the modern world and sensibly think about the future, we need to thoroughly study the past.

INTERVIEW BY ANNA KILIAN

This is an English translation of an article approved by the author only in its Polish version.