

TB in Captive Elephants

[Announcer] This program is presented by the Centers for Disease Control and Prevention.

[Sarah Gregory] Today, I'm talking with Dr. Barry Kreiswirth about TB in captive elephants. Dr. Kreiswirth is the founding director of the Public Health Research Institute, Tuberculosis Center, at Rutgers University. Welcome, Dr. Kreiswirth.

[Barry Kreiswirth] Thank you.

[Sarah Gregory] Dr. Kreiswirth, as the founder of the TB center at the Public Health Research Center, how did TB in elephants become of interest to you?

[Barry Kreiswirth] Well, it sort of found me. So, just as background, the TB center which I set up in January of 1992, it's actually 25 years, we just had our anniversary, was really a response to an emerging epidemic in New York City which started around 1991, which was due to the co-infection of tuberculosis among AIDS patients. And, that was my introduction to tuberculosis, and as a public health laboratory that does research, we decided to see if we could provide some help to this tuberculosis epidemic because many, many years had passed since we had tuberculosis as a major problem in New York City and it was really fueled by the AIDS epidemic because we now understand that the co-infection of HIV and TB is called the deadly duet. So having said that, we set up a tuberculosis center and the real goal was to, for lack of a better term, to fingerprint the *M. tuberculosis* bacteria from infected and diseased patients so we could provide help to the department of health to track and identify patients with tuberculosis in order they can perform better TB control. So, that's the history of the center, so any study that has to do with tuberculosis and someone could provide us with the actual bacteria we can characterize them and we've been doing this for 25 years, have over 35,000 bacteria in our collection, and the audience should remember, when you're dealing with tuberculosis, you need a very, very special facility to both house and study tuberculosis because the bacteria spreads via the air so you need bio-safety level 3 facilities which we had to establish in 1991/2 to respond to the epidemic. So, long way to say that we've been doing this a long time and any infection caused by *M. tuberculosis*, be it in humans or in animals—elephants, lions, elk, and also in cows, which is caused by related bacteria, *M. bovis*—we can fingerprint. So we have a huge collection and I was basically contacted by a colleague of mine from Arkansas who's also a DNA fingerprinter, named Don Cave, who asked me if I would do him a favor and speak with Dr. Gary Simpson who is an infectious disease physician who also consults for the Albuquerque Zoo because he had an elephant that had tuberculosis and he asked me if we could at least fingerprint the bacteria, and Gary contact(ed) me and it's been a very, very pleasant relationship with Gary over the last, now close to 17 years, studying this very, very interesting story.

[Sarah Gregory] Okay, well, your study now involves three elephants—elephants A, B, and C.

[Barry Kreiswirth] *laughs*

[Sarah Gregory] Can you tell us about them?

[Barry Kreiswirth] Yeah, so this story, and the audience should understand, we actually, when we talk about these elephants, we actually use their names, but believe it or not, for HIPAA

regulations, we weren't allowed to name the elephants as they are in the zoo. So we call them A, B, and C.

[Sarah Gregory] No kidding.

[Barry Kreiswirth] I'm not kidding. So the story begins in 1997, in July, and it should be known, in fact, that I lived in New York City and grew up in New Jersey; we didn't have small traveling circuses come through town. We had the big Barnum and Bailey ones in Madison Square Garden, but many, many towns have circuses that come through, spend a few days and they're sort of roving caravans and many of the key attraction is an elephant which everyone loves. But the downside is elephants are expensive, they live a long time, and if the caravan is having financial woes elephants, unfortunately, can pay the price. In this particular instance, there were three elephants who we'll call A, B, and X who were sent to the Albuquerque Zoo because they were being poorly treated by basically a caravan that went bankrupt. So, one of the elephants unfortunately died and the other two elephants, who we'll call A and B, were received by the zoo, and as soon as you receive, in this case, quite malnourished animals, especially ones that could be sick, they put 'em in quarantine. So, Elephant A was an Asian elephant of 31 years old who was also with Elephant B who was an 8-year-old African elephant. So, an Asian and an African elephant put in quarantine and basically just brought back to good health and they spent one year in quarantine together, each quarter—so every three months—they were tested. Now, what happens with elephants, similar to what we do in humans, there are ways to evaluate whether an elephant, in this case, has presumed disease with tuberculosis, and the way you do that is you do what are called trunk washes. So elephants are very, very smart and their trunks are very able to take, in this case, saline—about a liter of saline—you just put it at the bottom and they take in the saline and they basically slosh it around and they expel it back into the bath. And that's the way you can sample, if you will, to see if they are harboring tuberculosis, and that saline is taken to the laboratory and tested to see if there's any tuberculosis bacteria in that sample. So it's concentrated and tested, and that is done around the U.S. by the USDA to monitor elephant health. And in this case, it was done every three months so they could really evaluate, since these were presumed sick elephants, to make sure that they didn't have TB. And, after one year, so basically four samplings every three months, they were deemed to be “trunk wash negative,” which means they didn't have any active TB that they could test or they could evaluate, and so in 1998, after a year, they were sent into the general zoo population with the other elephants, and everything was fine. And then in October of 2000, that's when one of the elephants—in this case it was Elephant A—the 31-year-old Asian elephant, had a positive trunk wash, which means that the laboratory identified *M. tuberculosis* in her trunk wash. She had subsequent trunk washes and was deemed to be with TB. So, that's the way I got involved, so at that point, there was bacteria isolated from Elephant A and they sent it to Don Cave's lab in Arkansas and subsequently to my lab, and we fingerprinted them. So that's the beginning of the story.

[Sarah Gregory] Okay. And Elephant B? What happened to Elephant B?

[Barry Kreiswirth] OK. So, good question. So, the analogy, in this case, with humans, if mom has TB in the household, the first thing we do is to make sure anyone who has been exposed to mom, which is obviously everyone in the family, everyone she works with, everyone she has close contacts with, would be tested to see if they were exposed. In humans it's called the PPD test, or now we use something called interferon gamma. It's really to see if you've been in

contact, and have thereby been infected. Now, it's a very subtle difference when you become PPD-positive. It doesn't mean you have disease. It means you have now acquired the infection and potentially could come down with disease in your lifetime. So what we do, if we do find people who have been exposed, we treat them in a prophylactic way, which means we treat 'em with a drug to try to prevent them from coming down with disease, and that's called isoniazid prophylaxis. So, in this case, we had an elephant who had active disease—Elephant A who we treated, which we'll talk about next—and we have Elephant B who's been housed with Elephant A for a year. So, the presumption is, uh oh, if Elephant A has TB, we're worried about Elephant B, so they gave Elephant B isoniazid treatment—one drug for six months—in a similar manner how we treated Elephant A, which we'll talk about in terms of the treatment regimen.

[Sarah Gregory] OK. So, there was antimicrobial resistance in Elephant A. Does this resistance herald the same kind of danger that it does in humans?

[Barry Kreiswirth] Yeah. So, yeah, it's a good question about this. So, what we've learned, and this is something that we don't commonly see in humans but, and I knew nothing about this until we started sampling. It turns out, and that's one of the reasons we fingerprinted the TB from Elephant A. She actually was infected with three different strains. One of 'em was rifampin-resistant and the two were susceptible to all drugs that we use to treat TB. So, because one of 'em was rifampin-resistant and the other two weren't, they were cautious about the treatment regimen. The actual way bacteria, in this case TB, becomes resistant to rifampin is due to probably just exposure to the drug without a complete treatment regimen and it actually mutates and the strain from Elephant A had the same type of change that you would see in a patient, from a human patient, who had rifampin resistance as well, so it was a classic strain of rifampin resistance, the other two were susceptible, so it did present a problem much like in humans—what to treat with? Because rifampin is our best TB drug, it's bactericidal and it's the key to any treatment regimen which needs to be multiple antibiotics, in this case, so it did present a problem.

[Sarah Gregory] Ok so, in your study you talk a little bit about how Elephant A's treatment was believed to have probably been inadequate.

[Barry Kreiswirth] Yeah.

[Sarah Gregory] What would you have done differently?

[Barry Kreiswirth] Well, I think we should first talk about how we treated 'em, just because it'll explain some of the concerns about both what we treated with and how we treated with and for how long. So these are the same issues you have treating humans, so in this case, we knew we had a rifampin-resistant strain and two other strains that weren't, and it should be noted that the predominant strain that we covered multiple times was not the resistant one. So there was concern about the rifampin resistance but the likelihood was it was the other strains that we really had to deal with. So, the first regimen was rifampin, so they kept rifampin on just because the other two strains are susceptible. Added pyrazinamide and started with another drug, with INH. So there was INH, pyrazinamide, and rifampin, and they treated her orally. Now, the problem was that, how do you treat an elephant? So, if anyone knows, has either a cat or a dog, dogs are very easy to pill, you put usually a pill in a piece of cheese and a dog will eat it. Cats, not as likely. So, in this case, the elephants are more like cats. So they knew Elephant A really,

really enjoyed watermelon. It was her favorite food, so they very wisely took a large—now remember the other issue is, you give TB drugs in regard to the body weight—so a baby gets X amount of drug, an adult gets X amount more, so it's done by just simply multiplying the amount of drugs per the person's size, so you can imagine, an elephant gets a lot of pills per day. This is given daily. This is literally almost a baggie full of pills that they put inside—a baggie's worth, they didn't use a bag—a baggie's worth of pills inside the elephant, closed up -- I mean the watermelon—closed up the watermelon and gave it to Elephant A and they watched her proceed to gingerly pick apart the watermelon with her toes and then eat the entire watermelon and stomp the pills to destruction. So, Elephant A was very smart. And basically told the vets, you're not treating me orally. So, what they did, and this was really part of, I think, the success story and maybe a lesson for other people treating captive elephants, they literally had to develop a rectal suppository method, which they used these very, very Draconian cages to put her in, with literally her tail sticking out, and a vet, a very heroic vet, would wear these gloves up to his armpit, and literally push these drugs into her daily for two months and then every other day for the rest of the treatment which went on for a year. So,

[Sarah Gregory] Hmm.

[Barry Kreiswirth] This was the heroic way and they treated Elephant B with the one drug—isoniazid prophylaxis—the exact same way. So both were treated rectally. Now, the inside joke here is that one of the issues of treating tuberculosis patients is compliance, or adherence, to make sure you take your drugs for six or nine months because it's hard to do. Drugs don't make you feel well, so making sure people take their medicine is key to treating TB, in this case obviously, Elephant A and Elephant B didn't have much say in the matter.

[Sarah Gregory] *laugh*.

[Barry Kreiswirth] The good news was they actually, you know, were able to treat them. And now, getting back to your question about the resistance issue. So, they treated Elephant A for the entire year using the regimen I said which turned out to be, ultimately, so what they decided was, when they went rectally, they decided not to use the rifampin, they just treated with pyrazinamide and INH, and because that's only two drugs, they were always concerned, during the whole time period, even though she was trunk-wash negative and proved to be, they actually got rid of her bacteria with the treatment, they felt that maybe, due to the fact that it was only two drugs, that it took three months before her trunk-wash was cleared, which took a long time, and pyrazinamide, sometimes in humans, can act on a subset of bacteria and cause the other one, if there's only one additional drug, to become resistant. They were worried that Elephant A may not have had the best of treatment. And that was just in the back story but nothing—they decided that they weren't going to do anything, they were going to just wait and see. And that was what really, that was sort of the end of the story. So we had Elephant B prophylac, Elephant A now cured, and basically, Gary and I said, “Well, you know, this is a nice story.” I think the treatment regimen and the success of doing the rectal treatment was probably something that the readers, at least in the veterinary field, would probably find interesting and we discussed writing it up, but basically, time went on and we never did. And then we, and then there's a second chapter to this story that took place later, in December 2010.

[Sarah Gregory] Elephant C, right, right?

[Barry Kreiswirth] Yep. Elephant C.

[Sarah Gregory] Okay.

[Barry Kreiswirth] So, what happens? I get a phone call. So, Gary and I kept in touch over the years. You know, we talk occasionally, but then next thing I, ya know, it's now 2010, so literally ten years have passed. I think he's called me at the end, or maybe it's the beginning of January 2011 and he said, "I'm calling you because, you wouldn't believe it. The lab just called me from the zoo and they said that there's an elephant—not A or B, but another elephant, we'll call C—they did a trunk wash and they found one positive colony and it grew *M. tuberculosis* and they asked me if I could fingerprint it." And I said, "Of course, Gary." And the obvious question that he was posing, which I knew was, was this bacteria coming out of this elephant similar to the one we had found in Elephant A? And it's exactly what I do with humans, so it was the most logical question and something we do because we had the fingerprint profile of Elephant A, so we would just see if it matches. And that's what we did. And to my surprise, and, I think, Gary's surprise, we found that it was an identical match to the predominant strain that infected, and presumably caused disease in Elephant A.

[Sarah Gregory] So why did it take 11 years?

[Barry Kreiswirth] Well, so this is the interesting thing about TB. So, let's backtrack to Elephant A. So, Elephant A presumably—where did Elephant A get her TB? So, what we think, and this is true in humans, Elephant A comes from Asia. Elephant A probably was weaned as a very young elephant and then sent over to the U.S. for this, in this case, caravan. Elephant A then goes through her life, and then, at some point—age 31—gets in this caravan, gets ill from malnourishment, spends a year in the zoo, and then comes down with TB. So, literally 32 years at age. The first question is, why did Elephant A come down with TB then, and where does it come from? The assumption is that Elephant A, as a baby elephant, was infected in India or Thailand—wherever the elephant came from, but an Asian country—and then harbored the bacteria benignly, like one third of the world does in humans, and then, at some point, due to, in this case, stress, malnourishment, what's called "reactivated" her TB and came down with disease. So, same thing's true now, we believe, with Elephant C. In this case, the presumption is that Elephant A infected Elephant C, most likely during the time when they finally put Elephant A with the entire zoo after the first year after quarantine. So, the assumption is sometime at that point, Elephant A infected Elephant C and Elephant C then harbors the bacteria benignly, and for some transient reason, maybe just that time when the trunk wash was taken, maybe Elephant C wasn't feeling well, had some underlying illness, and that bacteria comes but Elephant C never presented with disease. So, this is perhaps a transient event? Maybe it was just, you know, maybe Elephant C was in the beginning of early disease? The point is, they caught it, and that's what these trunk washes are very, very useful for because, whether they were going to say Elephant C was getting or going to have major disease or not, there was no issue. You treat Elephant C because the Elephant C had a positive bacteria, so Elephant C then got put on, much like Elephant A, a treatment regimen with the notion that we have to treat Elephant C, you know, for completion as active disease. And that's the right thing to do and that's what you would do with humans if you had a positive culture in a human as well, even if it was one bacteria.

So, it was, you know in this case, it's a lot of experience of having Gary, who treats human disease, as a consultant with the veterinarians, who obviously know how to handle the animals,

but it's really the experience of putting two worlds together—the veterinary world and, in this case, you know, people who know how to treat active human disease—to really put your heads together. So, with Elephant C the question was, and we'll get back to the issue of the treatment, now they said, okay, now it's—and I've never had this conversation with Gary—now we have Elephant C, we're going to treat her. We were concerned we treated Elephant A incorrectly, so now we have an opportunity to revisit that. And that's what they did. So, no longer was it going to be just isoniazid and pyrazinamide rectally; they were going to give a third drug. And that third drug was something called enrofloxacin. It's a fluoroquinolone, it's related to ciprofloxacin, but in this case, it's actually used primarily for veterinary medicine. It's not used in human health. So, the laboratory at Denver Jewish actually tested the bacteria to make sure it was susceptible to this drug. It was. So, in this case, they used this enrofloxacin in combination with pyrazinamide, in combination with isoniazid, into the bolus that they used rectally. They treated Elephant C for 18 months with this regimen—very long time—and, if this was a human being, we probably would not have allowed this, because the patient probably would have said, “What, are you kiddin?” They went back to Elephant A and, basically in parallel, re-treated Elephant A with the same regimen.

[Sarah Gregory] Hmm

[Barry Kreiswirth] Just out of concern that they may have not had complete treatment on her. So, basically both elephants, A and C, were treated – in this case Elephant A again and Elephant C for 18 months with three drugs, and both elephants to this day are healthy and fine and still in the zoo and I have an open invitation to visit Elephants A, B and C whenever I want.

[Sarah Gregory] So what about the staff?

[Barry Kreiswirth] Great question. So, you can imagine, in the literature, there are actually documented examples of transmission of animal caretakers getting TB and presumably transmitting TB. So, yes, this was a big concern and for both periods of time, in the early event in 1997 or 2000 when the first Elephant A had disease, and then subsequently in 2010, they did full screening on the workers and I think 178 workers in total were tested and there were no PPD conversions during those times, which means that the elephants didn't infect them. And the other thing that's interesting is so, because we can fingerprint bacteria, the TB, we actually know what the fingerprint of Elephant A and Elephant C looks like and when we compare it to our database among human TB, it actually has what we call, it comes from, geographically, the sub-Asian continent, consistent with the idea that is probably where Elephant A got the strain. It's a “human”—quote unquote—Asian strain which we see in our Indian-Pakistani-Thai population—that area of the world—which is where these strains are more predominant. So, it's a nice story, and then, to finally convince people that we really knew transmission took place from Elephant A to C, we've even did much more sophisticated molecular biology which has actually sequenced the two bacteria to prove they're identical. So we did a lot of work to basically close this loop in this story.

[Sarah Gregory] So, maybe this is an obvious question, maybe not. Is TB in elephants, is it a by-product of living in such close captivity?

[Barry Kreiswirth] Well, I think yes. I mean, but you can also, just so we don't, I mean this has been going on a long time, but in the real African world of animals, unfortunately, there's

transmission that goes on among lions, but in this case, it's not any human intervention. It's due to the fact that there are animals that get *M. bovis* disease, like buffalo. Wild buffalo and wildebeest and these kinds of animals that the lions will eat on and get infected that way. So there was a, there's a big concern in Africa about lions and TB, so in that case, you're actually getting transmission in the wild due to animal prey, clearly in places where humans interact with elephants, much like in India where they train elephants, yeah, I think all of a sudden, put in close quarters like we do with other zoonotic diseases, that's the way we get what we call "jumps," meaning, you know, animal-to-humans and vice versa. But in this case, I think the uniqueness about what we found was, here we've actually documented a case which, you know, it would be hard to argue it didn't happen at the zoo at least, because A and C had never seen each other until they were co-quartered once they transmitted them from the quarantine area into the main zoo. So, they never had crossed paths. Now, one could say, well maybe there's some elephant out there or some other source that had the same exact strain that infected C. Yes. Is it possible? Yeah. You can't argue against it, but the likelihood is that A infected C in captivity.

[Sarah Gregory] Well. It's quite an ordeal that all of, everybody went through. Are there standard guidelines now for treatment of TB in elephants?

[Barry Kreiswirth] You know, there's constant revision of the monitoring, you know, by the USDA with the trunk washes, and I think the good thing that I've learned, just because I had to sort of jump into this field, I have colleagues who do this much more professionally than I do, in terms of overseeing animal health and just learning from Gary. I think these are the types—and that's why we were so keen on publishing it—I think the treatment issue of using rectal delivery, I think, now is something that people clearly consider because it's obviously one way to ensure treatment, and I'm not sure how many facilities have the capability of doing this. But I think the other issue, and this is true now, in many phases of, unfortunately, antibiotic resistance. We always have the concern of which drugs to use due to resistance and also be concerned about using drugs in animals that we don't use in humans. And, so the enrofloxacin, as an example, was an active drug that works in veterinary medicines, we don't use it in humans, so that was also, I think, a very good decision and one that we noted in the paper as being something that I think we advanced, at least, you know, step-wise, the treatment phase, providing another option, in this case, to cure both elephant A and C.

[Sarah Gregory] Okay. Thank you very much, Dr. Kreiswirth; I really enjoyed talking with you today.

[Barry Kreiswirth] Yes, same here. It's a fun story to present to people, especially ones that study TB, and when you can change the, in this case, the patients to elephants, people, for some reason, everyone does love elephants, and you actually get a nice sigh at the end when they see pictures of the elephants back in the zoo, both healthy, and so everyone's happy at the end.

[Sarah Gregory] Yeah.

[Barry Kreiswirth] That's not usually the story when you give TB talk.

[Sarah Gregory] Listeners can read the entire article, "*Mycobacterium Tuberculosis* Infection among Asian Elephants in Captivity" at cdc.gov/eid. I'm Sarah Gregory for *Emerging Infectious Diseases*.

[Announcer] For the most accurate health information, visit cdc.gov or call 1-800-CDC-INFO.