UNDERSTANDING PROBIOTICS AND THEIR DIFFERENT MODES OF ACTION IN THE INFANT MICROBIOME

Featuring:

Adam Baker, PhD
Karyn Wulf, MD, MPH

TRANSCRIPT

Maura Bowen: If you've been a Power of Nutrition Podcast listener for a while, you may remember a three-episode series we produced in September 2020 on probiotics. We created the series in partnership with Chr. Hansen, which is a bio-science company based in Hørsholm, Denmark, who among a host of other innovative solutions, develops best in class probiotics. Those are the live microorganisms that live in the gut and can positively influence health and cognition across the lifespan.

Chr. Hansen has an entire website dedicated to this topic, The Probiotics Institute at The Probiotics Institute.com. If you missed the series or you'd like to revisit it, you can find all three episodes on ANHI.org and on our Power of Nutrition Podcast channel on Spotify.

Dr. Adam Baker of Chr. Hansen and Dr. Karyn Wulf of Abbott served as our experts on those recordings, describing probiotics and their modes of action, strain differences among probiotic species and probiotic safety and quality in manufacturing. To make things easy, you can find link to the series on the transcript of this recording, which we host on ANHI.org.

Now, because we've received such great feedback about the series, I'm excited to welcome Dr. Baker and Dr. Wulf back to the podcast to continue our discussion on probiotic modes of action, because there's so much more to say about this topic. I'm Maura Bowen. I'm recording in the studio today and Dr. Wulf and Dr. Baker are joining us by phone. Doctors, thanks for returning to the podcast.

Dr. Adam Baker: Thank you so much for having us back, Maura.

Dr. Karyn Wulf: Happy to be here.

Maura Bowen: Quick note, when we have these two experts together, I like to step back so they can hold a

one-on-one dialogue and that's the approach we're taking today. Dr. Wulf will do the interviewing and offer her added commentary and Dr. Baker will provide his insights.

Dr. Adam Baker: Yes. Thank you very much. My name is Adam Baker and I'm the Director of Science for Human Health with Chr. Hansen. I've actually held this position for 10 years now. In that role, I've been studying the microbiome very intensely and understanding how probiotics play a role in human health through the different life stages.

Prior to starting at Chr. Hansen, I actually have a career that's been looking at therapeutic and diagnostic development and studying in the areas of oncology and complex diseases. I'm really pleased to be here this afternoon.

Dr. Karyn Wulf: My name is Karyn Wulf and I am a general pediatrician. I have, for the last almost three years, served as the Medical Director of Pediatrics for Abbott.

Dr. Baker, I'd like to set a foundation here today. Probiotics play an important role in helping to develop the microbiome. Can you explain to us what is the microbiome and why is it so important, particularly to infant health?

Dr. Adam Baker: Yeah, the microbiome, It's almost an organ of the human body and we're understanding more and more about it every year as the science is developing through infant ages, through a toddler into childhood, and then continues to change and be modulated through adult life all the way into old age. It's a fascinating, it's a dynamic organ.

One of the things we realize is, it's key how we develop a microbiome from birth, so the understanding about how it is first initiated, what bacteria strains are important at these stages, and what happens. As the microbiome is developing in infants, this is when the key processes, the immunological development is going on, the immune system is developing and then intimate interactions and connections between the microbiome and the development of these other key areas in health.

As we understand more about this, this is very, very important for us to understand how we can modulate and work with the microbiome to actually work and modulate with our health through infancy and how these have long-lasting health effects into adult life.

Dr. Karyn Wulf: Dr. Baker, I'm going to ask you a clarifying question there. When an infant is born, are they born with their microbiome or is that something that develops in the first few years of life?

Dr. Adam Baker: There are some discussions around this, but I think we would like to say that you're born with a blank canvas. The microbiome is the bacteria that live in and on you. What we're thinking about or

talking mainly about today, I would say, is gastro-intestinal microbiome, and that really is seeded from birth usually, where you get your first inoculations or introductions to bacteria and then it develops from there.

Dr. Karyn Wulf: Are there some strategies that we can use to help infants build a healthy microbiome, especially a healthy gut microbiome?

Dr. Adam Baker: That's a very, very interesting question, Dr. Wulf, because what we're learning from the medical science is that the way our environment is can affect the microbiome. We need to make sure that we're exposed to bacteria. That can be through feedings, through touch, through the environment.

If we're born in a more sterile environment, we're not going through a natural birth, but more through a cesarean section in a hospital environment. Some of these things can affect the ways that we can interact with bacteria and start to build up this microbiome. I think it's getting clearer and clearer that the way that we can interact with good bacteria, trying diverse bacteria and get these into our system and start building our microbiome, the better.

Dr. Karyn Wulf: When you're thinking about the infant gut microbiome, how do you identify probiotic strains that you think are important for this development?

Dr. Adam Baker: The microbiome is seeded from birth, and we can see how it's set up and how it's affecting many aspects of health in early and even later life. What we start to understand then is what types of bacteria do we think are important? What types of bacteria can we see that's developing in the infant microbiome, and what types of bacteria would you like to see there and what types of bacteria can really support and help and propagate and accelerate that healthy microbiome?

We want this microbiome to be healthy. We want it to be strong. Mother's breast milk has HMOs or human milk oligosaccharides. They're there to actually feed bacteria, so that's one aspect where we can actually see the types of bacteria that feed off this nutrition that's coming from the mother's milk. They're the types of bacteria we sometimes would like to see within the microbiome, but there are many different types of functionalities we want to see.

Dr. Adam Baker: I talked to you as well about how bacteria can support the immune development, how bacteria are involved in many different things, maybe displacing other types of bacteria. These are the types of bacteria we're focusing on when we think about the infant microbiome.

In particular situations, like when a child is premature, this is even more critical. That child, I would say, is not being introduced to the normal types of bacteria. There's potentially antibiotics around. There's other bacteria, which can cause unbeneficial bacteria. That point is even more key the younger we are and when

we're born more fragile, we introduce the right types of bacteria with these functionalities that I'm talking about so that they can take hold and help build a very robust microbiome.

Dr. Karyn Wulf: Dr. Baker, that was a lot of fantastic information. Just to clarify, you mentioned something called HMOs or human milk oligosaccharides, and that they could be used as food for certain types of bacteria. Is this only for good bacteria or can all bacteria in the gut use these HMOs?

Dr. Adam Baker: What we see is that certain types of bacteria can utilize these HMOs very, very well. In particular, they're called *bifidobacteria infantis* strains. We've studied a lot of these strains and they have the machinery, the genes in their genome, that they can really utilize most of these HMOs.

They may not be the only bacterial strain that can. There's other bifidobacteria strains in the infant microbiome that are also capable of utilizing different types of HMOs. HMOs come in many different flavors and the infantis strains are able to utilize pretty much all of the different HMOs available, whereas other types of bifidobacteria are very interestingly able to utilize very well certain types of these HMOs.

Dr. Karyn Wulf: From what I understand you are saying so far, is that the gut infant microbiome is really important to develop, and then that there are certain strains of good bacteria, these probiotics, that can be helpful in the gut. The ideal food for babies, human milk, actually can support the growth of these good bacteria if the baby is exposed to them. You mentioned something about genomes. Could you explain what a genome is and how, at Chr. Hansen, you study the genomes of the different strains of probiotics?

Dr. Adam Baker: When we're looking at bacterial strains and probiotic strains, we look at the genome and that's the blueprint, the genetic makeup of the bacteria, just like we look at the genetic makeup or the genome to understand a little bit more what's going on inside us. At Chr. Hansen, we're actually studying this in many, many different bacterial strains.

To begin with, you sequence the genome of the bacteria because you want to understand: Is it safe to use? What types of genes are present? You want to make sure there's no deleterious-- or genes that could be construed as being bad for humans like antibiotic-resistant genes. Then what you do is you look at the genome and you try and understand some of the biology, understanding what genes are present on the bacterial genomes and what are they actually doing.

So, when you study the *bifidobacteria infantis*, you can sequence it and you can see they have some special gene clusters, which are the HMO-utilization clusters. These are the genes that are involved with using the HMOs from mother's milk and actually utilizing that to make other food. There's thousands of other genes present on the genomes of all of these bacteria as well and some of them are doing very, very interesting

functions. These are the functions that actually are also called probiotic and may be actually working in how the bacteria interacts with the immune system. They may be working in ways so that that bacteria can out-compete or fight off other bad bacteria.

I think one really, really important point that we need to think about and understand is you can't understand the biology of a bacteria by just sequencing it and understanding of the sequence. You have some very good ideas by doing that, but what you really need to do is you need to take that understanding, that hypothesis you've seen from the genome sequence, and then put it in the lab or put it in the clinic and understand actually, what is the bacteria doing in reality.

If we take the example of HMOs, we've actually sequenced 180 different bacteria, mainly bifidobacteria, to understand how many of them have these HMO clusters, how do they look in the genome. Then what we've done is we've taken these bacteria to the lab and we're growing them on these different HMOs or other prebiotics to understand how do they actually grow.

Dr. Adam Baker: This is the way you bring true power and understanding to the science, because we're not just looking at one bacteria and saying here's the genome and this is how it uses HMOs. We're actually studying it in many different bacteria and understanding how these different bacteria actually do work in different ways.

We've actually made some great videos, if anyone's interested, where you can actually go and see we actually have equipment so that we can grow the bacteria without oxygen at all they're called anaerobic chambers, which is almost like the vacuum of space. There is absolutely no oxygen present.

That's the equivalent of what's actually present in the gut, deep in the colon of the infant. You're studying how did the bacteria actually grow within these environments with these HMOs exclusively. Then you can actually understand how the bacteria grow.

It's not just that functionality we're looking at. We're looking at many of the functionalities as well. We don't just study it on one strain or one strain in isolation. We can study it on the combinations of strains.

Dr. Karyn Wulf: That was just fascinating. You presented us a lot of information there. What I heard you say is that you start with understanding the genome, the genetic structure of these individual probiotics and study those in the lab to better understand how these probiotics might work. Then you actually test them in simulated situations in the lab as well, so that you understand not only what genetically they're able to do, but how they actually function in systems.

You mentioned a lot about the HMO, but there's something else that I wanted to ask you about. That is the survival of the probiotic when it's given orally. If you're given a probiotic and it has to go through the digestive system, how do you know if that probiotic actually makes it down into the colon where that anaerobic chamber mimics?

Dr. Adam Baker: That's a really good question, Dr. Wulf, and I think that almost comes into what we would actually discuss as some of the other functionalities. We actually have artificial gut systems within the lab that actually mimic and actually are validated systems to replicate the stomach and the upper intestine.

What we can actually do then for the individual strains, we can understand how they would survive as they go through the stomach, are introduced to the gastric acid, and are then introduced to bile in the small intestine.

On top of that, it's not just the single strains that we can study in that way. We can study products and product formulations. In the case if we have two or three bacterial strains, we can actually study how the product itself is able to survive and the individual strains can survive as they go through the stomach and the small intestine. This is a very powerful, functional technology for us to understand more about how our products, and the quality of them, and the science is actually working.

Dr. Karyn Wulf: You mentioned the importance of not just single strains, but increasing diversity of the gut microbiome. What else do you know about the functionality of some of the strains that are beneficial to the infant gut microbiome?

Dr. Adam Baker: There are other things and other functionalities that we're actually looking at and are able to actually study within the lab.

One of these is actually studying what is actually happening in the colon in the deep microbiome. We actually have artificial gut systems, which represent the colon, and within these gut systems, we can actually introduce fecal microbiome, so fecal material from infants, newborns. Then what we can do is we can study how that microbiome, how that complex ecosystem, is interacting and what actually happens when we add probiotics to that ecosystem, what actually happens in that young infant microbiome when we add a strain.

What we can do is we can see what would happen when we add the *Bifidobacteria infantis* (DSM 33361) or the *Bifidobacteria*, BB-12® or *Streptococcus thermophilus*, TH-4®. When we add these strains together, what effects are they having on other bacteria that are present? How are they behaving? What are the metabolites

being produced? How is the microbiome performing, or how is it going, so we really start to understand how the infant microbiome can develop.

Dr. Adam Baker: Other types of experiments that we're able to do is we're very, very lucky that we actually have access to blood banks. We actually take human blood and are actually taking the PBMCs (Peripheral Blood Mononuclear Cells) out of this blood and then go through a five-day maturation process where we grow the dendritic cells. These are key immune cells that's part of your immune system. They are actually able to interact directly with bacteria or communicate. What we can do within the lab is actually study the individual strains. What types of immune-signaling profiles are they giving when they interact with dendritic cells, or we can use T-cells within the lab. How do these individual strains communicate with the immune system? This is very, very powerful information for us and starts to give us an idea: are the bacterial strains communicating through a TH-1 type pathway or TH-2 type pathway? Are they maybe producing a pro-inflammatory or an anti-inflammatory type of response?

On top of that, we can actually start to put the bacterial strains together. In the case of the infant microbiome, in one of our formulations, we have the *Bifidobacterium infantis* (DSM 33361) and the *Bifidobacterium*, BB-12® and *Streptococcus thermophilus*, TH-4® together. What we see is that we actually get interestingly different immune interactions when the three strains are put together. We get a very, very nice anti-inflammatory response when we have these three strains together as they would be in the microbiome of the infant. It's not even good enough just to understand one strain. We can start to understand how the different functionalities of the strains communicate together.

Dr. Karyn Wulf: Information that you gave about inflammation and immunity, this all stems from having these good probiotic strains in the gut, and then studying the impacts that they have outside of the gut as well. You're saying having a healthy gut microbiome with the right combination of strains can actually impact the infant's health beyond the gut itself. Is that correct?

Dr. Adam Baker: Yes, exactly. If we just take *Bifidobacterium*, BB-12®, we can also see how that actually works, even in not just preterm infants, but actually then in term infants and we see how they communicate. So we're layering all of this information together. We have examples where *Bifidobacterium*, BB-12® has been demonstrated to reduce the colic in newborns up to seven weeks old.

Dr. Adam Baker: Why we can see what's happening there is, again, the understanding that we have from the laboratory and the genomes. There we're seeing that actually the *Bifidobacterium*, BB-12® is decreasing crying time in the clinical outcomes from this clinical trial, but it's increasing the numbers of bifidobacteria in the gut. We're seeing through cross feeding, that seems to be increasing other bacteria, which is increasing

butyrate levels within the gut, which is a very beneficial metabolite.

The *Bifidobacterium*, BB-12® is creating a bifidogenic environment, encouraging other bifidobacteria to be present. It's not utilizing HMOs itself. We know it can't do that, but yet we can see some of the other functionalities, excluding bad bacteria that's bifidogenic. This is how it's having its function in the infant microbiome. It's so important.

Dr. Karyn Wulf: This is absolutely fascinating to me, Dr. Baker. Thank you for this explanation. As we're wrapping up our conversation today, what key takeaways would you like clinicians to think about when selecting probiotics or thinking about probiotics and the use in infancy?

Dr. Adam Baker: I think it's absolutely key that not all probiotics are the same, that we have to understand and we need to look for and absolutely research and look for probiotics that are backed by science in the sense that the science is there and descriptions are there of how these bacteria are working. The more scientific understanding, the better.

We are looking to develop an infant microbiome that is healthy and is diverse over time. The way that we start with the infant microbiome that we're producing and have a, we call it a bifidobacteria-enriched microbiome, that is good, but that's still a diverse microbiome we're trying to build up with different types of bifidobacteria and other bacteria coming up.

It's important not to think about a single type of bacteria. When you have single types of strains taking over, that's also when you have these pathobiomes in the NICU, which is taking over. They're stopping the diversity. That is not a healthy situation to be in.

I think we are looking, but if we can add one, or even better if we're adding more strains with different functionalities, this is the way that we're approaching to try and start to build that robust infant microbiome, which is seeding and predisposing us to have a healthy microbiome through life, which is very diverse. Really, really important not to think about just one strain but maybe a combination of strains or how these strains are going to interact and build up and support the healthy microbiome overall.

Dr. Karyn Wulf: Well, it sounds like the work that you're doing at Chr. Hansen, from genome to functionality and understanding mechanisms of action and complementary mechanisms of actions when strains are put together is just an incredible burgeoning science. It's been so fun to hear you talk so passionately about these probiotic strains and what you've learned in the laboratory.

Dr. Adam Baker: Thank you so much.

Maura Bowen: Well, as always, a great conversation between you, Dr. Baker and Dr. Wulf. I'd like to thank you both for coming back to share your thoughts on probiotics and how they can be beneficial in infant health. It goes without saying you are welcome on the ANHI Power of Nutrition Podcast any time.

Dr. Karyn Wulf: It's always a pleasure to be here, Maura. Thank you.

Dr. Adam Baker: Thank you so much. It's been really, really great to be back again.

Maura Bowen: For our listeners, if you're looking for more podcasts, we have dozens and dozens across a variety of different nutrition science topics, and you can find them on ANHI.org by clicking resources at the top of the page, then podcasts and videos.

We're also on Spotify now, so be sure to subscribe to ANHI's Power of Nutrition Podcast series to hear the latest nutrition science news and share us with your colleagues. Be sure, also, to visit Chr. Hansen's The Probiotics Institute. That's TheProbioticsInstitute.com to learn more about how probiotic strains can benefit the microbiome across the life cycle. It's full of informative, incredibly engaging content, including the fantastic new video Dr. Baker referenced that was filmed in a Chr. Hansen lab to show the actual science behind the creation of these probiotics. Visiting the site will be solid time spent. Thanks, everyone.