

Detection of *Orientia* spp. Bacteria in Field-Collected Free-Living *Eutrombicula* Chigger Mites, United States

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

[Sarah Gregory] Hello, I'm Sarah Gregory, and today I'm talking with Dr. Loganathan Ponnusamy, a principal research scholar at North Carolina State University. We'll be discussing detection of *Orientia* species bacteria in chiggers in North Carolina.

Welcome, Dr. Ponnusamy.

[Loganathan Ponnusamy] Thank you for having me on your podcast, Sarah.

[Sarah Gregory] I've heard of chiggers as little red things that burrow under your skin, particularly in the South, and they are really itchy. But what exactly is a chigger?

[Loganathan Ponnusamy] Chiggers are larval-stage mites. They are very tiny, and about one hundredth of an inch in length—in other words, four to five times smaller than a mustard seed—and they are difficult to spot with the naked eye. Their typically bright reddish-orange color helps in identifying them. So among the mite life cycle, it is only the chiggers that bite humans or animals. Unfed chiggers usually aggregate on leaves, grass blades, and wait for people or animals to pass by. When chiggers attach to the host organism, they find a suitable area on the skin where they cannot be easily seen and start to feed. The other life stages of the mites are either inactive or predatory, generally feeding on arthropod eggs or small arthropods.

[Sarah Gregory] What causes the itchiness in people?

[Loganathan Ponnusamy] First, I need to say that chiggers do not burrow into skin and feed on blood as many think. Instead, chiggers attach to humans, and they use their sharp, blade-like mouthpart to cut into the skin of the host and then inject salivary fluid, which contains a protein-digesting enzyme, to liquify epidermal tissues for feeding. In the feeding process, a tube-like structure called a stylostome is formed by the salivary secretions. The host's immune response to the foreign enzymes that causes the itchiness.

[Sarah Gregory] I see, okay. So what is their geographic range? As I said, I've only encountered them in the South of the United States.

[Loganathan Ponnusamy] You can find chiggers almost anywhere throughout the world. In particularly the US, chiggers have been reported in all 50 states, but prevalently in the sun belt region of the nation, which includes California, Texas, Utah, Colorado, Nevada, Oklahoma, Georgia, Florida, and the North Carolina State that I live. It makes sense that chiggers are predominantly in the sunbelt region because chiggers are most commonly encountered during warm spring and summer weather.

[Sarah Gregory] I never knew they carried diseases. I thought they were just itchy, as I said. What kinds of pathogens can they carry?

[Loganathan Ponnusamy] Some species of chiggers are known to carry the intracellular bacterial pathogen *Orientia* species that causes potentially life-threatening disease known as scrub typhus. In addition to *Orientia* species, several other pathogens

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like *Anaplasma*, *Bartonella*, *Borrelia*, and *Rickettsia* species being also reported in the chiggers but not being reported to cause with increasing frequency but not being reported to cause any illness to humans.

[Sarah Gregory] So what kinds of pathogens can they transmit to people when they bite them?

[Loganathan Ponnusamy] Yes, when chiggers bite, they can spread bacterial pathogens during the feeding process to people. As of now, we know only *Orientia* is being transmitted via an infective chigger bite. Scrub typhus threatens one billion people globally and causes illness to one million people each year. And because scrub typhus shares some common clinical symptoms with other rickettsial diseases like Rocky Mountain spotted fever, it is easy to misdiagnose it. In fact, a recent review of the literature from 1999 to 2017 estimated that over 60 percent of rickettsial diseases outside of the US were misdiagnosed.

[Sarah Gregory] Why did you do this study? What were you looking for when you began?

[Loganathan Ponnusamy] Well, until 2010, scrub typhus was exclusively reported in the so-called 'Tsutsugamushi Triangle' region stretching from Pakistan in the west to far-eastern Russia in the east to northern Australia in the south. But scrub typhus was found recently in the Middle East, southern Chile in South America, and Africa. As I mentioned earlier, we also know that chiggers are present in the US, but we know nothing about the epidemiology of potential chigger-borne diseases in the US. So we were wondering, therefore, curious to know whether chiggers in the US harbor any pathogens. So that led to do the study.

[Sarah Gregory] Okay. So what are the dates of the study and where did you look?

[Loganathan Ponnusamy] This was started in 2020. We collected free-living chiggers from 10 sites in North Carolina state parks. So the molecular work was done in our lab at North Carolina State University.

[Sarah Gregory] And how did you go about collecting the chiggers?

[Loganathan Ponnusamy] Well, for collecting chiggers, we basically laid black tiles in the woods or tall grassy areas. The black colored tiles are used because they make it easy to see the reddish color of the chiggers on the plate, especially when they are crawling. We picked them up using camel brush and then transferred them into a tube containing ethanol.

[Sarah Gregory] Take a moment here and tell us briefly about your study.

[Loganathan Ponnusamy] So the collected chiggers were taken into the lab, and then we extracted DNA of the chigger sample, then PCR amplification was carried out using a specific part of the bacterial 16s rRNA genes. Then we sequenced amplified genes using next-gen sequencing technologies and compared them with already established sequence database for confirmation of the bacteria species present in the chiggers.

[Sarah Gregory] What did you find?

[Loganathan Ponnusamy] So we found *Orientia* DNA sequences in five of the 10 sampling sites with high prevalence of this bacteria in two of the outdoor recreation areas that we sampled. This is the first time, we identified *Orientia* species in free-living chiggers within the United States. So subsequent phylogenetic analysis showed sequences we obtained from our study matched 99.5 percent to 100 percent in nucleotide identity with *Orientia tsutsugamushi* strain, which was isolated from a human scrub typhus case in Kurosawa village, Japan.

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[Sarah Gregory] Is this pathogen spreading and, if so, why? Or is it just that it wasn't properly identified before?

[Loganathan Ponnusamy] Short answer, Sarah, we don't know if this is a recent introduction into the United States or if the bacterium has been here for years and has not been identified.

[Sarah Gregory] What about animals getting bit—could they create a zoonotic transmission chain?

[Loganathan Ponnusamy] It could be very well. It has been demonstrated outside of the US that small mammals, especially small rodents, are the most common host for chiggers. But in the US, it is yet to be determined if *Orientia* species can be transferred from chiggers to other animals or even if they exist in animals at all.

[Sarah Gregory] What's the epidemiologic significance of what you found?

[Loganathan Ponnusamy] So our results from this study indicate probable local circulation of *Orientia* in chiggers, because the chiggers we used in this study were unfed, meaning that they had acquired the infection from the previous generation through the mother. This suggests that *Orientia* is vertically transmitted and raises the concern for potential exposure of local residents to this potentially serious vector-borne disease.

[Sarah Gregory] So they're born with the pathogen, is that what you're saying?

[Loganathan Ponnusamy] Yeah, that's correct. It goes through the eggs, the mother provided to the egg and the transfer basically happens. That's correct.

[Sarah Gregory] I see, okay. Did anything you discovered surprise you?

[Loganathan Ponnusamy] Well Sarah, I will say no and yes. I was not very much surprised because up to date, *Orientia* species has been discovered in almost every continent but the United States, which made me think that it was here but undiscovered. On the other hand, finding very, very high levels of *Orientia* prevalence in two of the parks really surprised me. I was not expecting that high level (80 to 90 percent prevalence) in two of the parks. That really makes me surprised.

[Sarah Gregory] One of the authors on the study works for the Georgia Museum of Natural History. How was the museum involved?

[Loganathan Ponnusamy] Dr. Dac Crossley is a well renowned chigger systematist and taxonomist. He helped in identifying the species of the chigger that we collected from our sampling sites. He also was instrumental in getting this project started as he offered a great deal of knowledge on how to successfully collect the free-living chiggers using the black tile method. Thank you very much, Dac. It was really very helpful for us.

[Sarah Gregory] I see. And what are the public health implications of what you discovered?

[Loganathan Ponnusamy] Yes, this is a very good question. As it stands now, we do not have concrete evidence that *Orientia* infected chiggers in the United States pose a public health risk. But we do know that all the three known species are known to cause disease in the outside of the US. Although we found the *Orientia* species in our study to be closely related to *Orientia tsutsugamushi*, we cannot say for sure because we don't have any clinical data. For now, the general public should be aware of symptoms when bitten by chiggers and seek medical attention.

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In addition, public health workers and medical practitioners should at least be on the lookout when diagnosing vectorborne diseases with similar symptoms to scrub typhus.

[Sarah Gregory] So you mentioned further studies. What further studies do you think need to be done?

[Loganathan Ponnusamy] So while currently, we know that all three *Orientia* species identified in overseas causes human disease, but we do not know the health impacts of *Orientia* species that we identified within the US. Also, we do not know the prevalence of *Orientia* beyond our sampling area, so we need to do further research to determine the prevalence within the whole US, maybe one or a few states at a time.

[Sarah Gregory] Dr. Ponnusamy, where do you work, what's your job and what's great about that job?

[Loganathan Ponnusamy] So I work at the Department of Entomology and Plant Pathology at North Carolina State University. I am a microbiologist with research interest in arthropod-microbial interactions. Discovering new things and overcoming scientific challenges that at the long run are directly relevant to positively shaping people's livelihoods is what I love to do.

[Sarah Gregory] In a world of climate change, seemingly more and more natural disasters and, as we know, an increase in emerging infectious diseases, what worries you the most?

[Loganathan Ponnusamy] This is a really very good question, Sarah. As a microbiologist, I see climate change causing the geographic expansion of infectious diseases. Rising temperatures is making originally unfavorable environments favorable for pathogens, vectors, and hosts to interact, causing an emergence of infectious diseases. For example, tropical countries like India, where I grew up, have scrub typhus almost all year round. In the US, the temperate climate leads to seasonal occurrences of vectorborne diseases, but climate change is changing seasonal periods, which eventually affects the life cycle of pathogens, vectors, and hosts by allowing infectious diseases to emerge in periods that were previously unfit for those organisms to survive and spread disease.

[Sarah Gregory] Well, on that somewhat unhappy note, thank you so much for taking the time to speak with me today, Dr. Ponnusamy.

[Loganathan Ponnusamy] Thank you very much, Sarah. Thank you for having me on your podcast.

[Sarah Gregory] And thank you for joining me out there. You can read the August 2023 article, Detection of *Orientia* Species Bacteria in Field-Collected Free-Living *Eutrombicula* Chigger Mites, United States, online at [cdc.gov/eid](https://www.cdc.gov/eid).

I'm Sarah Gregory for *Emerging Infectious Diseases*.

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