Genome Insider Episode 3: River Microbiomes From Around The World

ALISON: Hey! I'm Alison Takemura, and this is Genome Insider, a podcast of the US Department of Energy Joint Genome Institute. I usually report these stories from the beguiling city of Berkeley, California. But for this episode, we're going on a field trip.

ALISON: This is the Cache La Poudre river, near Fort Collins, Colorado.

LEAUNDRA: So we're standing next to the river. It's pretty low right now because it's the end of the season. The river is sort of a light greenish color. But it's also really clear. So you can see all of the pebbles and stones at the bottom of the river as well.

ALISON: That's LeAundra Schöpflin, a PhD student at Colorado State University in Fort Collins. And I'm tagging along with LeAundra and a small group of scientists today who are sampling the river. It looks pristine and the scientists tell me it's the only federally designated "wild river" in Colorado. Which means it's undammed and free-flowing. I did wonder about the river's name, though: the Cache la Poudre? Here's what I dug up about the river's history.

LeAundra Schöpflin (left), grad student at Colorado State University, Fort Collins, sampling the Cache la Poudre river with help from Rebecca Daly (right) in 2019. (Massie Ballon)

Way back in the 1820s, a couple of French-Canadian fur trappers were hauling gunpowder along the river, when they got caught in a blizzard. They needed to get to safety, but the gunpowder was slowing them down, so like pirates, they decided to bury it. That's where the river gets its name: because they buried a cache of powder — or cache la Poudre — as someone who can't speak French might try to say. Eventually, even the locals must've given up trying, though, because the name went from Cache La Poudre to the much easier-to-say "Cash La Pooder" river.

Zooming back to my present-day jaunt to the Cache La Poudre, the scientists I'm with are exploring much more about rivers than just their names. They're trying to understand the ecology of rivers, because rivers do more than you might think.

REB: They're responsible for a lot of nutrients cycling, right — carbon and nitrogen cycling. They're also very sensitive to anthropogenic disturbance, whether it's farmland and runoff from agricultural chemicals, climate change.

Rebecca Daly, microbiologist at Colorado State University, Fort Collins. (Massie Ballon)

ALISON: That's microbiologist Reb Daly, one of the PIs on this project. She and Leaundra work with Kelly Wrighton, who's an assistant professor at Colorado State University in the soil and crop science department. And she studies microbiomes in diverse ecosystems. Here's Kelly.

KELLY: People used to think of rivers as just kind of conduits, so you just transported — they didn't actually think of them as actual places where microbial processes were contributing to things like greenhouse gas flux, or carbon and nitrogen or remediating toxic pollutants. But now we're realizing that it's more than just a conduit for chemicals on this planet. But it's actually a place where there's active processes, but we really have no idea what the microbes are that are catalyzing those reactions.

And so that's why we need to be out here studying it. And there's not just us, but a group of people across this globe, working to understand what these processes are, how uniform they are, how consistent they are, so that we can develop ideas and models - to track and predict the flow of contaminants in river systems.

ALISON: So few rivers have been studied, in fact, that if you do a literature review — which is exactly what grad student LeAundra did just a couple weeks before our interview — you'll find that there are fewer metagenomic papers on rivers than there are on faraway deep, Antarctic lakes. There are only about 40 papers on rivers! And, of those, how many had sequenced the transcriptome to look at what the microbes were doing? Just a handful.

KELLY: So there's not very many genomes or kind of community, like how these organisms are processing carbon and nitrogen in the system, we really can't capture that from the information we have today.

ALISON: Kelly has collaborated with JGI for years, and now, she’s working with JGI to investigate river microbiomes. Kelly loves underrepresented ecosystems.

KELLY: Right, I mean, that's like kind of a theme in the lab. Right. So we've worked with JGI on looking at waters that come out of 2,500-meter deep fractured shales, we've worked with JGI looking at the microbes that live in Alaskan fistulated moose. So we, we aren't scared of ecosystems that are undersampled.

But when we started this project, I think the most surprising thing to me was that something like rivers are so poorly represented in the databases. And so when we think about the United States alone, we have 3.5 million miles of rivers.

So to put that in context, that's like, going from the Earth to the Moon 15 times. So this covers a huge area compared to something like fistulated moose, or fractured shales. Yet, when we worked with JGI to look and say, what is in your database? What kinds of samples have you sampled?, it was on the scale of three to five rivers. And so we really can do better, and we can do better quickly. And the way that we can get there quickly and create a resource that's a community resource for everybody, anybody working in river systems, is by all coming to the table, and bringing our samples and our data sets, and saying let's work together to get them all sequenced, so that we can start asking bigger questions that we're not even capable of even formulating today.

ALISON: So, how does an idea like this even get born?

KELLY: This started out of a rapid fire talks at a DOE meeting. And we were asked, what- what kind of science can we not do alone? What kind of science do we need a scientific community, and what are those kinds of challenges? And I proposed this idea that we needed to do more community microbiome science. And little did I know that right after me, James Stegen got up and talked about this WHONDRS project. And so it was this, I sat back down and I was like, why aren't we? Why aren't we working on this, like, this is the perfect kind of experiment for us to do this global microbiome analyses.

ALISON: James Stegen is working on a collaborative science project sampling rivers — a project called “WHONDRS.” That’s w-h-o, n-d-r-s, which stands for

REB: The worldwide hydrobiogeochemistry observation network for dynamic river systems.

ALISON: And the goal of WHONDRS is to take filtered water samples from rivers to get at their dynamic chemistry.

KELLY: And so the first question I asked was, so what are you doing with those filters?

Kelly Wrighton, microbial ecologist at Colorado State University, Fort Collins, on the Cache La Poudre river being interviewed by host Alison Takemura. (Massie Ballon)

ALISON: And James said they weren’t analyzing the filters, which Kelly thought might mean they were just getting tossed. For the next audio clip, I just want to say that when Kelly was telling this story, she was re-enacting her original reaction, and it was really funny, so you’ll hear me laughing in the background.)

KELLY: Wait, you're doing all this work and you're collecting all this beautiful data (laugh) and you’re throwing the filter with the microbes away? I'm like, Send it to my lab! We'll look at it. So that's how this all started. It's like a grassroots, like, wait, we want your waste, we want your trashy filters because those are full of all these microbial cells that could be linked to this beautiful data.

ALISON: James thought that was a great idea. And he clarified that the filters weren’t actually being thrown away, they just weren't being processed yet. But with Kelly's new-kindled interest, the two scientists could work together to really understand river ecosystems in a new way: at the microscale.

ALISON: We're going to take a short break from the river, but I promise, we'll be back. Let's meet another PI on this project -- a grad student, actually. I caught up with her back on campus.

MIKAYLA: I'm Mikayla Borton. I'm a graduate student in Dr. Kelly Wrighton's lab.

ALISON: Mikayla's interested in the tangled web of metabolisms that microbes weave. And she's studied microbes in really different environments, including wetlands, deep underground, and the human body. And now, rivers.

ALISON: What do you think of when you see a river?

MIKAYLA: Well, I will tell you that I didn't think very much of them when, before I started this project, but now I am.. I am very much interested in what is happening there. And actually now when I see a river, I'm like, Oh, I should take a sample because we should get it sequenced and try to expand our database that we have now. But I, yeah, I have a new appreciation for rivers since this project. (Laughing)

Mikayla Borton, grad student in Kelly Wrighton's lab and a co-PI on this JGI-supported project. (Massie Ballon)

ALISON: The project is not only fueling more love for rivers, but also for more collaborative science. As Kelly put it, it's not just a bunch of genome geeks -- it's also hydrologists, biogeochemists, environmental scientists, ecologists... It's a scientific smorgasbord.

There are a lot of PIs on this project. Several of them are at Colorado State: There's Kelly, Reb, and Mikayla, as well as Ed Hall and Mike Wilkins. And then, as previously mentioned, there's James Stegen, along with and Bob Danczak from Pacific Northwest National Lab; Byron Crump, Jerome Payet, and Stephen Good from Oregon State University; Chris Henry from Argonne National Lab, Annika Mosier from University of Colorado Denver, Matt Sullivan from Ohio State, Jacob Hosen from Purdue, and Peter Raymond from Yale. They're all working on this project.

And they're wrangling samples from rivers around the world.

MIKAYLA: So these include 250 rivers worldwide. That's 8 of the 10 largest rivers in the United States. Beyond the United States, we have sampling in over 20 countries, including the Amazon River in Brazil, the Congo, in the Republic of the Congo, the Jordan River in Lebanon, and even 15 watersheds in the McMurdo dry valleys, Antarctica. You can imagine, these are a lot of rivers in a lot of places—

ALISON: Which means: oodles of samples. The team plans to have 1,100 samples collected by the end of this project.

MIKAYLA: So by doing the sampling this way, we can really ask big questions like worldwide questions like what is the biogeography of specific microbes across the world? What is the biogeography of specific metabolisms across the world? And so, you know, all of our samples are collected in the same manner and treated the same way, and so we can really get out big picture questions.

ALISON: It's rare that scientists get to scale across sampling sites, like they're doing in this project. The Department of Energy has a history of supporting research on microbes, but in individual places. One example is the East River in Colorado, where Lawrence Berkeley National Lab has supported the work of Jill Banfield at UC Berkeley.

KELLY: Jill Banfield's metagenomic data has really put, kind of, microbial processes on the map, And the same thing, you can extend that to Pacific Northwest National Lab—

ALISON: —they sample the Columbia River—

KELLY: —you can extend that to even permafrost, in Sweden.—

ALISON: —We actually talked to Gary Trubl earlier about his work on soil viruses in this site—

KELLY: —So I think individually, in localized areas, we have the tools, and we can link these microbial processes to really important, you know, greenhouse gas emissions, and even incorporate microbial processes, potentially into models. But the problem comes when we try to actually scale across systems. And the reason why we can't scale across systems is because the data is not collected uniformly. It's not collected in a format where it's intended to share. And it's not collected in a format where we can actually do these meta analyses to ask, what kinds of traits are shared across watersheds?

So if we look at East River in Colorado, and we look at the Columbia River in Washington, do we need to go out and sample so extensively? Maybe we only need to be looking at a couple target organisms or a couple target processes. But we can't even begin to start sampling or thinking about these questions until we understand what bugs are there, What are they doing? Are they even shared? And we don't have that kind of connectivity in our research programs.

ALISON: But Kelly says collaborating with the JGI changes that. JGI has a Community Science Program, or CSP for short, which gives scientists a foundation for collaborating in the way that Kelly envisions. With JGI, Kelly says that the samples can be processed in the same way, with the same amount of sequencing, to the same high-quality standards.

KELLY: JGI is a, is more than just a sequencing facility for us. They're actually a collaborator that is a key member sitting at this table. And the reason why is that, you know, each one of us has little small pots of money to do this research. But none of us have really been funded to do a lot of microbiome research in this space. And so we may be extensively looking at the chemistry and the hydrology and the biogeochemistry or the fluxes of carbon and nitrogen. But we don't have the resources in any one of these grants to really do the microbiology and do the microbiology the same way across all these 19 or so different funded projects.

And so what JGI gives us, is they give us basically this resource allocation that allows us to kind of standardize the microbiology measurements across this massive team of collaborators. And I want to stress the fact that these collaborators are all not all funded on one funding source. This is patched together from research questions that we each have in our own labs. But JGI is really the glue that provides the resources that allows us to kind of stitch together and do this work.

ALISON: In the past, working with JGI looked different for Kelly's lab.

KELLY: When we've interacted with JGI in this CSP format, we've just taken sequences and processed them ourselves. This project is so big that we can't do that this time. We're going to really need to work with JGI to kind of do the assemblies and craft the data in a way that makes it translatable as quickly as possible to the larger community. I really see this as a true collaboration with JGI being another set of PIs on that proposal.

ALISON: And all the PIs are making the data publicly available. That's not a typical move for scientists. But Kelly and her peers are giving it a shot.

KELLY: And I think what's fun for all of us is everybody who's on this project has sort of agreed that, you have to implicitly trust if you're going to do work like this, like, right. I mean, we all have to come to the table and say — I'm going to bring all my data and, and I have early career people. We all have grad students that we're looking out for, and we want to make sure they can tell their individual stories. But then what is our data-sharing policy? How are we going to use this data? How are we as a team going to do this? Who's going to lead it? You know, all of those questions. I think we've had all to agree, like, we don't know each other. And we're not all friends on this team. I mean, some of us are, but we sort of are friends of friends. So we're tackling that and those are very honest conversations that we're having, and we're going to have to figure out as we go, but I think this new formula for doing science and —this is a really powerful way that JGI can work to really support lots of questions across ecosystems — is if we all kind of come together and say hey, we're going to put aside our own solo stories and think about what we can do together that maybe is better than what we could just do by ourselves.

ALISON: Kelly and Mikayla hope to use data from this rivers project to build on some work they've already done in the Columbia River in Washington state. There, with Pacific Northwest National Lab, they're looking for microbes exhaling greenhouse gases. They set up what are called peepers which are these pore water dialysis samplers to collect the greenhouse gases.

MIKAYLA: What this does is it actually allows us to collect within the sediment of the river collect depth profiles. So greenhouse gas emissions at very fine scale depths, 10 centimeters.

ALISON: Initially, Mikayla had been thinking, "We'll probably see methane coming out of the river sediment, because that's what we've seen in wetlands." Rivers, maybe not that different. But actually, the team found another gas was more dominant: nitrous oxide or N_2O. N_2O is more commonly known as laughing gas when you're in a dentist chair, but it's also a greenhouse gas, which like methane, contributes to climate change. And who's making that gas? To identify the microbial culprits, the scientists were taking gas and metagenomic samples at the same time.

MIKAYLA: Once we have this data, we can start to tease apart what is important and what greenhouse gases we're seeing released from the same sites that we have metagenomes from. And so we can pair that data together and ask, every time we see this organism or this organism's at this abundance, can we predict this amount of methane is produced or this amount of N_2O is produced — and so, this is really important, because the models that we have right now don't incorporate this sort of data. By just building this database, we can ask those questions that we haven't been able to ask before, I mean I'm not sure what will be important, you know, in Antarctica, but it's going to be really cool, whatever we find. (Laughter)

ALISON: For now, this worldwide river project is just getting started, with the first few dozen samples being extracted for their DNA and RNA. But more samples are going to be coming in. And you can help!

Let's get back to sampling on the river and I'll explain there.

So, remember how this worldwide river microbiome project all started because Kelly heard James Stegen speak about his river-sampling project, WHONDRS? James' project was and is a collaboration. And if you're a scientist or part of a citizen science group, you can contribute to the effort! Just reach out to James' team at Pacific Northwest National Lab, and they'll send you a river sampling kit. Here's Reb.

REB: That's the wonderful thing about this, anyone can do it, you can contact WHONDRS through their website and request a kit of your own and send it in and help contribute to this database that we're making.

KELLY: They design these kits to be so user friendly, I mean that anyone can take them off the shelf and use them in the river system is amazing. And let's just look at how well organized they are. You know, it's so fascinating. Little puzzles in there that they're all like, intricately bound in that set..

REB: Oh my goodness, they even give you a pencil.

KELLY: It's making our field packing look put to shame here, Reb! And you're pretty good at that.

REB: I know!

Rebecca Daly opening the WHONDRS sampling kit from Pacific Northwest National Laboratory. (Massie Ballon)

ALISON: Reb and Leaundra then get calf-deep into the river, and they're ready to demonstrate the kit. Let's wade in! Just kidding! That's a figure of speech; I was not prepared to get wet during this interview, so I stayed on shore. What?

LEAUNDRA: In the kit, there are three different vials, a filter, and a syringe. So you're filtering water, and aliquoting it into the different vials and then also preserving the filter for later DNA and RNA extraction.

ALISON: Nice.

LEAUNDRA: Uh, sorry.

ALISON: Uh, that was the syringe.

Now LeAundra takes a syringe-full of river water, and puts the 0.2 micron filter onto the end. Kelly gives some more detail.

KELLY: So the filter is a 0.2 micron filter. And the idea is, is that microbes are present in these waters. And as we push the water through that filter, we're going to capture all those cells. And then what we do is we add a preservative here in the field so that the microbes stay preserved. And then we take it back to the lab and we break open the cells and we take out their nucleic acids, and then we can look at the DNA and the RNA in that microbial community that we've captured in that filter.

ALISON: And the team isn’t just going to be analyzing the metagenomes and transcriptomes of samples; they’re also collecting the metabolomes, or the collection of microbial metabolites.

KELLY: James Stegen has developed some great online tools to look at the transformations of these metabolite profiles so we can see what the microorganisms are eating in these systems.

ALISON: Reb takes some samples as Kelly looks on.

KELLY: Look at those beautiful microbes retained on that filter.

REB: I know! I want to see who's living there!

ALISON: As I mentioned, you, too, can sample a river near you. Just first check that there's a stream gauge collecting data on the changing water level and flow of the river.

ALISON: Okay, so if I'm a really interested excited citizen and I want to sample the river that's right next door, should I double check online, ”Does my river have stream gauge data?”

REB: Yes, we did that not too long ago for sampling, and I simply went into Google and typed “Cache La Poudre river stream gauge.” USGS has a website where you can find the locations of all of those. They show them on a map. So it's really easy to find. There's actually one right in the town of Fort Collins. Next to a bridge that's very accessible. Anyone could go there and sample.

ALISON: Reb and Leaundra finish getting their samples, and once Leaundra is safely on dry land again, I go up to them — Leaundra goes by the pronouns they / them. And they say that they love how the project has crossover appeal with the public. Rivers are so familiar and accessible. And the kit they were using made it easy to contribute to the research.

LEAUNDRA: Using kits like this for citizen science is a good way to feel like you're part of the scientific community without having to be in a lab or work in the field. People love science. I mean, I know so many of my friends aren't scientists, but they always get super excited to talk about the stuff that I'm doing or other scientists are doing as well. Because I think everybody wants to know more about the world around them.

ALISON: Isn't that so beautiful?? I'm going to end the show there.

ALISON: This episode was directed and produced by me, Alison Takemura, with editorial and technical assistance from Massie Ballon and David Gilbert. Genome Insider is a production of the Joint Genome Institute, a user facility of the US Department of Energy Office of Science, and we're located at Lawrence Berkeley National Lab in bodacious Berkeley, California.

So much thanks to our guests LeAundra Schöpflin, Reb Daly, Kelly Wrighton, and Mikayla Borton for sharing their research. If you're interested in river sampling, you can reach out to WHONDRS at pnnl dot g-o-v.

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