DANNY LENNON: Gab, thank you so much for joining me.

GAB FUNDARO: Thank you for having me.

DANNY LENNON: Yeah, it's been an absolute pleasure, first of all, to meet you, and get to chat a bit. I've lots of questions, so I'll try and pace myself and build up into some of the more detailed stuff. But maybe, just to get started, how would you introduce yourself and the work you do to people?

GAB FUNDARO: I would first say that I'm an educator and a science communicator. I was in academia for four years and was really enjoying my time as a professor there of exercise science. So I primarily taught anatomy and physiology and sport nutrition. Prior to that, my dissertation research was looking at the link between the gut microbiome and skeletal muscle metabolism. But really, while I was teaching for those four years, I was pretty far removed from any of that research and that area, and it wasn't until I switched to working with RP full-time that I started to revisit that work and start to communicate more about the gut microbiome. It was just really good timing because gut health is such a big, kind of hot topic right now, really full of just pervasive myths and misinformation. So people are really kind and

calling me a gut health expert but I sort of prefer the term gut science communicator; or I was on the bodybuilding dietitians podcast and they called me a, I believe, it was a gut health professional. So I kind of prefer those terms because I don't really consider myself necessarily to be an expert, and I think a lot of people call themselves an expert in gut health and they aren't. And because I've taken a break from doing active research in the field, now I have the opportunity to read and immerse myself in a bunch of research, synthesize based on my experience having done it myself, and be able to deliver that with context and nuance that are really so important in the field to really kind of understand what's going on in a big picture sense.

DANNY LENNON: Yeah, I thought a good place to start would be around the idea of bacterial diversity, because, as I think, I said to you the other day, it's pretty difficult when you start looking at individual different types of bacteria, and it's hard to know what the optimal microbiome is, and I'll probably ask you about that. But one thing that seems to be fairly consistent is around the benefit of some degree of diversity. Now, I think I've probably being guilty in the past of thinking that was too simplistic as probably many other people do, and one of the great points of your talk was talking about things like evenness and richness and exactly what we mean by bacterial diversity. So first, maybe can you explain that term to people and then what does the literature to this point seemed to suggest either good, bad, or what we do or don't know?

GAB FUNDARO: Yeah. So when we look at the microbiome as a collection of organisms and all of their genetic material, we can identify who's there and also what they're doing. So we can look at taxonomic diversity, that's who's there. So that's where we're identifying the specific species and strains of bacteria, and then also the non-bacterial organisms like the archaea, the fungi, the protists, and we can even look at

the viruses and the bacteria phages. So we can look at all of the genetic material, look at who's there, we can look at species richness or the number of species, the evenness is the relative abundance of species, so what percent of species we find of various species, what's like their relative contribution to the total population. We can also look at phylogenetic diversity, so in terms of their relatedness to one another, as in, looking at relatives for example. So ideally, we want to have a large number of species, we want to have a relative abundance that is going to help to control the overgrowth of potentially pathogenic organisms, and then we also want to make sure that they are not too closely related; and that's important because these specific strains have sort of niche functions, and what we want to have is a large amount of genetic or functional diversity as well. So what the research has shown is that individuals with obesity, they tend to have lower genetic diversity, so that means there are fewer genes there, that means that there's fewer functional capacity. So we can think about genes as sort of a recipe to make something and we want to have a lot of different recipes; we want to be able to make German food and Italian food and Chinese food rather than just only being able to make German food, we only have the recipes for that. The other thing that we find is that in individuals with obesity or those who are eating a low fiber diet or a diet that's low in microbe accessible carbohydrates, so it's not just fiber, it could also be specific starches and some sugars, that they also tend to have less bacterial diversity or a shift in diversity that may not be favorable, because then we start to see higher levels of potentially pathogenic bacteria.

DANNY LENNON: Awesome. So one of the things that I think you had touched on in your talk was not only do we look at this kind of diversity in it, but when we start to try and piece apart, again, back to that question, what is the optimal microbiome, and correct me, but we seem to see that there are

certain species of bacteria that may be beneficial for specific things, others that may be problematic, but we don't really know what optimal looks like, or if there is such a thing, I guess. So one question that I had that kind of maybe pulls from two opposite directions is, on one hand you made a great point about how that, on average, the microbiome that someone has doesn't change as much as maybe people think over the course of time; however, another thing that I've heard is around how dynamic it can change in response to acute changes in diet, for example. So how do we kind of reconcile those two things of how quickly does it change your response to diet but also, in the grand scheme of things, what does it actually do to our composition if that makes sense.

GAB FUNDARO: Yeah, and that's a great question, and I think, one, that we still sort of have to parse out in terms of what are the actual effects on health, like, why does it matter; and looking at it comparing or differentiating between the time course of the change, so how rapidly do things change versus the magnitude of things changing, so if we consider that we have potentially thousands of species in the gut and there may be more that we just haven't been able to classify yet, and we see that perhaps after a dietary or exercise intervention that we see maybe an entire genus has changed, and so that's a number of various species. So we may see that dozens of species have changed, but that could mean that really, in terms of functionality, not much has changed; or we could see that in functionality a ton has changed despite a relatively small change in taxonomy. Because even if we have a couple dozen species change out of thousands, it's probably not a significant difference in terms of the overall taxonomic diversity. But the functional diversity is one that we really haven't taken as much of a close look at. It's just starting to be more prevalent that they look at both taxonomic and functional diversity and changes. And we do find that as – and it makes sense, that there are changes in gene

expression in response to dietary changes. So if we have a diet, if we have a change from a higher carbohydrate load to a higher animal protein load, then genes change in response and that can indicate that we're also seeing changes in bacteria. But there are some bacteria like bacteroides that are really dynamic, and so they can very easily adapt to dietary changes, so they may not actually change in terms of taxonomy who's there but their gene expression changes, just like we do; like, if we have a diet that's higher in fatty acids versus a diet that's higher in carbohydrates, we'll see the parallels in gene and changes in gene expression as well.

So how quickly things can change, some studies have shown that in diet, in the extremes of dietary changes if we're comparing vegan versus a diet that includes animal foods, just within days they do see changes in taxonomy and gene expression. But when you revert back to your previous diet you kind of go back to where you were before. So the changes that we see in response to diet or exercise do also seem to be transient.

DANNY LENNON: Right, and again, we may not know this or this may be a kind of more speculative question, but if there are those transient changes that occur with a change in diet that can change back, does a change, let's say, someone changed their diet and reduced their carbohydrate intake, maybe their fiber drops, and then you see a change in the gut microbiome because of that dynamic nature, can we say, a certain change is a bad change or does it just mean that's a normal adaptation to this type of diet that may not be problematic or how do we even answer that?

GAB FUNDARO: I think we could probably say both, that there can be changes that are simply changes; like, if we see, you know, I've seen in some cases, the interpretation of data on changes in response to increased protein intake, for example, causes just a change in diversity, but it's not

necessarily one that we've been able to characterize as deleterious. But there are some changes that we have been able to characterize as potentially damaging to the actual anatomy and physiology of the gut. So, when we're looking at sort of more long-term changes, especially when we have like long term studies in rodent models, which we have to be very cautious about how we're extrapolating those things, or if we're looking at just an observational study comparing individuals eating a westernized diet versus those eating a more rural or agrarian diet, that we do see a reduction in diversity, which could certainly be an adaptation, but I think arguably is sort of a problematic adaptation. Losses of diversity have been associated with autoimmune diseases, with type 2 diabetes, even with the potential for developing food allergies if we see that a child has started with a reduction in diversity early in life. So I think that when we look at it in that respect, yes, there are some aspects, some situations where we could say those changes, those adaptations are problematic, especially if we see that we're having then a reduction in the protective mucus layer, covering the colonocytes, that is almost certainly problematic; or if we're seeing chronic intestinal permeability that's leading to greater levels of circulating endotoxin, and we can see that after a single high-fat meal challenge and that has been in humans and that has been replicated and we can also see that in individuals who habitually eat a high fat diet and/or are also individuals with obesity. So we have seen these correlations that have been replicated multiple times across human studies and we've also been able to recapitulate those in rodent studies as well with the microbial transplants.

DANNY LENNON: One of those other kind of dietary strategies that gets talked about in this area quite a lot is the impact of a high-protein diet, and you kind of see this – I think, there's been at least some that would suggest it's harmful; others maybe not so; and that may be down to confounding

of, as you mentioned in your presentation, fiber intake. So how should we think of this area of research with the impact of high-protein diets on microbiome?

GAB FUNDARO: I think that, as you mentioned, the caveat there is the fiber intake. So in our observational studies on athletes that are eating a habitual high-protein diet, but also are getting a high carbohydrate diet and perhaps also a high-fiber diet or at least one that's not so deficient, those studies did show a positive correlation between protein intake and bacterial diversity. But it could also be because they're all physically active as well, so there are a lot of confounded factors there. And then in a recent study they Illustrated that athletes that were in a known fiber deficiency, their protein intake was negatively correlated with diversity. I think the other thing that we really have to touch on that I, you know, because I didn't get in so much into like the background, the methodologies, and the technology that we use to measure diversity, that part of the problem with replicating the studies is that, if we use different methods, we may have very different representations of diversity even with the same sample. So part of that I think is just methodological differences between these studies. So if we are looking at DNA versus 16S RNA markers to identify bacterial strains, we may have over-representation of non-viable bacteria. Depending on the region of the RNA that we're looking at, we can get a closer or less clear resolution in being able to identify to the strain level versus only being able to identify at the genus level. And in some cases, papers are only reporting at like the phylum level, which is like looking at animals with versus without a vertebra. So it's like, that's not really going to tell us much of anything.

Now, technology is improving and more and more we're starting to look at the regions that are allowing us to look at the species level specifically, so we're getting clearer resolution on that data. But I think that is part of it. And

then even how we are storing those samples, how we are processing them to find those bacteria; bacteria that have a cell wall tend to be a little bit harder, and so they can be underrepresented. And then also looking at stool samples is a little bit of an indirect measurement as compared to being able to look into the actual mucosal or luminal contents in the intestines, and really almost no studies are actually doing that. So there are really significant limitations in just how we're collecting and how we're processing our samples; and then if we're comparing our rodent models to our human models, it's postulated that when we are – even if we were to transplant a human fecal sample to a mouse that a vast majority of the microbes actually would not thrive in that mouse. And so we still get a different representation even though at the phylum level we're pretty similar when we look down at the genus and species level that they're pretty significantly different. So I think that's part of the problem is that we just can't replicate because the methods are not exactly the same.

DANNY LENNON: Right. So keeping with fiber, that kind of punch line of fiber is important and fiber can be very beneficial, how would you distinguish between different types of fiber and the roles they because, I mean, there's different ways to divide them up, people hear about insoluble, soluble, they hear about prebiotic or resistant starch, all these different ways to classify it? Are there some ways we should think of as fiber that's more beneficial for the gut than other types or how do you get people to think about that question?

GAB FUNDARO: Yeah, so for the purposes of working with a population that wants something that's pretty actionable, the easiest way that I kind of differentiate between soluble and insoluble fiber – and it's not an exact science, but I say, it's usually a soluble fiber, if you mix it with water, it will get mushy; and then an insoluble fiber will keep its form. So an insoluble fiber

would be something like a vegetable or fruit peel, you put it in water, it doesn't change at all; whereas oatmeal is a great example of something that contains soluble fiber, you put it in water, and it gets really mushy. Now, in most cases, both fruits and vegetables will have a mix of both soluble and insoluble fibers. And so, I stress just having a variety of both. So fruits, vegetables, legumes as well, those are all excellent options for people to just add a little bit to each meal.

The soluble fiber, and also the resistant starch, tend to be more beneficial in terms of looking at supporting the growth of specific bacteria – bifidobacterium is one that's cited quite often – that these bacteria need to have a fermentable source of carbohydrate that is not going to be sequestered by the host. So because we don't have the digestive enzymes to break down these fibers or to break down resistant starch, they pass through our digestive tracts, reach the colon, and then they can be metabolized by those bacteria. So I really promote having a large amount of soluble fiber, and then the insoluble fiber helps to create bulk in the stool and just make for easy comfortable transit. And then there are other carbohydrate sources that could also be utilized like lactose; if we don't have the lactose enzyme, it kind of has the same fate as fiber; sugar alcohols, those can be kind of problematic, they cause a lot of gas and bloating and have a laxative effect also because they're fermentable and so the bacteria can utilize those as well, although that's usually not my first recommendation because they're so gas forming. And so, when we, you know, a lot of people are probably familiar with FODMAPs, those are the fermentable carbohydrates that are usually cited to be kind of problematic, but they're still really important, and it's not about eliminating them entirely, it's just including them at levels that are comfortable for you. But I even recommend, if you want to have a good list of things that will really feed your gut microbes, go for those FODMAPs.

DANNY LENNON: Right. So a couple of questions on prebiotic fiber. So essentially, those fibers that can feed some of these organisms and therefore can lead to short chain fatty acid production, so the big one that gets talked about most I would guess is probably butyrate and people talk about butyrate is good, it helps the health of the intestinal lining and so on. In your talk, you mentioned about also, we kind of forget some of the others, so like propionate, I know there's been some data on like acetate as well. So can you maybe talk about, just generally, an overview of a short chain fatty acid production, why that may be beneficial, and then some of the differences between each of those?

GAB FUNDARO: So yeah absolutely, butyrate is one, and this was something that I had read about even when I was still in grad school, like, before this was such a hot topic, it was just a little rabbit hole that I went down, and they had characterized butyrate as being able to bind certain receptors that help to regulate appetite and insulin sensitivity and even fatty acid oxidation. And so I was like, wow, this is really interesting, this seems like something I want to supplement with, and so I did for a while. I don't even remember if I had any effects from it or anything like that, but it did smell really bad, it smelled like really, really gross. But butyrate production, in general, has been – we see that it's enriched in the guts of people who are physically active, and those who are athletes. So even in general pop, just in recreational exercisers, the more physically fit they are and the more physically active they are, the greater levels of both butyrate producers and fecal butyrate that are seen. So that's promising to show that we see that correlation, and that butyrate production does seem to be a prominent product of the fiber fermentation. And when we increase fiber intake IN individuals with obesity, even through something like inulin supplementation, inulin being a very fermentable fiber, they see increased butyrate production, they also see

increased insulin sensitivity, reductions in appetite and sort of spontaneous weight loss. So if you can withstand the gas that would probably result from that, then maybe inulin supplementation being a “in-the-toolbox” thing for helping to regulate appetite or something like that.

Now, it's interesting because the other two fatty acids, the propionate and the acetate really hadn't gotten that much press. And previous studies showed that there was actually sort of a negative correlation between those fatty acids and physical fitness and butyrate production, and there have been some conflicting findings in sort of baseline fatty acid production in individuals with obesity. So to sort of rewind a little bit, when I was still in grad school, one of the theories about the potential of an obesogenic microbiome was that in individuals with obesity, potentially the microbiota were more efficient at harvesting energy from the diet. So they were fermenting fibers to short-chain fatty acids, thereby increasing the energy availability by up to about 10%, so that could have led to obesity. But at the same time, you have all of these great benefits of butyrate, and I remember thinking, I don't know how to present this in my dissertation in a way that makes sense because these two things are just completely at odds with each other. And there was a recent study done in individuals who were lean or obese, they actually did, I believe, it was an eight-week exercise intervention, and they measured diversity and short chain fatty acids at baseline – no, it was after six weeks of exercise, and then six weeks after a washout period, and they actually found that at baseline, the individuals with obesity had higher levels of fecal butyrate. And I can't remember if acetate and propionate were also elevated, but it was sort of, well, that's puzzling, because these people are physically active but the individuals with obesity have higher baseline levels. Now, the people who are physically active had a greater magnitude of increase – or excuse me, the people who are lean had a greater

magnitude of increase in fecal butyrate because they started lower and they finished higher. So the individuals with obesity didn't really respond to the same magnitude, they kind of, you know, their butyrate levels climbed a little bit but not so much; and they really couldn't explain why. I mean, the researchers were just kind of like, well, this is really interesting, we don't know why that is, it needs to be explored further, because it was sort of at odds with previous literature.

So I think that's still just an area that we're really not quite sure about. And also, if we're measuring fecal butyrate, we aren't sure what's going on with the butyrate that's being produced in the colon and potentially being absorbed. So perhaps they're producing so much more that even if they're absorbing a lot of it, they're still excreting more in the feces potentially. But who knows. Now, in a very recent study, one that I talked about this weekend which was really interesting, they found that in elite marathon runners, so they're – I know that they had 15 individuals who had run the Boston Marathon and then they did follow-ups in two groups, and one of them were Olympic rowers, I can't remember the third one. But they found that a specific genus of bacteria was enriched in their intestines especially following exercise and that these bacteria specifically can only utilize lactate as a substrate, as a carbon source. So they theorized that well, perhaps those athletes are then a little bit more efficient at clearing lactate because they have this bacteria enriched, that could be part of it, but probably more likely is that they're fermenting that lactate to propionate which is then an alternative energy source. So it's a little bit perhaps – it's a little bit better than the Cori cycle where we're converting lactate back to glucose, it's very costly, and so it's just a way for us to really prevent lactic acidosis but it's not very efficient at producing glucose in us for a way that gives us like a net ATP gain; whereas, if these bacteria, now we don't have to exert any sort of

energy because the bacteria are doing it for us and they're clearing that lactate and then producing propionate which is another energy source, so that was super interesting.

And then what they did after that was, in mice, first they confirmed that lactate was able to pass from circulation back into the lumen of the intestine, they did that in mice just by injecting them with lactate. Then they also did a gavage of some specific species within that genus, a specific strain, and the mice that received that strain versus lactobacillus was their control, a lactobacillus strain, had increased exercise performance. And then they did an enema of propionate in those mice...

DANNY LENNON: Wow.

GAB FUNDARO: I know, I was like, wow. I had to do oral gavage, like oral tube feeding in mice when I was in grad school and that was very delicate, I did that for like 20 to 40 mice everyday for three years, and I thought, I don't know if I would trade that in to do mouse enemas. But that also increased their endurance performance. So it's really interesting to see that. It's another potential benefit for those short chain fatty acids, but is still sort of in contrast somewhat to what we've seen in the past or is just another perspective perhaps. So it's not necessarily that propionate is negatively correlated with physical fitness, it's just that in that specific group, that's what they found; and now, in this new group they're finding that there was just another utilization for it.

DANNY LENNON: Yeah, that's really interesting. I know it's something completely separate, but we kind of saw this thing maybe with exercise science and fuel metabolism years back, where it was presumed lactate is this bad thing we don't want. And then we realized, oh hey, we can actually use lactate as a fuel, and it's not necessarily the bad thing that we thought it was, it was just kind of correlated with that. So it's kind of clearer that prebiotic fiber for its

ability to help with the short chain fatty acid production is beneficial. When it comes down to practicality of what people should know about that, there's obviously some, maybe fruits and vegetables we can give people an idea of, particularly some of the vegetables are high sources of that. But I'd be interested to hear your thoughts on – I've seen over the internet over the last few years more people have gone to this area; again, they latch on to this idea of we want prebiotic fiber, so like this high dose resistant starch or particularly people using raw potato starch for example and going super high dose in that – how does that compare with getting that from, say, food sources of that? Is that a problem? Where do you come down on that type of stuff?

GAB FUNDARO: I mean, I usually, you know, and this is sort of a personal bias, and I'm very food first. I have seen some interesting stuff in terms of using resistant starch in rodents as supplemental to a ketogenic diet, and it helped to reduce some of the loss of the diversity that they saw specifically with bifidobacteria which seemed very sensitive to a low fiber intake. But I don't think that we probably would need to supplement if the diet is already sufficient in soluble fibers from fruits, vegetables, whole grains, and legumes, to then add more resistant starch on top of that, sort of like taking BCAAs when you're getting sufficient dietary protein. It's probably not going to cause any harm, but is it really that necessary? Probably not. And then when looking at other fiber supplements like inulin, which is also sold as chicory root, that so often causes GI distress that I really don't think that the benefits outweigh the discomfort that people feel. I do see it quite often in clients that will be really – they'll feel so distressed because they're having really chronic severe bloating and GI distress already from just a high FODMAP load in the diet alone, and then we have ways of kind of addressing that and making sure that we can modify things so that they're more comfortable, still having a very diet. But it's just, I think, it

goes to show that you absolutely can have sufficient fiber from just a plant centric diet without necessarily having the need of supplementing.

DANNY LENNON: Right. So on those kind of food sources, beyond kind of variety of vegetables, fermented foods get talked about quite a bit, things like kimchi and kefir maybe and so on; recommendations around there in terms of, if people have never perhaps eaten them before, how would you suggest kind of including them in the diet?

GAB FUNDARO: Yeah. I have to say there's a funny systematic review that I've posted on my Instagram, it was a systematic review on the beneficial health effects of kombucha, and there was one study in it. So it was like this is not a systematic review, but I think they kind of – they did it in sort of a cheeky way, saying that this is something that is promoted to be super beneficial for health, and like, we don't have any data on it.

DANNY LENNON: Right, we did one okay study, yeah.

GAB FUNDARO: Yeah, exactly. And that has sort of happened with quite a few of the fermented food products. So we don't really have – there's almost no data on anything to do with fermented fruits. Obviously, you can talk about alcohol. It looks like a little bit of red wine might be slightly bifidogenic, meaning increasing the growth of bifidobacteria. But as a whole, alcohol does not seem to be beneficial for the gut. And then in terms of our vegetables, like sauerkraut and kimchi, they do seem to, you know, obviously it's going to increase shelf stability, it does increase the growth or they do contain levels of some probiotic bacteria, so probiotic meaning an organism that could confer health benefit to the host. But we aren't really sure about the load of bacteria there. So is it something that's going to be a high enough dose to even matter? We just don't know that. And then there are going to be plenty of strains that really only

grow well in that food and they're not going to thrive in the gut. So thus far, we don't really have any promising data on fermented fruits, vegetables, or grains in terms of their magnitude of effect on human health. It's not going to be harmful, but it's not going to be like a panacea. Where we probably see the most promising data would be in fermented and – oh, and the other caveat that I have to make, if you've had those foods but they've then been pasteurized, they're not going to contain any live bacteria, and that's what most people are getting, like sauerkraut that's been pasteurized.

DANNY LENNON: Yeah.

GAB FUNDARO: Now, with fermented dairy, that has received really much – there's just more research on that for one thing. So we don't have so much of a problem of the absence of evidence, but there does appear to be solid evidence that dairy in general and that includes fermented dairy, does seem to have either slightly neutral or positive health benefits on a number of indices. So that would be the one that I would say, probably has the strongest evidence for its regular inclusion in one's diet as long as there are no issues with severe lactose intolerance or ethical reasons. They do usually contain, if it's been something that's been cultured or fermented, it usually does contain less lactose. So it's a little bit more digestible if people do have lactose intolerance. So that one's probably the most promising, but it's not something that I would say you'd have to go out of your way to eat a specific amount because we really don't have recommendations for the amounts that people should be eating. I think with regards to dairy, even people who are lactose intolerant, can usually handle up to about 150 mils of milk without too much issue, so you might want to start there; or you could have it as a replacement for one of your protein sources if you wanted to, instead of having like an animal flesh protein source or a plant protein source, you could have fermented or cultured dairy. And when it comes to adding in the fermented vegetables and grains,

potentially just having those as one of your veggie sources, but not thinking that like you absolutely have to go out of your way to include those in your diet. There's really nothing out there on fermented meat. Across the board, we might see some modest changes in antioxidant potential or some micronutrients; but if we're looking at miniscule amounts, and it's a small percentage change and a minuscule amount, the overall magnitude is going to be so small it's not even going to matter. So, I would say, focus first on have very plant centric diet and eating plants at every meal and then use those other things as sort of fun additions throughout kind of smattered throughout your diet, but not thinking that it's going to make a really huge effect.

DANNY LENNON: Right. We can't go through a conversation on the gut microbiome without bringing up probiotics. So open that can of worms. May be a good place to go is any of the positive literature would indicate that certain strains or maybe even a genus is associated with a certain particular outcome, so there seems to be at least hope for targeted therapies in the future, but how most people end up using it is as almost, like prophylactically to stay healthy, I should be continuing to have this. Where would you come down on that? Is there anything that suggests to you that there is inherent benefit to doing that? If so, is there a particular type of product, specific types of bacteria that we should be looking out for in these products or where do we start on this topic?

GAB FUNDARO: Yeah. So I think one of the big points that I like to make is that, as a whole, probiotics research sort of has a problem of replicability. So we may find that a specific strain works in one trial, but then four trials show that it has no effect. And because of the overall positive publishing bias in the literature, we have to realize there are probably a lot more negative findings that just haven't been published yet. So it's important to not go by just one

randomized control trial, we have to look at a whole, you know, the breadth and the depth of evidence that we have for each potential strain. And also, there are so many different strains of probiotic, and even if – so if we're looking at a species, that's very specific; but we can have a subspecies/strain, and that can be something that's different from lab to lab. So we have like lactobacillus rhamnosus is species and then we have these different sub strains, so we have like a GG, an R11 – I mean, there are so many different ones, and so they may potentially have different effects. And then, even the population in which they are effective can differ. So one that's I've seen hugely significant differences in lactobacillus rhamnosus or LGG, L. rhamnosus GG, that one is specifically very effective in pediatric diarrhea, nothing else. So when we're trying to determine what probiotics are effective, it really depends on, okay, effective for what and for whom.

But that being said, yeah, we have seen that there are some strains that are pretty consistently effective for certain things, like, S. boulardii, that's actually not a bacteria, it's a yeast, and it seems to be very, very effective for the prevention and reduction in symptoms of diarrhea. So that's one that I feel very comfortable recommending to people. If they're going to be traveling or if they're having diarrhea after antibiotics or something, that seems to be a very safe and effective way of reducing those symptoms. Likewise, there are some that are fairly effective for reducing symptoms of irritable bowel syndrome. VSL-3 is one that has been shown really effective for inflammatory bowel disease and Pouchitis. And then there are some that have been shown to be fairly effective for reducing or promising for reducing upper respiratory tract infection incidents. But it's really a pretty short list of strains that have been shown consistently effective, that have a net positive balance of findings.

A lot of people I think are also taking these in hopes that it's going to help them lose weight, there's really no evidence to show that they help with weight management at all. I know that some people will say, oh but they even cited in studies on like reduction of waist circumference is one that I see fairly often, but it's not with a concurrent change in body composition or body fat. So there I think it's probably we're looking at a reduction in bloating more than anything else. And then everything else, I mean, there's just really not supportive evidence. There was a recent systematic analysis on mood disorders and they looked at probiotic supplementation and it was, I not kid you, like a 50-50 split, for everyone positive study, there was a negative. So that is just, it's just a mixed bag, and I think with that, because mood disorders are so complex, the problem is that we identify a link between the gut and the brain and then it's like, oh, well, the gut, that's the solution, we just fix that and then everything else gets better. And that's really not the case. And that even when we are using probiotics to help reduce the severity of a disease that is an adjuvant therapy, that means that it's in addition to traditional therapies, that this helps to make the therapy either more effective or reduce the symptoms associated with that therapy. So that's very important.

Also, some probiotics are being sold as like a broad spectrum or kind of like a kitchen sink probiotic one to take every day. There are even gender specific probiotics. There's no evidence to support that a probiotic is going to be super effective for males versus females. Yes, we do see some differences in some bacteria in males versus females, they tend to be higher in males versus females. But, as far as I have seen thus far, we don't have anything to show that we have to have a female specific probiotic. I think that people may be using some of the strains that are common in the vaginal microbiome and then saying, this is the probiotic that you should be taking because they are in the

vaginal microbiome. But the probiotics that you're going to be taking orally are not going to be reaching your vaginal microbiome. And when we look at studies on probiotic supplementation in healthy individuals, if a person is healthy and they don't take antibiotics, nothing else is going on, it appears that the probiotics literally go in one end and out the other and do absolutely nothing on the way through, which is ideal, because that means that you have a very diverse resilient microbiome withstanding perturbations of outside sort of invaders coming in. There's nothing wrong with that. Some individuals may be actually resistant to probiotic enrichment. So they've been able to actually do mucosal samples and luminal samples throughout the gut in individuals and found that in the cases, it sort of depends on what's going on in the cecum, so that's where the small intestine meets the large intestine sort of a little pouch and sort of a hotbed of bacterial growth. And if you have a really diverse cecum, you tend not to be enriched by probiotic bacteria, so that's super interesting to see as well.

And then in other cases, they've actually found emerging research that probiotics may actually inhibit or interfere with cancer drugs. So that's one thing we have to realize. And there are interactions between the microbiome and other drugs as well – metformin is one that they've recently identified. Now, that's actually a benefit, but we still don't really know if there are potential interactions between probiotics and other drugs. One thing that you learn about in medical nutrition therapy is that you have to be really aware of the fact that there are things in the diet that can have significant interactions with pharmaceutical drugs. And so I have no reason to believe that probiotics would be any different; that if you have something that is super effective and in some cases probiotics are, that's a great thing, but it's also a double-edged sword, because that means that they can have interactions with other things.

DANNY LENNON: Right. Some of the points you made there relate to, in cases where we do see a positive benefit, the, I suppose, mechanism or the way in which they're having the benefit. Is that, I think, at least from my understanding, and again, correct me, probably different from what most people think is going on, so I think a lot of people think I'll take this certain probiotic for X length of time, this type of beneficial bacteria gets into my gut and now will proliferate and now will be part of that microbiome; whereas the kind of opposite side to that would be, as you said it probably doesn't hang around, but if you're getting a positive benefit, it's more acutely as you take that, would that be an accurate way to...?

GAB FUNDARO: Yeah, absolutely. So we can actually see that in individuals who are receptive to the probiotics that there is a short term enrichment of those probiotic strains and we could assume that, perhaps we are seeing benefit during that time. There has been some data that's also shown that even if they pass through, in some cases, even if they don't stay, that they can also cause transient changes in just gene expression. So they could still, because bacteria are interacting, all these organisms are interacting with one another, that there's potentially some interaction with them. As they're passing through, they're interacting with the other microbes and causing some changes in functionality. But again, I think part of the problem there is that we just haven't looked into that possibility quite as much as just in most cases we're looking at are there changes in some markers of human health or are there changes in the actual profile of the microbial profile who's there. And then the other issue is that we're often looking at that in fecal samples. And in the one of the great studies that Illustrated that resistance, they showed that everyone's fecal samples were enriched with those probiotic bacteria regardless of whether they were actually enriched in the gut, because those fecal samples are really

representative of the luminal bacteria and especially those in the distal colon, and they actually are significantly different from what we see in the small intestine or even upwards in the large intestine; and we really don't know how they compare across many studies to more of the mucosal samples. So there's like kind of three different things that we're looking at, there are three different populations and trying to assume that they're all going to be the same and really we kind of know that they're not.

DANNY LENNON: Right, that transient effect kind of relates to the antibiotic question that I was going to ask you about, because, again, it's quite common – at least, I've seen two different hypotheses related to this, and I'm not sure how valid they are, so I can ask you – kind of, originally, the logical thing for people to think was I have to go on a course of antibiotics, so when that's over, I will take a probiotic to help again get some good bacteria back in because this antibiotic will have decimated my gut microbiome, there's no point in taking that probiotic during my course because they essentially cancel each other out, that's what people would logically think about it. I then remember seeing an idea proposed, and I think there's some literature behind it that someone that's taking antibiotics could still take a probiotic and get a benefit because of this transient impact I believe on reduced inflammation even though it wasn't going to be colonized.

GAB FUNDARO: Yeah, so there are a few different studies that have looked at this in various ways. One of the most recent, and one of my favorites because it was so invasive, because they were looking at fecal samples and then also took multiple samples through the small and large intestine. They actually found that taking a potent lactobacillus containing probiotic after a course of antibiotics, delayed reestablishment of the native microbiome. The best course of action for those individuals was actually an autologous stool transplant so that was where they ingest their own fecal transplant from

before the bout of antibiotics, so they were re-established within a few days. The people who just took the wait and watch method, so they didn't do anything, they just went back to their previous habits, they fared much better than the people who took the probiotics and were just a little bit delayed compared to the autologous fecal transplant in getting back to normal. So those would be really the two best options most likely. I think part of the problem with taking a probiotic, a potential problem with taking a probiotic after a course of antibiotics, once again, comes down to sort of the species, the number of species that were taking versus the number of species that we have there. So those individuals actually did experience what would be considered dysbiosis because they had a very low species richness. So they were predominantly enriched by those probiotic bacteria, so yes, they absolutely had been enriched. The problem was that, now, for whatever reason, there was a suppression of the other microbes being able to thrive then as well, perhaps because of competition for nutrients and for real estate. So that was really kind of problematic.

In other studies that have looked at healthy individuals taking a course of antibiotics and then taking probiotics, in some cases, they did find that it sped returned to what would be considered a normal non-dysbiotic microbiome. And in other cases, there was no effect. So it could be they were looking at individual characteristics, individual differences, and then also strain specific effects. It could be that some of these bacterial strains are going to interact with others more than some other bacterial strains. They can suppress each other's numbers, they can also encourage each other's numbers. So if we are enriching or if we are taking a probiotic bacteria that produces a substrate for another bacteria, well, now, as one goes up, the other goes up. So we have positive correlations that can enhance the proliferation. But, in some

cases, we have negative correlations between these strains, so that's another issue.

And then there's also the issue of a great number of these studies, that are looking at antibiotic use, are looking at individuals who already have a preexisting condition. So if an individual has dysbiosis associated with irritable bowel syndrome or an inflammatory bowel disease, and they go on antibiotics, their reestablishment of their previous microbiome isn't necessarily a return to a healthy microbiome, it's just reestablishment of what they had before which could still be considered dysbiosis or an unfavorable relative abundance of species. So I think that's, when we are trying to take a look at all the literature and come up with a recommendation, it's really hard to say because our populations are so different and then the potential probiotics are so different. The other thing is, if you consider your microbiome as a selection of organisms that you kind of have to take care of, and you have to feed and water, you have to provide them with nutrients or they don't thrive, if your diet is not sufficient in the nutrients for those organisms, adding more organisms is not going to help. So now you're just going to have much more competition for a bunch of nutrients that aren't there, and so that could be problematic as well. So I would say that probably the best thing to do is to make sure that your diet is very plant centric and you're including all of the fibers that the microbes might want to feed on, getting in physical activity because that is also correlated with increased diversity, and really only taking a probiotic if you have side effects associated – like if you have antibiotic associated diarrhea, then probably you'll benefit from taking the S. boulardii, and you would take that for the period of time for which that you have diarrhea and then you kind of stop. Or other situations in individuals who have had C. diff., there are some strains of bacteria that have been shown pretty effective for reducing recurrence of C. diff. infection, because you need to be taking really, really

strong antibiotics. I would say, that's one of the worst perturbations to the gut that we definitely would see overall loss in diversity, significant dysbiosis, and in that case, supplementation may actually be warranted along with everything else.

DANNY LENNON: Sure. Where do you come down on the current abundance of consumer microbiome testing that we see now of people getting their microbiome sequenced, getting this big bundle of raw data, and then probably interpretive reports too – do you see any usefulness in there, and if there is, what is that?

GAB FUNDARO: I would say, that it's cool and it's fun. So funny story, I had a client who was vegan and they told me that they had one of these, like the GI map tests done. And I was like, oh that's really cool, and I just predicted – I thought, I bet they're going to have a high prevotella load, and they're going to have pretty low bacteroides, they're probably going to high bifidobacterium. And sure enough, they sent me their analysis, and that's exactly what it showed; and I was like, well, I could have told you that based on your diet. So in terms of accuracy and validity, so they're taking a fecal sample that people collect at home, so we already know that the fecal sample is going to be different from what we see in the distal colon, and significantly different from what we see in the rest of the GI tract. So we have to say that, okay, that's going to be one limitation, it's not going to show you the whole picture of everything that's going on there. The other limitation would be this sample is now going to be at room temp for some period of time, that's going to cause the death of some bacteria; they're going to be without nutrients for some period of time, that's going to cause the death of some bacteria; depending on how that lab is then storing the sample, processing the sample, and then depending on whether they're looking at DNA versus the 16S RNA and then the regions of that 16S RNA, those are all potential limitations in the resolution of the data that

we're going to get. Then we know that there are potentially thousands of species of bacteria and these are probably going to give you, a redoubt of like several dozen.

So there are definitely going to be significant limitations in how accurately that data is going to portray, what is going on in your intestinal tract. It can still be kind of fun and interesting to see. I think the problem is when people try to use that as a diagnostic tool to say you have dysbiosis, well, we don't have a specific profile of dysbiosis. And we are also, you know, are we looking at just the relative abundance of species or are we actually trying to quantify them, that's another kind of – there are differences in ways that we can represent our data. And then, in other cases, people are using it as sort of evidence for like a pathogenic infection. Well, there are plenty of bacteria that are potentially pathogenic, that are important inhabitants of the gut, and it's not just the presence of the pathogenic bacteria, but the relative abundance. So they are they're constantly keeping count of their numbers, because a pathogenic bacteria is not going to, on its own, exert all of the energy to produce virulence factors to try and cause a disease in the host without having sufficient numbers to overcome the host's immune system. So if we have some pathogenic bacteria, but we have plenty of non-pathogenic bacteria, it's really not a problem. Or people will use it to say, like, oh you have the presence of candida – well, that's also a normal inhabitant of the gut, so it's one yeast that gets like a really bad rap. So people are using that as a way to say, this is a bad bug or this is a good bug, and there's really no good or bad, it's all about context and the relative abundance, and they all have an important role. In some cases, we don't even know what the role is, it's very hard to point out just one single species and say, this is a bad guy because it could be that, yeah, maybe he's kind of a bad guy, but he's suppressing a worse guy; and so getting rid of him, it's like the old lady who swallowed a spider, and then she had

to swallow whatever else to get the spider. So we kind of get down that rabbit hole.

And I think it really can lead to individuals to down the path of orthorexia just like someone's giving a food sensitivity test, also totally invalid, now you get a list of foods that might be bad for you; or they have these tests that are saying, we can sequence your microbiome and then tell you which foods you should or shouldn't be eating – absolutely, we cannot do that. We are so many decades behind being able to do that. It's absolutely inaccurate. Or people – I think someone just sent me something today on like a breath test that you can take...

DANNY LENNON: That was the one I was going to ask about hydrogen breath test for malabsorption and these are like consumer devices now.

GAB FUNDARO: Exactly, yeah. Now, in some cases, if you have that done by an actual medical practitioner, keeping in mind that there are no gold standards for these breath tests, and that the accuracy ranges anywhere from 20 to 90%, it could be perhaps one additional tool along with a bunch of other diagnostic stuff to determine if potentially you have an issue with small intestinal bacterial overgrowth or malabsorption problem; or even, you know, there are some assays that we can use to determine intestinal permeability, we can also look at just markers of intestinal permeability. And at the extremes, like if a person has something like ulcerative colitis, those are going to be so far outside of the normal range, you're going to be able to say, okay, something is up here. But in many cases, yeah, we do see some changes over time perhaps, you might say that you're at the higher end of a normal range or a lower end of a normal range and these ranges are really, really wide. Even when I was in grad school and we were measuring levels of circulating endotoxin or LPS, it was very difficult to determine where our cutoff was for metabolic endotoxemia, because in some cases

it would be, this person should be septic, this person should be like in the hospital dying according to the concentration of endotoxin, but they're sitting here perfectly fine. And there's really not an agreed-upon range of what is metabolic endotoxemia or a low grade systemic inflammation associated with increased intestinal permeability in LPS versus we know what sepsis is. But below that clinical cutoff, there's just such a wide range of what would be considered normal. So we really can't, you know, I don't think we should be using just this one thing as a diagnostic tool or, here's, what you should eat or shouldn't eat.

DANNY LENNON: So is it essentially the risk of something like a hydrogen breath test that it's, like mechanistically it makes sense and there's some validity to it, but the risk here is of false positives maybe.

GAB FUNDARO: Yes, exactly.

DANNY LENNON: That you're showing something that someone probably doesn't have that much of an issue with, this thing flags, and now it's like, I can't have this thing.

GAB FUNDARO: Absolutely.

DANNY LENNON: So you would only marry up if they would reliably see that and observe that in some other markers perhaps.

GAB FUNDARO: Yes, exactly.

DANNY LENNON: Awesome. You've already been far too kind with your time, so we'll start wrapping up. But before I get to the very final question, let people know where they can find you on the internet, where they can follow you on social media all that type of stuff.

GAB FUNDARO: Yes, so I'm on Instagram and Facebook as vitaminphd. I am also on the Renaissance periodization website. On my Instagram, I have a link to my personal website that lists all of the

podcasts that I'm on, anything that I've written. I have a chapter in the RP Diet Book 2.0 which is available online. I also have several hours worth of video on RP Plus which is Renaissance Periodization's subscription to videos that contain a bunch of lectures. Mike and James do webinars pretty much every couple of weeks, we have forums there, so there's a lot of educational material. If people want to check that out, you can use the code FUNDARO, so my last name, for 20 bucks off a year-long subscription to that. I also have videos posted up on Stronger Experts, so it's another similar sort of education dissemination service. And again, all of those links and all of the discount codes that I have are linked up on the profile on my Instagram. So those are probably the best places to find me.

DANNY LENNON: Awesome. The final question that I always end the podcast on can be completely disassociated from what we've discussed if you wish. It's simply, if you could advise people to do one thing each day that would have a positive benefit on any area of their life what would that one thing be?

GAB FUNDARO: I would say wake up and think of something to be grateful for.

DANNY LENNON: I love it. What a way to end, and Gab, with that, thank you so much for talking to me.

GAB FUNDARO: Thank you.