Genome Insider S5 Episode 3: A Redesign for Yeast's Genome, Chromosome by Chromosome

Menaka: Today we're starting with a piece of art. It was made in the lab we're covering, and it shows the entire baker's yeast genome, in chromosome form. So that's 16 pairs of chromosomes, and in this piece they're squiggles in bright yellow and light blue, against an eggplant purple background.

Jef Boeke: This particular artwork was hanging in a gallery, and it's about six feet by three feet.

Menaka: Oh wow, so it's really, really big.

Jef Boeke: Yeah, it was blown up, you know, so that people could look at it in the gallery. And it's actually amazingly detailed and beautiful to look at.

Menaka: That's Jef Boeke, the director of the Institute for Systems Genetics at New York University Langone Health. His group made this piece. And the whole picture has a nubby texture, like it's an elaborate tapestry made of tiny beads.

It hangs in Jef's office, because for years, he's worked -- with a bunch of collaborators – to make completely synthetic versions of these yeast chromosomes -- to replace native yeast DNA with human-generated sequences, at large scale.

And if that wasn't wild enough -- the picture above Jef's desk shows these yeast chromosomes, "painted" with yeast. Each dot that makes up the picture is a yeast colony, engineered in Jef's lab to produce a pigment.

Jef Boeke: So six and one half of the chromosomes are colored yellow because they're expressing a beta carotene. And the other, nine and a half chromosomes are colored blue because at the time they hadn't been synthesized yet.

Menaka: This piece was made in 2017, to mark that this team had made synthetic versions of those 6 and a half yellow chromosomes.

Jef Boeke: But if we were to make that again today, they would all be yellow.

Menaka: At the end of 2023, this group published results from the final synthetic chromosomes -- different labs have now made all 16 chromosomes, one at a time. So today, we'll hear what it took for them to get there, and what they've learned as they've designed their own chromosomes for yeast.

This is Genome Insider from the US Department of Energy Joint Genome Institute. Where researchers discover the expertise encoded in our environment — in the genomes of plants, fungi, bacteria, archaea, and environmental viruses — to power a more sustainable future. I'm Menaka Wilhelm.

Our species today is yeast -- the workhorse Saccharomyces cerevisiae, or Baker's yeast. Yeast is a model of model organisms. It's quick to grow and cost effective, in a lab setting, and it's everywhere.

It's the microscopic fungus behind bread and beer -- where I live in Portland, Oregon, Saccharomyces cerevisiae is the official state microbe in recognition of the craft beer industry.

But this yeast is behind a bunch of other industrial products too -- ethanol biofuel for one, and plenty of other chemicals that get made in fermentation. So engineering yeast to be more productive or efficient is a win-win.

And that's one of the ideas behind creating synthetic yeast chromosomes -- if it's possible to replace yeast's native DNA with sequences generated by humans, that could help us engineer yeast to do new things, and understand way more about genomes in general.

Making these new chromosomes opens up all kinds of possibilities for creating stuff we need -- fuels, chemicals, enzymes.

So we're taking a look at these synthetic yeast chromosomes that Jef Boeke and his collaborators have been working to build. We'll specifically get into how they created a synthetic chromosome number 4, because JGI helped out there a few years ago.

But let's start at the beginning. This is a project that's years in the making -- or, millenia, depending on your definition of engineering. As humans, we've been using yeast to make things for a really long time. Here's Jef.

Jef Boeke: it's the first domesticated microorganism, so to speak. We humans domesticated it back in the days of the Fertile Crescent.

Menaka: With such a long history of getting yeast to make things we like, Jef sees it as totally logical that these days we're coaxing yeast to produce things beyond food.

Jef Boeke: And now of course we have yeast that have been engineered to make all kinds of useful products from Jet fuels to vaccines.

Menaka: yeah, so it's both this really interesting model system and also a genome that we could really sort of stretch the limits of what it could do, it sounds like.

Jef Boeke: Exactly.

Menaka: And if we really want to push the limits of what yeast can produce, it would be great to engineer multiple processes at once, to get yeast to be their own little factory, rather than a step in our production line.

Jef Boeke: But typically people are doing this type of work one gene at a time.

Menaka: So one gene, one length of DNA, on one chromosome -- which is great, but Jef and his collaborators want to scale this approach up. A lot.

Jef Boeke: You bring in a set of genes that collectively work together to produce something or to effect some sort of biological change. And so a good example of that would be the pigment pathways that we use in our yeast art project.

Menaka: Like the artwork hanging in Jef's office -- the one that shows all the yeast chromosomes. It's got a dark purple background, for example, because the yeast colonies in that backdrop are pumping out a dark purple pigment: violacein. And those colonies have the multiple genes they need to create that pigment themselves.

Jef Boeke: So the violacein pigment from Chromobacterium violaceum, is synthesized, starting with tryptophan, through a series of five steps into a dark purple colored pigment.

Menaka: It's a little like that proverb about giving a man a fish, and eating for a day, versus teaching him to fish, and eating for life -- modifying one gene in yeast might let them convert one ingredient into something new, but changing a whole set of genes? That lets yeast alter ingredients in multiple ways, to create more complicated and interesting stuff.

Jef Boeke: And this sort of thing isn't by any means limited to making metabolites, we can also use it to make much more complex molecules, you can make structures, viral particles and things like that.

Menaka: So altering a whole chromosome, or the entire yeast genome opens up some really interesting, adaptable end goals. It also asks giant questions about how well we understand the biology of these bits. Leslie Mitchell worked on this project with Jef. And she's now CEO of a startup called Neochromosome, which spun out of this project. Here's how she phrased one of those questions.

Leslie Mitchell: Given all of the more traditional work that has been done, you know, to understand genomes and cells and biology generally, have we like encapsulated or collected enough information to then sort of take it forward and, and, build, design and build — I'm not going to say from scratch because it's really a redesign.

But can you take all of that information and consolidate it in a new version of a genome the world has never seen before and have it work?

Menaka: So that's a giant question -- but Jef and Leslie and their collaborators started with building individual chromosomes, one at a time, not the entire genome. They started out by figuring out how to make and replace yeast's smaller   chromosomes, since the 16 chromosomes in the genome aren't uniform in size. Once that worked, they had bigger dreams.

Jef Boeke: And we were trying to scale the project up to all 16 chromosomes, which at the time seemed like almost an impossible task.

Menaka: They wouldn't be able to do it alone. So they reached out and found partners around the world -- in China, the UK, Australia, and Singapore. And those partners agreed to take on the chromosomes that hadn't been done yet -- many of the medium to large ones, as it turned out.

Jef Boeke: And so I felt a little bit like Tom Sawyer, you know, here I was trying to recruit people to do my work for me, and make much bigger chromosomes than we had ever made. So I felt guilty, and we decided to assign Chromosome 4 to ourselves because it was the biggest one, and it would be challenging.

Menaka: That's the fourth chromosome in the yeast genome, and it would be challenging to the tune of 1.4 million base pairs.

So this chromosome four holds a special place in this episode, because it's very large, and because Jef turned to the JGI to help get this one off the ground.

In 2012, Jef and Leslie submitted a Community Science Program proposal where they requested DNA synthesis -- so JGI would make the starting materials that they'd need to produce a completely synthetic chromosome 4. Here's Leslie.

A yeast art print showing the JGI logo! (Courtesy of Aleksandra Wudzinska)

Leslie Mitchell: It turned out to be a collaboration between Hopkins, and NYU Jeff's lab, JGI, who did a lot of the work to build the smaller segments of DNA. So JGI played this like, incredibly foundational and critical role in construction of the world's, what is largest synthetic chromosome, to date.

Menaka: And Leslie agreed with Jef that taking on Chromosome 4 was the right choice.

Leslie Mitchell: I think he made a good call in taking on the biggest one.

For the following reasons — the biggest one is going to naturally just by chance have the most number of bugs. So you need to have the best crack team, thinking about solving those bugs. And there is no one better in this world than Weimin to be the leader of that project.

Menaka: That would be Weimin Zhang, who's now a research assistant professor at NYU with Jef. Before Weimin worked on the yeast chromosome 4, he was working with partners in China and Japan, to complete a different synthetic chromosome. Here's Weimin.

Weimin Zhang: I was super excited about the idea of using computer-designed sequence, and then to see if the DNA works. So I was fascinated by this idea.

Menaka: Next, we'll get to how Weimin put this giant yeast chromosome together, and what this team learned as they built that big synthetic chromosome 4. But first, a quick break.

Allison: This fall, the JGI will host its annual meeting in Walnut Creek, California -- that'll be September 30th through October 4. You can register at jointgeno.me/JGI2024, and we hope you will. It's a great meeting, but you don't have to take our word for it -- here are some of last year's meeting attendees who kindly shared their thoughts with us.

Bruce Hungate: The meeting's been wonderful. The talks are fantastic, captivating.

Serina Robinson: It's so cool to be here just walking in. I'm looking forward to the tour tomorrow to actually see more of the facilities, but so many people that I've just been corresponding with and finally get to, to meet them and be a part of this community in person. It's been such a joy.

Jerry Tuskan: The talks and posters are all wonderful, but really the greatest outcome of the meeting is the one-on-one discussions and the face-to-face interactions that you can't have really in a Zoom call. And so it's created a platform for all of us to get together and just talk to each other.

Aditi Sengupta: And that leads to very fruitful conversations about science, about technology, about serving society. And all of that has been happening since the last three days, which I'm very excited about.

Allison: That was Bruce Hungate , Serina Robinson, Jerry Tuskan , and Aditi Sengupta . And again, you can register at jointgeno.me/JGI2024.

Menaka: Ok -- back to our synthetic yeast chromosomes. So creating completely synthetic yeast chromosomes is interesting, because it could let us engineer yeast to make all kinds of things -- but it's also a big job. So let's get back to exactly how this team sets that up.

Jef and his team are creating all new, synthetic chromosomes, then putting them into yeast, replacing their existing DNA. And really, they do this bit by bit -- not a whole chromosome swap at once. So they start with the stuff that makes up a chromosome -- a length of DNA. To be specific, a roughly 10 kilobyte chunk of sequence. This kind of chunk is what the JGI put together to let Jef and his team build chromosome 4. Here's Jef.

Jef Boeke: They were designed in such a way as to sort of snap together into what we call megachunks, five to six of these chunks would go together into a megachunk.

Menaka: Those megachunks are what they swap into their yeast strains.

Jef Boeke: And then we would take over from there and, section by section, replace, a megachunks worth of yeast DNA with the synthetic version.

Menaka: So the yeast's native chromosome works almost like a scaffold -- and megachunk by megachunk, they replace pieces of the scaffold until they have an entirely synthetic chromosome, brand new and human-made.

For chromosome 4, our favorite yeast chromosome here, it was Weimin who worked on this. He really specifically optimized the process of replacing this native DNA with synthesized DNA -- and he said it was neat to get to see what was possible.

Weimin Zhang: The cool idea of this project is to ask whether the genetic code we write using a computer can actually have functions in a living organism. So, by synthesizing the chromosome, we are testing this idea. I think so far, we get a positive answer.

Menaka: As Weimin built this chromosome, he also tested what he could change about this genetic code -- since he was generating the synthetic bits himself, he could make all kinds of changes and see if they still worked in yeast. So for example, completely changing the organization of this chromosome, shifting the 3D position of genes, as well as the shape of the chromosome.

Menaka: In terms of like, trying to figure out what will work about computer generated chromosomes, it kind of reminds me of the game Minesweeper.

It's a, like a gridded game where you click through different squares but you don't see, you don't know what's behind the squares and they could be fine, or sometimes the squares have like a bomb and your game is over.

Weimin Zhang: Yeah. After you described it, I think this is exactly the metaphor I would put in.

Yeah. So when we put the synthetic sequence in, into the yeast, sometimes it works, sometimes not — exactly like you described. That's another reason why it took such a long time to publish this long chromosome, because the longer chromosome you have, the more bombs you may have. And once you hit one, you have to, kind of try to figure out why and then redesign the sequence and put it in a harmless way.

Menaka: Yeah, yeah, you're, you get a chance to test the boundaries of what can be changed and what can’t.

Weimin Zhang: Yeah,

Menaka: But it sounds hard.

Weimin Zhang: But I think that's also what we learned, during this process, what can be changed, what cannot.

I mean, the yeast genome already evolved for millions of years. There must be a reason why it is like that, but before actually changing it, we don't know.

Menaka: But now after changing the sequence and the shape of this chromosome, Weimin does know a few more things that are important. So for example, when he reversed the order of the genes in the synthetic chromosome and changed its shape — they called that an inside out chromosome — it was totally fine, viable, but sometimes putting in a small piece of buggy DNA was not.

Menaka: So there are all these things to learn from an individual chromosome. And at first, these chromosomes have all been created separately, by teams around the world. They've produced yeast with a synthetic chromosome 4, or a synthetic chromosome 12, and then native DNA otherwise. Which is super exciting and a long time in the making -- but there's another phase of this work coming up. Getting these synthetic chromosomes all together in the same yeast cell -- so it has an entirely synthetic genome.

Jef Boeke: And so we're well past the halfway mark now, so we have strains that have eight or more of the synthetic chromosomes combined. But that is a somewhat time consuming process, so we think we're still about a year away from having all 16 together in one, single yeast strain.

Menaka: And as a result, that's pretty amazing. But Leslie also pointed out that the process of getting there has been remarkable in its own right.

Leslie Mitchell: I think there's so many incredible achievements associated with this project that are sometimes forgotten in the context of, “Hey, the chromosomes are almost done and the organisms almost built, and then we're going to unlock all this new science.” But like, the path to get there has sort of broken ground on a lot of learnings. And so congrats to Jeff for being that visionary.

Menaka: It's really neat to hear about teams all over the world, doing all of this hard work and then getting it together.

Leslie Mitchell: And to the extent that the groups like JGI had the foresight to say that “That is a meaningful project and we're going to participate.” Groups like Genscript who built the DNA for SYN6. BGI in China has sort of built a whole division and, and, you know, It's launched, careers and new projects and new directions, around the world.

Menaka: A big project with some big returns.

So eventually, when all those chromosomes are together, maybe they'd remake that yeast art in

Jef's office. That original piece from 2017 has the purple backdrop, with finished chromosomes in yellow, and unfinished ones in light blue. Like Jef said, they'd all be yellow now -- but they could really be a lot of colors. The yeast art color palette has expanded a bit since then.

Jef Boeke: We've now engineered yeast to produce pigments of, probably 25 or more shades of, different colors.

Menaka: And as we wrap up, I'll just note that along with providing starting materials for a synthetic chromosome 4, JGI helped out with the beginnings of yeast art also. These art projects started in a class Jef taught, called Build a Genome.

Jef Boeke: And some of the students were able to make little dots of colored yeast appear on an agar canvas, using a special type of robot, which I actually saw for the first time when I visited the DOE facility in Walnut Creek.

Menaka: Aka, the JGI! I was surprised to hear that, and I was not the only one!

Leslie Mitchell: Really? I remember him coming back to the lab and saying, "You guys have to hear about this thing," but I did not know it was at JGI!

Menaka: Yep -- but of course, the art idea was all Jef and his students.

Leslie Mitchell: And I love the circularity of talking about engineered yeast in the context of art that is driven by engineered yeast. It's beautiful.

Menaka: I agree. It's perfect.

Leslie Mitchell: It's perfect.

Menaka: It's a super visual way to see exactly how adaptable these yeast we've been working with for millenia really are. Sure -- they ferment sugars, but with a little coaxing, these microbes can also become van gogh's starry night, or a portrait of a researcher and of course, a representation of yeast. And that's really just a start -- with an entirely synthetic genome, there's probably plenty more that yeast could do.

That was Jef Boeke and Weimin Zhang from New York University Langone Health, and Leslie Mitchell from Neochromosome. We've got links to this synthetic chromosome work wherever you're listening to this episode, and a transcript of this audio posted on our website, also linked in our episode description.

This episode was written and produced by me, Menaka Wilhelm. I had production help from Allison Joy, Massie Ballon, and Graham Rutherford.

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Thanks for tuning in – until next time!