Ancient DNA

[MUSIC PLAYING]

CORY: Hi, everyone. I'm Cory with WSU Global Connections. And welcome.

I'm proud to introduce Dr. Brian Kemp. He's an assistant professor in molecular anthropology. He received his PhD from the University of California, Davis and has been teaching at WSU since 2007. His area of research includes ancient DNA and New World archaeology, specifically addressing the peopling of the Americas and the rates of molecular evolution. We are very excited to have him with us here today to share a bit of his research and area of expertise.

BRIAN KEMP: OK, thank you, Cory. And hi to everybody who logged in, there. This is a new experience for me. Let's see how it goes. If you have questions, I believe you're supposed to text them in. And then, [? Sarah ?] can let me know when questions come in. And I will respond to them.

OK. So what I'm going to talk to you today about is some of my research and some research of other people, in which we're using ancient genetic data to address anthropological questions-- and hence, the title, "Anthropological Applications of Ancient DNA." And what I would like you to get out of this lecture over the next 50 minutes or so, is first, what is ancient DNA? And importantly, how is ancient DNA of great value? What can we learn from ancient DNA that we can't from any other source of information?

Two, what are some good sources of ancient DNA? From what can we extract and analyze ancient DNA? And then, third, I'm going to list a whole number of applications of ancient DNA, things that people have done-- some of which I have done, here at WSU. And we'll look at a couple of these. I've listed far more applications than I can get through. But we'll see what I can do in time we have left.

So what is ancient DNA? For this, I like to turn to this individual, here. I've got to be careful not to go too fast because this screen's taking a while to load up. This individual here at the top, that's Svante Paabo. And it was work by Svante Paabo in the late 1980s that led to this intellectual revolution-- what if we could extract and analyze DNA from things that are no longer living?

Svante Paabo is who I consider-- and most consider-- to be the godfather of the field. And interestingly enough, in 2007 Time magazine considered him to be one of the 100 most influential people in the world. And this is largely because of his efforts to reconstruct the Neanderthal genome, which we have a draft of. And it's because of his lab.

According to Svante Paabo-- and this comes from a review paper that he wrote-- he defined ancient DNA "broadly as the retrieval DNA sequences from museum specimens, archaeological finds, fossil remains, and other unusual sources of DNA." So you can see, it includes quite a few different sources. Now, when he says "fossil remains," that's a bit of a misnomer. So, if something is completely fossilized, we can't get DNA out of it. Sometimes we say we can't make stones bleed. There is no organic left in it. But the quote is nevertheless fairly appropriate.

Now the way to I like to think about ancient DNA, and really the importance of ancient DNA, we can look at this map here. And it doesn't really matter precisely what it's depicting here. But it's some mitochondrial DNA types that are distributed in populations-- indigenous populations-- across the American Southwest. And these are some data I collected when I was working on my PhD back in California.

Now, we know that the variation exists today. And the way I like to think about it, is to be able to take this map and see how this works. But we'll take this map. And in a way, we're turning it on its side. And then, we're thinking about a map of the same area that existed sometime in the past. And I put a question mark here.

So where we can find human remains in this case, and type them for, say, mitochondrial DNA, we can say, what were the patterns exhibited by populations in these same areas 1,000 years ago, or 2,000, or 5,000, or 10,000, from wherever we have good radiocarbon dates. And if these patterns are different, we can ask, how different are they? And what explains that difference? And it's really evolution. And so, we can see evolution in action this way.

And another way I like to think about it is this. So let's pretend that some time in the past, some woman carried a mitochondrial DNA type, whatever color this is. It's sort of bluish-gray. And mitochondrial DNA is passed only from mothers to offspring. And so we look at the second generation. And we follow the mitochondrial transmission through all these generations. And we see that first woman-- the woman at the top-- gave birth to two females. She had two daughters that carry her mitochondrial DNA type.

Where we see on the right side, we see a mutation accumulate. So blue turns into red. It doesn't mean a person is red. It just means it's a different type. And you can see, a blue color evolves from that. And then we can see a green color on the left. And then we can see, for instance, a yellow color in the middle.

And we notice something here, in that that yellow color didn't make it to the present. So the bottom line is what we would call modern DNA. We can sample from people living today. But that yellow type, well, it either went extinct, or that female-- the last female that carried the yellow type-- didn't have any daughters. She may have only had sons.

And so, extinction of lineages is actually an important part of a population history. But we can't see it from the present. So we can't see extinctions in the present. We have to look in the past.

What ancient DNA allows us to do is sample from any one of these generations. And we can actually see things go extinct, such as that yellow lineage. And we get a better estimation of what happened to that population over time. So I think that's where the real power lies.

And according to me, this is how I would put it. I'd say, "Analyzing ancient DNA allows one to directly test hypotheses that are based on theory from 'modern' DNA studies through the ability to precisely date the age of remains that exhibit genetic types of interest." So what we can do, is we can find, for instance, human remains. And we know where that person died. And we can use radiocarbon dating to say when they died. And then, we can sequence them in their DNA and tell what their genotype was. So we know where they died, when they died, and something about their genes.

And it's really hard to get that much information from a single sample. The other kind of question that I would want to know is, what did the person speak? But we can't know that, right. We've gained a lot of information, nevertheless-- where they died, when they died, and something about the genotype.

Now, what are some sources of any ancient DNA? From what can we extract and analyze DNA? These pictures are depicting good sources of ancient DNA-- so bones and teeth. We can also get DNA out of mummified tissue, although we know today that both bones and teeth are superior in their preservation of ancient genetic material.

Another source of ancient DNA-- and this comes from a recent study by a student, actually, out of the University of Chile, she studied DNA that was contained within what is called "dental calculus." And you're seeing a picture here. So, what dental calculus is, is just a fancy name for tartar. And you can see how much tartar is deposited on the teeth. So what her and her colleagues did, was scrape off some of that tartar, and then extract DNA from it. And they can tell, not only thinking about the human DNA, but about the bacteria that were living in the mouth of that person or those people.

Another excellent source of ancient genetic material is from what archaeologists would call "coprolites." And what this is, is just a fancy name for poop. And what we're seeing here is a number of feces or poop samples they have yielded good ancient DNA.

On the top left, we see those poops are about 6,000 to 9,000 years old, from a cave in Utah. To the right of that is a early farmer poop from the Southwest. It's about 2,000 years old.

To the right of that, are some poops that I studied from a cave. They're human, from a cave in Southern California. To the right of that, those are some ancient turkey droppings that are about 2,000 years old.

And all of these samples-- the one from the bottom, as well-- have yielded good, preserved DNA. So we can tell something about people and the animals from their feces. And we don't even need necessarily their bones and teeth.

So what I'm depicting here are artifacts that we call "quids." Quids are, in prehistoric times, in places like the American Southwest, Southern California, and parts of Mesoamerica, people would chew on yucca. We're not exactly sure why they were doing this. But a lot of people did this. And so, when they would shoot on yucca and spit up fibers, when we find them thousands of years later in rock shelters, like the ones that shown here, they just looks like wadded up pieces of fiber. But some of these have yielded DNA, presumably from cheek cells that get incorporated while the person was chewing on those.

So in this picture, what I'm depicting is a bag of what archaeologists call "midden." Midden is just really another fancy name for garbage. You can see that it is an exceptionally rich bag of garbage. This comes from a site in southeast Utah called Turkey Pen Ruins.

And you probably can recognize a number of things in this picture, such as those corn cobs and those corn stalks. You can see a little piece of deer hide there. There's a quid in the middle, if you can see that-- that chewed up yucca. And then, all those big, gray spots that you're seeing, those are all human feces. And those are some of the ones I showed you in the couple pictures ago.

And why we get such good preservation? I also, on this computer, I can see turkey feathers-- if you can see that in the middle as well. And there's lots of human hair in there too, which you can't see, but I have seen in there. Because of the preservation in the southwest, so all of this was found in a midden that was protected by a very large rock shelter. So it protected these artifacts from the elements. So they haven't been wet since they basically were deposited there.

So all the things you're seeing here are 2,000 years old, approximately. And all of these things have the potential of yielding DNA. So the questions are almost limitless. What do you want to study? We studied the turkeys from this site. We studied the humans. We could study the maize. We could study the animals that they caught and ate. There's a lot of information here if you know how to get to it.

So the gentlemen here that I'm depicting on the left, this is Eske Willerslev. And he's one of the leading ancient DNA researchers in the world today. And he and his team have been pushing the idea of studying dirt DNA-- or sometimes we call it "sedi-DNA" or "sediment DNA"-- and also, DNA that's held in ice cores.

And he's holding there some kind of a machine in which they will go up to see the permafrost, which is more or less frozen soil. And they just core right into the permafrost, and take the columns of dirt back to the lab, and then cross section it by stratigraphic layer, and then sequence all the DNA that's in the soil. And the plants and animals that were alive when that soil was being made contribute to that soil.

So parts of their DNA get incorporated in the soil. And you can reconstruct what kind of plants and animals were there at the time when that soil was produced. And some of the DNA from the permafrost samples date as early as over 400,000 years.

And on the right here, you see a depiction from one of his papers that's the study of ice cores-- DNA from ice cores. And the DNA from that is in excess of about 700,000 years. So these are the examples of the oldest DNA that has been recovered from things.

Now, before we jump into some of the applications here, I like to put this slide as a warning, just to say, I think I'm going to show you some flashy results and things that I think are really cool. But you have to understand that this doesn't come without a lot of attention to detail. And it takes a lot of time and a lot of effort to get the data. And I don't have time in this lecture here to go into this specific problem. But just appreciate that this is not easy. If it was, everybody would be doing it.

There's all kinds of different applications. And we'll look at a few of them during this lecture. One is dietary reconstruction. We could study the genetics of extinct populations and/or species to determine their evolutionary relationships-- not only to living populations, but also to other, extinct populations or extinct species.

We could study ancient diseases, for example, Black Plague. There's a number of diseases that will actually incorporate their DNA into bones, and sometimes, for instance, we can recover DNA from Yersinia pestis, which is supposed to have caused Black Plague, and study the evolution of diseases that we lived with for thousands and thousands of years.

We could test for population continuity versus replacement. Archaeologists have long been interested in tracking migrations of people around the world, and anthropologists have, as well. And archaeologists can look in the ground and say, well, at this time, some time, these people were doing something. And then, some time later, they were doing something very different.

And the question is, were those different people later? Or did those people evolve into a people and just adopt, say, a different life way. Well, we can test this directly by testing the biology or the genetics of these different populations and pose the question, did one evolve from the other? Or can we say that that isn't possible?

Molecular sex determination-- sometimes is very hard to tell the sex of a skeleton. If the skeletons are in very good shape, and you're looking at adults it's easier. But when the skeletons are fragmentary or they're children, it's very difficult to tell the sex. We can do that with genetics.

Domestication of plants and animals-- because ancient DNA gives us-- we have time on our hands, we can actually see the process in effect. So domestication is not an event. It just doesn't happen. It's a process that occurs over many, many, many generations, in which humans are controlling the breeding and the biology of the plants and animals. And we can actually see that in real time with ancient DNA studies.

And lastly, but it's really not the last application-- it's the last one I'm going to list here. But, we can measure the rate of molecular evolution by looking at radiocarbon dating samples and seeing how one type evolves into another. We can literally watch evolution occur.

So I'm going to start here with dietary reconstruction. So there's this really interesting paper that was published by Hendrik Poinar in 2001. This was published in the Proceedings of National Academy of Sciences, or PNAS. And him and his team in this study had recovered some human feces that you see depicted here-- three human fecal samples from a cave that was located in Hinds Cave Texas. These date to in excess of 2,000 years old.

And so, not only were they able to determine that these were made by humans, they checked the mitochondrial DNA. And it came out with types that are known to be exhibited by Native Americans, which makes perfect sense, because that's who lived in Texas 2,000 years ago. Well, they took it one step forward. And they wanted to ask the question, what do these people eat? Was there dietary DNA left in these fecal samples?

So the way that he did this, is they thought about every plant undergoes photosynthesis. And many of these genes are coded for in the chloroplast DNA. One in particular, which is called rbcL, all plants have this gene. And different species, or different genera of plants have slightly different sequences. And so what they did, is they used a method called polymerase chain reaction to make many, many copies of the few remaining copies that are in these feces. And then I'll what they did to that second.

And then, they wanted to also look at that animal component. So they looked at some mitochondrial DNA and the gene here, called the 12S RNA. So they PCR amplify that, as well.

Now, because it's very likely that the humans ate more than one type of plant or one type of animal before the feces were made, they have to tease this out. And they use a method called cloning here. They take an amplified DNA. And they have bacteria pick them up. Bacteria love to pick up pieces of DNA.

And then, you can grow up those bacteria in plates. And each of the little colonies would start from a single bacterium. And then, you can sequence the DNA from that colony. And it ultimately originated from a single sequence. And that must have come from one plant or one animal that that human had eaten.

And then, basically, you'd see a bunch of diversity of sequences. Here I'm just making up some sequences. But you would basically sequence the DNA, see the pattern of As, Gs, Cs, and Ts and ask from what kind of creatures could this DNA have originated?

Now, they also use a more traditional approach in a macroscopic look at these feces. So this is more traditional, in which you would just take the fecal material and start breaking it apart. And you'd look for things in there, such as seeds or parts of plants that you could recognize as being distinct in one species or another. And sometimes you find little bones in there, too. And you might able to identify to what animal that belonged.

So here's the results in the first sample. So the feces that was made by that human 2,000 years ago included pronghorn antelope, cottontail rabbit, packrat, squirrel, something in the hackberry, sunflower, yucca or agave, and cactus. Sample II, we see also of a large diversity of things. In many cases, they couldn't get down to the species level. But you're seeing here something in the sunflower family, something in the nightshade family. What exact species it is, we don't know. But you can see that this person also ate quite a variety of things before defecating in the cave 2,000 years ago. And then number III, we can see evidence, for instance, of sheep, packrat, something in the buckthorn family, et cetera.

So what's really neat here is that there is some theory-- archaeologists like to use some theory called "Optimal Foraging Theory," in which you make predictions about what humans should have eaten, given what was available. And here, I just want to highlight again, the power of ancient DNA. These are direct observations. We don't have to guess what the people were eating or say they should have been eating something. This is what they ate, what? A day or two before defecating, however long it takes for all that to get through the system. A really unique approach to understanding what people ate in the past.

And what this indicates, I think you'll all agree, is that it indicates that the inhabitants of this cave had a very diverse and well-balanced diet. And it seems like they were eating everything that they could get their hands on. Wasn't one particular thing or a few things. They had a very rich, well-balanced diet.

Now, what's neat here, is because they took both a macroscopic approach and a molecular approach-- that is, a genetic approach-- macroscopically, three plants were confirmed. So three plants that they saw, with the DNA were confirmed. They could see it, whereas six were not.

And conversely, they could only see cactus macroscopically. They couldn't detect cactus DNA. They could see something that looked like cactus in there.

So by using both approaches, we get a better idea of what these people are eating. We learn something from both methods. And when we combine them, we learn even more.

Now-- I like this part because it's kind of funny to say-- but macroscopically, no large mammals were detected, whereas small mammal bones and teeth were detected in the feces. Now, oftentimes you will find little rodent bones or little fish bones in ancient feces. But you don't see deer bones because people don't swallow deer bones. OK, they're just too big. And so, what this study shows you is really if you want to study the diet of humans through their feces, and you want to know what they were eating-- at least the large mammals-- you have to use DNA. Because humans just don't swallow large bones, OK.

So back to the applications, I think we'll move on to a case study that I was involved in, looking at population continuity versus replacement-- testing if a migration occurred. What I'm depicting in this picture is the distribution of what was called the Numic languages. And people who speak Numic languages are found distributed throughout the Great Basin, which is the area I'm depicting here.

And most linguists believe that these people originated somewhere in Southern California, such that when we're depicting the Northern Paiute's closest linguistic relative is the Mono, south of them. And the Panamint is most closely related to Shoshone. And Kawaiisu is most closely related to Ute. And this fan-like pattern has been interpreted as, these people began somewhere in Southern California. And at some point in the past, some people speculate around 2,000 years ago or so, spread out into the Great Basin.

Now, there were people in the Great Basin before the hypothesized migration. And these are people we call "pre-Numics." Though some archaeologists also see evidence for this migration in terms of people moving different kinds of technology through time and space. But the real question is, how do the pre-Numic people relate to the Numic people today? And with ancient DNA, we can directly evaluate this.

And when I was in graduate school, and I had read these papers on ancient feces, I really had a hard time believing it. Because I thought, first, it's probably the coolest thing I've ever heard. And secondly, I thought, just how could this be possible? It must be difficult or something.

Well, at that time I met a woman named Wendy Nelson who had studied macroscopically some human feces from the cave that I'm depicting here, Fish Slough Cave. And you can probably barely see the opening of the cave is that little, tiny black hole there. And in this cave, the cave's full of feces. And you can see some human feces depicted below that.

And when I met her, I told her about my interest in trying out a genetic study of these coprolites. And she gave me eight samples. And every single sample yielded human mitochondrial DNA. And so it wasn't as hard as I thought it was going to be. And it really spurred me to think about the usefulness of just feces in general for reconstructing population histories and relations in areas like Southern California and the Great Basin, et cetera.

And these samples all date to about 700 to 2,000 years. The cave's a mess, really. The stratigraphy is all turned around. So these poops weren't directly dated. But things in the cave were directly dated to about 700 to 2,000 years.

What I'm depicting in this next slide are some mitochondrial DNA frequencies of people living in the Great Basin today that are Numic speakers-- the Northern Pauite Shoshone. And that's what their pattern of mitochondrial DNA looks like. The Washo to the left of them, they speak a language that is not related to other languages around. It's a very unique language.

On the right there, you see Fremont. Those are some samples you can see that date from 650 to about 600 years before present. Those were ancient farmers that took farming way to the north of Utah, where nobody else could farm. It's pretty amazing. And that Ancient Western Great Basin, those are pre-Numic peoples, studied from a collection of bone in the Western Great Basin.

And then you can see at the bottom, the eight samples and what I discovered from Fish Slough Cave. Now, a sample size of eight is pretty small. So I wouldn't put much weight on the frequencies that I'm depicting here.

But I'm going to tell you that, for instance, that yellow color-- Haplogroup C-- there's not just in one form of Haplogroup C. And if we get more and more sequences from these things, we can say specifically is it one type of C or another, what variant of C it is. And that's when it got very interesting-- even more interesting.

So when I sequenced the DNA more from these coprolite samples, the Cs were all of a type that are very, very rare. And in fact, they've only been observed in the Northern Pauite today, and the Washo, and a few other-- a Northern Pauite some Pima-- Pima is a population in the Southwest-- and another population in Southern California.

So the circle I'm showing you here are the relationships between the types I saw in the poops and their relationships to the direct matches to people living today and their relationships to other people living today. It's an extraordinarily rare type that has only been observed in Southern California and the Great Basin in the American Southwest.

And so, what does this mean? means-- I'm going to put these little asterisks there. It means that some relatives or ancestors of the Northern Paiute Shoshone and the Washo pooped in that cave in Southern California about 2,000 years ago.

And that might sound funny. I think it does sound a little bit funny. But just start to imagine the potential here. How else would you have known that their ancestors or relatives were down in Southern California 2,000 years ago? And we're fortunate for this great preservation of feces in these caves.

And this is just the beginning. We can start to look at movements of people, well, in areas where you can find feces or bones, too. And we can start to reconstruct migrations like this.

So the Northern Paiute Shoshone are thought to have originated around where Fish Slough Cave is, about 2,000 years ago. Now, I'm not going to say that this proves anything. But it certainly is consistent with the hypothesis. And a lot more work would have to go into really testing this. But I think that it's an interesting start.

OK, back to applications. And we're going to move on to this issue of molecular sex discrimination. And this was a study I also got involved in. And this is a paper. We published this paper a couple of years ago, in 2008. This was work that I did with some of my colleagues that worked down in Mexico City.

And they contacted me because they had these Aztec sacrificial victims. And they were trying to determine the sex of these individuals. And they contacted me when I was a grad student. Said, could i help them out? And I did. And we put out this paper that I think is very interesting.

So the Aztecs had founded their empire at a site called Tenochtitlan. That was the capitol. And neighboring Tenochtitlan was a trade city, Tlatelolcol. It was a market city or a trade city.

And we see depicted here what's called Temple R, from that site. And before that temple was built, a number of individuals were killed-- a number of kids, mostly kids-- and sub-adults. What I mean by "sub-adult" is not an adult, so juveniles or teens. And a lot of kids were killed, placed in the ground, and then that temple was built, OK. And that temple was dedicated to this god, here. This is the god of wind and rain.

I'll show you some examples of the types of remains that were unearthed from under that temple. The first one there is a six-year-old. You can see that was buried directly in the ground. And the one on the right here is estimated to be about two years old. And that individual was buried in one of these ceramic urns. They're called "ollas," OK.

Now, my colleagues wanted to know what was the sex profile of these sacrificial victims. And as I mentioned earlier, the skeletons of children are very difficult the sex. And it's mostly because they haven't developed their secondary sexual characteristics. If you think about it-- little boys and little girls-- if you put them in gender neutral clothes and maybe give them a similar haircut-- little boys and little girls, even when they're alive are hard to tell apart. And their bones are even more difficult to tell apart.

Now, one question you might have-- and I think it's an interesting one-- is about how do we know how old these kids were when they died? Obviously, they were small. So those were kids. But some of these sub-adults, I mean, how do we know they're not adults?

Well, people who understand this far better than I do-- one of the tools they use is the eruption of the dentition. And I'm showing here patterns of a dental eruption of people that are aged at different ages. And some of you out there might have your wisdom teeth already in. If you have your wisdom teeth in, and we find your mouth many thousands of years later, we will know you're an adult. If your wisdom teeth have not erupted, then we don't consider you to be an adult, no matter how you feel about yourself.

But I think you get the point. This is one way that we can tell approximately how old somebody is. It's the same thing that forensics people would use, too. And so, in this box here, that's the real difference between adults and sub-adults, whether they have their wisdom teeth or not.

Now, I was talking about sex determination from the skeleton earlier. Now, if you're provided with a complete skeleton-- a skeleton that's very good shape-- and you have the pelvis-- and I'm depicting here a male pelvis on the top, a female pelvis on the bottom. Men and women have very different-shaped pelvises. And it has everything to do with birthing babies. Men don't birth babies. Women do. And so the shapes are quite different. So if you had a complete pelvis like this, you could probably tell the difference.

Now if we look at the head, depicted on the right there, women tend to be more gracile. Men tend to be more robust. And I just mean, the bones are a little bit thicker. They're a little heavier-looking. Men tend to have a bit more of a brow ridge. Those are some things that can clue you in on whether somebody is a male or female.

But kids just don't have these differences. So we really want to know something about the kids. Well, as a molecular anthropologist, as a geneticist, I know the difference between men and women. And that is at the genetic level.

So men having an X chromosome and a Y chromosome. Females have 2 X chromosomes. So if we can look at those chromosomes and look at the DNA from those chromosomes in these ancient remains, we could potentially tell the sex. If we see a Y chromosome in there, it has to be male.

And fortunately, there's a gene called "amelogenin," that I've just called "amel" here, in which all humans, the X chromosome, the gene is six base pairs-- precisely six base pairs-- shorter than the Y. And so, when we PCR amplify that piece of DNA-- and let me show you. There is a female. And there's a male. And we amplify it. And we'll run them on a gel here.

We're going to put DNA in those little holes there and run the gel. We'll run the gel for a while. And then, we'll have two bands. And females will have one band. And they're precisely six base pairs different, with the X chromosome in this experiment is 106 base pairs. And the form on the Y chromosome is 112. It's pretty straightforward.

But I was thinking, well, I did detect some males in there. I have to be able to tell the difference between me and them. So we'll get to that point. Oh, wait. It is the next one.

So I have a Y chromosome because I'm a male. So if I identify any of these kids as little boys, I could have potentially contributed to that result. It could be contamination.

And so what I did, was I screened all of the ones I determined to be males for two mutations that are commonly found among Native American males. But I know I don't have them. So if I see these mutations from the ancient sample, that can't be me. I used that as a safeguard.

We also wanted to see was I getting the right signature from the mitochondrial DNA. So here was the profile of the 14 individuals I was able to test. And you can see a lot of Haplogroup A, that red color. And we can compare that to a study I published three years earlier of Aztecs from the same city.

These weren't sacrificial victims. But those profiles are very, very similar and suggest that the data are authentic. They make sense. If they didn't look too similar, I'd be very suspicious. But they looked very similar.

And of the sex termination-- my colleagues at UNAM in Mexico City, they said, well, there were 26 of 26 they were able to determine were male. My study was of the 10 that I was able to sex type, I thought that 9 were male, and 1 was female. Now, in this case, we might contrast. Or I might be wrong. They might be wrong about one of those individuals. It doesn't really matter, because overall the vast majority of these sacrificial victims were little boys, right.

And what does this mean? That's a non-random pattern. If they were randomly killing people, you should get about a 50/50 mix. Here there's intention. There's planning. OK, we don't know exactly why they were doing it. But it's a non-random pattern. There was intention here. They intended to kill little boys.

Now, why? As an anthropologist, sometimes we have to speculate about these things. And in that paper, if you care to take a look at it, we speculated a little bit. So the wind and rain god has all of these little boy deities that help him out. And our best guess is maybe they were trying to impersonate these little deities with these little boys.

But it's a total guess. We don't know, OK. Maybe somebody else has a better idea. I mean, there's only certain things that DNA can tell us. It can't tell everything.

OK, back to the applications here. And this is good timing. I think I have just enough time just to finish looking at this section of domestication of plants and animals. And I'm going to talk to you today about our work with turkey domestication. It's something that we're actively pursuing a WSU. We're trying to learn more and more about initial turkey domestication in the Americas.

So you may or may not know this, but there are very few animals domesticated in the Americas. The turkey is one. Dogs were brought to the Americas from Asia. So there's no independent domestication there. So we have domestic turkeys in South America. We have domestic camelids, like llamas and alpacas. They domesticated Guinea pigs, muscovy ducks.

But there were very, very few animal domesticates in the Americas, compared to the Old World where we had pigs, cattle, donkeys, camels, all the classic ones. So the impact of turkey domestication for people who achieved this in the American Southwest can't be underestimated. And we didn't really know much about it-- the genetics of it-- until very recently.

What I'm depicting here is-- there are two species of turkeys. And the ones that I'm showing you here, located in the United States and in Northern Mexico and Central Mexico, those are all one species. It's Meleagris gallopavo. And the different colors I'm depicting here are different subspecies. So they can totally interbreed with one another, but they have slight morphological differences, where an expert can say, well, that's one subspecies or another.

Now in Central Mexico, there is today a subspecies that is either extinct or extraordinarily rare. I cannot actually determine this because different reports tell me different things. They haven't been seen in a while. And some people think they're extinct. And presumably, this would have been the turkey that the Aztecs had domesticated. And it's called subspecies gallopavo. There's also a subspecies called the Tularosa bird, or subspecies Tularosa, found in the Southwest that is thought to be extinct today.

And that light blue color in the Maya region, the Yucatan there, in Mexico, that's a totally different species of turkey. It's called the ocellated turkey. And you can see from this picture, it looks to me more like a peacock. But it's really a turkey-- different species, though. And it'll be shocking one day to know if those were the domesticated. But there's no evidence of that yet.

So what I'm showing you here are four views of the same turkey. This is the type specimen of the Tularosa bird that's suspected to be a different species, or subspecies, of turkey. But nobody's seen it today. So we think it's extinct.

This is the type specimen held at the Smithsonian. And it was found in 1905 in a cave called Tularosa Cave and then scientifically described for the first time in the 1960s, early '60s. And what differentiates this from other known turkeys, other subspecies of turkeys, is the fully-feathered neck that you can see here. And you can see, if you look at the talons-- this fiery red talons-- it's actually, I mean, it's a very beautiful-looking bird. And what's cool here, is that we actually see the phenotype.

How old is this bird? We'd have to directly radiocarbon date it. But it's probably at least 1,000 years old.

So it was found in a cave. And some other interesting things about it-- when they cut open the crop, it was full of corn. So corn just poured out of the crop.

And they also believe it may have died of dehydration. Doesn't explain the mummification. But they think it was a turkey that was left in there and may have just died of dehydration. It's a really amazing specimen.

So aside from that extinct species, or subspecies, we decided to do a study of turkey variation, the samples that we could find, from all over the Southwest. And this is work that I competed here with a number of co-authors you see here, and some people we worked with from Simon Fraser University. And what we were really trying to do with this paper-- we did a few things. But the main thing we were trying to address is, were turkeys first domesticated in Central Mexico and moved up to the Southwest? So a lot of archaeologists believe this is how turkeys got to the Southwest. And we wanted to test that.

So what we did, was we identified a site. And what I'm depicting here is the Four Corners region. So you can probably see that. But we found an archaeological site, or knew about an archaeological site in Cedar Mesa there, in Southeast Utah, right there, around there. And this site is called Turkey Pen Ruins.

Now, this is the same site from which that midden had come, where all those ancient feces had come and all that cool garbage. And what you see here, is on the right, you see a pueblo. So that's an old building, likely at least 1,000 years old or so. And that guy there in this picture, he's standing on top of a midden. It's a garbage pile, basically.

And if you look on the left-- just to give you an idea of scale about this rock shelter-- you look on the left, and you see that guy. That's that guy. So you can see how protected this site is from the elements with that big rock overhang. And that's what causes the preservation here to be so great, because nothing gets wet.

Why do we call it "Turkey Pen Ruins"? Well, at least in pueblo times, they had turkey pens. And when this loads up, you'll see the turkey pens. We find these all over the Southwest. Usually it will be an enclosure of a number of sticks, like I'm showing in this case. Or sometimes they were subterranean, where there might be a little pit.

In any case, that would have been a place where they would have had a bunch of turkeys. And where there's a bunch of turkeys, there's a lot of turkey poop. And that's what we were focused on to study here. OK, that's all loaded up. So you can see that turkey pen there.

This was built in pueblo times. Well, we were interested in earlier times. So we had to dig a little deeper.

The people before the puebloans, are what we called "basket-maker peoples." Why we call them "basket-maker peoples" is that-- and you'll see in this picture-- they made incredible baskets. OK, this is pre-pottery. They hadn't figured out pottery yet. Puebloan people had pottery in which they could cook. These people made these really exquisite baskets. You could cook in them, but not by placing them-- basically you would heat up rocks, throw them in some water in these water-tight baskets, and then cook some corn in there, make some kind of gruel, OK.

What you're seeing here is a picture of a guy. And this was taken in 1891. So people have known about this site for a very long time. And what that guy's doing is looting the site. I mean, he's basically stealing stuff from there. And he's going to go sell it.

And here's another picture from that same excavation of looting. These guys, these cowboys here-- you can see them with their shovels-- they're digging through midden. That's all garbage. It's about 1 and 1/2 meters deep of garbage-- looking for cool stuff that can be sold, right.

And what's incredible to me is, the sample midden we have in our museum here, when you open that bag up, the first thing you smell is mouse urine. It's disgusting. And what's crazy about this picture to me is, these guys are out there with no ventilators on, digging through feces-rich soil and urine-soaked soil, looking for pots. And it would have been very, very dangerous and pretty disgusting.

I'm going to actually skip this slide. This is a picture of the midden that I had already shown you where all the human feces were. There's a lot of turkey droppings in there. And that's what we were focused on.

So in-- was it 1971 or 1972? RG Matson went and put a controlled column through that midden and bagged it up by stratigraphic layers. You can see the profile here. This is what they drew. And brought that back to WSU.

We dated two turkey droppings directly. So one dates at about 1,000 years old. These aren't corrected. They are radiocarbon years. In real years, it's a little in excess of that number.

But that is a turkey that lived there in pueblo times that pooped. And the poop made it in that pile. And at the bottom, that's 1,900 radiocarbon years. When that's corrected, that's over 2,000 years.

And as I'm going to argue here, we believe that was made by a domestic turkey. And if that's true, that's the oldest evidence of turkeys in the Americas. It's certainly older than this. We did not find first domestic turkey poop ever to exist. But this is the earliest known to exist.

And I get asked this question when I give this lecture at different universities. And I give this lecture also to some of my students here, on campus. Some people want to know, well, how do I know that what I'm looking at-- which, I'm showing you three examples-- how do I know that these fecal samples, or these poops, are from turkeys?

Well, they look like bird poop. That's for sure. They're a large bird's poop. And there's no other birds around there that could make that. And when we extract DNA from them and screen them for turkey mitochondrial DNA, virtually every single one of them has turkey DNA in them. So we believe strongly that they are turkeys.

Another thing that tells us about turkeys-- and maybe some of you know about this, but I learned about this during the process-- is that male and female turkeys make different-shaped poops. And so, I'm trying to get picture here when it loads up. So on the top right there, hens typically do these plops, what we call plops. And gobblers, or the males, typically deposit, or they lay these long, J-shaped poops.

And you can see at the bottom left, where it says "TPC 19X," that's the end of a J-shape. So that's a male's poop. So the morphology-- not only do most of them have turkey DNA in it. They look like turkey poops morphologically.

Now, OK, maybe I've convinced you that they're from turkeys. But how do we know that they're domestic? And this brings up a really great question. What do we mean by "domestic" in general? So could these turkeys do tricks? Right, would they roll over? Is that what we mean by domestic?

Well, we have to look at all kinds of evidence to say were these things controlled by humans? Were humans controlling the biology? That's the real question? That is domestication, whether or not they could do tricks.

One of the things-- I had a student take the same poop sample and then extract and purify pollen from them. And what I'm depicting here is the profiles of the number of those samples. And really, what I want you to focus in on is this yellow color. That's all maize pollen.

And the guy who helped us do this said that the only way that much maize pollen could get into, say, that top right sample would be by having been provisioned capsules from the maize. So if you're feeding an animal, it's pooping where you live. That's one thing. And the humans seem to be OK with this.

So they're living side-by-side. And they're provisioning them. And if you're feeding an animal, you better be getting something back from it. That's absolutely true.

So we believe this evidence suggests that there is a mutualism here. The turkeys are living with the humans. The humans are eating the turkeys and using their feathers for, say, feather blankets.

So we analyzed a bunch of turkey droppings from the Four Corners region, from that Turkey Pen Ruins. So we know about the genetics there. And I'll show you the results in a second. The thing is, we wanted to test if they were from Central Mexico, but that bird today is extinct. So how were we going to test that?

Well, this guy I worked with named Bill Lipe, we were thinking about this problem. And he said, well, if we want to know something about the now-extinct Aztec bird, we just need to go to the grocery store. This guy is Bill Lipe-- and I thought, Bill, I think you're crazy. What do you mean? Right, the guy is actually really brilliant.

But I didn't know anything about the history of turkeys. And he said, well, look, if our history books are correct, after Europeans had reached the Americas and interacted with the Aztecs-- and this map is loading up here-- they took the Aztec bird once they encountered the bird. OK, so they took the bird from Central Mexico. They took it back to Europe. And they bred all kinds of fancy breeds out of this.

If you have time and interest, Google search "fancy turkey breeds." It's incredible what humans have done to these things. It's kind of like dogs.

So they brought them back to Europe, bred these fancy breeds. And then, Europeans reintroduced them to North America, which would now be our commercial bird. So it's possible that when you go to the grocery store that your Butterball and any of your commercial birds is actually that extinct Aztec bird today.

So what we did, is we went to the grocery store. And we bought some turkeys. And we sequenced their DNA. And we didn't know how to contextualize it at that point. But that's what we did.

My lab manager is pretty crafty. And she tracked down some other samples that are held at the Smithsonian of the now-extinct or very rare subspecies, gallopavo. These were samples that were collected by people over 100 years ago, maybe when this bird was more common. They happened to be in a different part of the collection. She tracked it down and had these samples sent to us. So we actually had a direct comparison between that bird and the birds that were pooping up in the Southwest and our store-bought turkey.

And so, when we put it together we produced a diagram that kind of looked like this. And what I'm showing you here, is each circle is a different sequence-- kind of like that picture I showed you earlier with the Fish Slough Cave coprolites. Each circle here is a different sequence. And the lines connecting them are mutations that separate them. And they're color-coded by the subspecies.

And so, when we looked at our turkey droppings, the most common type that we saw matched to a type that we see in the Southwest today, where we're putting that star. It's not very common in the Southwest. But we found it in about 85% of our turkey droppings.

The second type we saw here, depicted in this star, is actually the most common type that we see in the Southwest today. These are both Merriam's-- subspecies merriami. The store-bought turkey was here, where I'm putting that black star. And it matched to some of those Smithsonian birds. And we also saw another haplotype, or another sequence, in those Smithsonian birds.

And what this means here, is that out of all the poops we looked at, we only saw two types. That indicates that humans were controlling the breeding in these birds. They went under a dramatic reduction of their genetic diversity. That happens when humans control breeding of animals.

And the types that we see in the Southwest are very, very distantly related to those that we saw in the grocery store and from the Smithsonian samples from Central Mexico. This indicates that the turkey was domesticated at least twice in North America-- once in the Southwest and once in Central Mexico. It also confirms that our history books are correct. And every time you-- if you eat a Butterball turkey, or if you go to Subway and eat a turkey sandwich, you're eating turkey that descends from the now-extinct, Aztec, domesticated bird, OK. If you eat something like a heritage turkey for Thanksgiving, it's going to be a different subspecies.

And our colleagues up in Canada had the same idea. They went to the grocery store in Canada. All their domestic or commercial birds are the same type. They are that now-extinct, Aztec domesticate, maternally-speaking.

When we put all this information together, with the poops that we had studied that are about 2,000 years old, and then we combined that with observations made on bones from about the next 1,000 years-- so we looked at birds over roughly from 2,000 years to 500 years ago-- almost 200 bones from these sites that I'm showing you in the Southwest-- all those numbers-- 85% or so of those birds are one genetic type. And the other 15% are a separate genetic type. And there are some rarer types. But what this shows is long-term management of these bird's biology-- so for 1,500 years, the humans had extreme control over the breeding of these birds.

So I'm going to end with this slide. And I hope you enjoyed the presentation here. And I really hope that you learned some things.