# TRANSCRIPT

episode #2: Beyond the Bite: Exploring Autoimmune Breakthroughs at the Garvan Institute

### **Zac Stritch-Hoddle** Host

### 00:00

Welcome to Tick Tick Boom. I'm Zac Stritch-Hoddle, your host, and in this series we'll be exploring the mysterious and often misunderstood world of alpha-gal allergy, a unique condition sparked by tick bites. Join us as we unravel the intricate science stories and far-reaching implications of this fascinating medical phenomenon. It's a journey through the complex relationship between humans and nature, a look into how our bodies react in the most unexpected ways. Whether you're intrigued by medical mysteries, passionate about nature or simply looking for an engaging story, this series has something for everyone. Thanks again for joining us for our second episode, where we visit the Garvin Institute in Sydney, Australia, to speak with three leading figures in the field of alpha-gal research. The Garvan Institute is one of Australia's leading medical research centres. It has a staff of around 600 people dedicated to studying cancer, inflammatory diseases like rheumatoid arthritis and multiple cirrhosis, immunity to and vaccines for infections like COVID-19, and allergic diseases, including reactions to ticks and mammalian red meat.

### 01:13

First, I would like to introduce Dr David Langley. Dr Langley was originally trained in DNA techniques. While studying the intricacies of bacterial DNA replication during his PhD, whilst doing a postdoc in the US, he discovered the magic of proteins and crystallized one by accident. This piqued his interest in structural biology and he has been attempting to crystallize proteins and solve their structures using x-ray crystallography ever since. He currently works in the Antibody Therapeutics Lab at the Garvan Institute of Medical Research, where he characterizes the molecular details of antibody-antigen interactions. Good morning, Dr Langley. Thank you for taking the time to speak with me today. Could you tell us why the Garvin Institute is interested in alpha-gal research?

## **Dr David Langley** Guest

### 02:01

Well, I suppose initially because Sheryl van Nunen came to us with her discovery of the link between tick bites and developing mammalian meat allergy, which intrigued us from an immunological perspective, and one thing we decided to look at was whether we could get a molecular understanding of the sugar actually bound to an antibody. So this is what we set out to do, and I'm what's called a structural biologist, which means I try and get a molecular picture of interactions and I use a technique called crystallography. So this involves first growing a crystal of the antibody in complex with the sugar,

and then we hit this with x-rays and collect diffraction patterns and then use a bunch of funky mathematics to back calculate, if you like, and give us an understanding of the molecule within the crystal that produces the pattern that we collect. So that's exactly what we did, and we managed to get a molecular picture, if you like, of how antibodies actually bind the sugar.

## **Zac Stritch-Hoddle** Host

## 02:59

So how does this relate to Professor van Nunen's research on mammalian meat and tick allergies?

## **Dr David Langley** Guest

## 03:10

This is one of the big questions, I suppose, is how does mammalian meat allergy and tick anaphylaxis actually get triggered by actually being bitten by a tick?

## 03:15

So humans don't make the alpha-gal sugar, but we're constantly exposed to it. We have it on the surface of the bacteria that live in our guts, and whenever we consume red meat we are exposed to it as well. In fact, we estimate that about 1% of our serum antibodies recognize the alpha-gal sugar to some extent, yet for some reason that we don't fully understand, this is tolerated without any adverse effects. So the big question, then, is how do we transition from tolerance or energy, which is another word for tolerance, to the potentially more dangerous anaphylactic response, and what role do ticks actually play in this transition? So one possibility is that the previous meal that the tick consumed before you get bitten might be the source of the introduced alpha-gal, or it may be the alpha-gal present on the tick itself, which also makes this sugar and gets injected into us. So why does alpha-gal in the context of a tick bite produce this transition, whereas normal exposure to alpha-gal seems to be relatively benign? So that's what we're interested in.

## **Zac Stritch-Hoddle** Host

## 04:20

So why is it that our body has an allergic response to this sugar in particular?

## **Dr David Langley** Guest

## 04:31

Humans and great apes, as I mentioned previously, don't actually make alpha-gal, but we used to because all mammals generally, apart from the great apes and humans, make this sugar.

## 04:36

So within our genomes we actually have the enzymes there to actually make the sugar. However, they carry some mutations so that we can't do it anymore, even though we've ostensibly got most of the

machinery in place. And the reason for this might be traced or we believe is traced back about 30 million years ago, when the great apes diverged away from the rest of the mammals and we accumulated this mutation. So we no longer made the sugar and, as a consequence, we could develop an immune response against this mutation. So we no longer made the sugar and, as a consequence, we could develop an immune response against this sugar. And it could be that there was a selective pressure 30 million years ago that helped cement this mutation as a good mutation to have, and it could be that this selective pressure was provided by an infectious agent such as malaria itself, which then conferred us with our ancestors, with an advantage to being able to produce an immune response against this sugar.

## **Zac Stritch-Hoddle** Host

## 05:33

I understand that this research has now evolved to have much broader implications for the field. Could you tell me a bit more about that?

## **Dr David Langley** Guest

## 05:40

Having gained a molecular understanding of the way antibodies bind this sugar, we're now striving to develop antibodies that bind this sugar much more powerfully, and once we develop these antibodies, it's possible that this might be useful not just in a diagnostic sense but also as a preventative against infection. So antibodies that bind alpha-gal very tightly might conceivably also be useful in the context of combination therapy with other antibodies, for instance used to fight cancer. So some of these antibodies used to fight various diseases are derived in mice, for instance, and mice, being mammals, also make this sugar, and there have been instances where people getting an antibody treatment then have a nasty reaction against the treatment itself because of the introduction of alpha-gal into the bloodstream. So it could be that you then might co-treat with an antibody that masks this sugar as a combination therapy for certain other treatments, for instance.

## **Zac Stritch-Hoddle** Host

## 06:42

So, Dr Langley, have there been any particularly surprising or eye-opening moments in your research on alpha-gal?

## **Dr David Langley** Guest

## 06:50

I suppose the eye-opening moment for me was solving the structure of the antibody in the context of the sugar, because it revealed that a whole class of our antibodies, without actually going through any of the fine tuning that antibodies normally go through, are capable of binding this sugar to some extent right from the get-go, and this perhaps explains why such a large percentage of our circulating immunoglobulin proteins are able to recognize this sugar to some extent.

## **Zac Stritch-Hoddle** Host

07:21

Dr Langley, if you could resolve one outstanding question about alpha-gal, what would it be?

## **Dr David Langley** Guest

07:27

So the big question really is what triggers the switch from an IgG response to an IgE response. And this is not just a problem in relation to alpha-gal, but with all sorts of allergies: peanut allergy, dust mites, etc. So it's the elephant in the room, in the sense that we don't actually understand what triggers this switch from an IgG switch, which our bodies have fairly good control over, to an IgE switch which is potentially quite nasty. So unravelling that in the context of alpha-gal might actually give us the answer to why this switch happens with all sorts of other allergies and now for a discussion about the health implications of alpha-gal allergy.

## **Zac Stritch-Hoddle** Host

08:15

I would like to introduce Professor Antony Basten. Professor Basten is a medical doctor and distinguished figure in the field of immunology. His career includes prestigious appointments such as Officer in the field of immunology. His career includes prestigious appointments such as officer in the General Division of the Order of Australia, fellowships in the Australian Academy of Science and the Australian Academy of Technological Sciences, and the role of an inaugural executive director of the Centenary Institute of Cancer Medicine and Cell Biology. Professor Basten has over 270 publications to his name and is currently a professor of medicine at the University of New South Wales, as well as an honorary senior principal research fellow in immunology at the Garvan Institute of Medical Research. So, Professor Basten, at the centre of all this research is the allergic reaction to ticks and alpha-gal in red meat and other mammalian animal products. Could you describe the underlying process of an allergic reaction to alpha-gal?

## **Professor Antony Basten** Guest

09:15

Well, allergenic sensitising antibodies cause allergic reactions, and they do so by attaching to the surface of what are known as mast cells in body tissues like the skin, the airways and the intestine. These cells contain packages of inflammatory molecules like histamine. On exposure to alpha-gal in tick saliva or mammalian red meat, the alpha-gal binds to the allergenic antibody which is sitting on the surface of those cells. As a result, the mouse cells burst open and release histamine, causing acute inflammation and an allergic reaction involving those tissues like skin, lungs and intestine. If extensive enough, anaphylaxis occurs.

## **Zac Stritch-Hoddle** Host

10:13

You mentioned sensitising antibodies before. What does it mean to be sensitised to alpha-gal, and do we know how many people are affected by it?

## **Professor Antony Basten** Guest

## 10:21

Well, alpha-gal sensitization means that a person has been exposed systemically to this sugar, the most common cause being a tick bite. As a result, the person has the potential to make allergenic or IgE antibodies to it on subsequent exposure. This can be in the form of red meat, other foods of animal origin such as gelatin, and even some vaccines. The best estimates we have for sensitization of people living in the Sydney Basin is around 12%, although it could be higher among those living near the coast, where ticks are more common living near the coast, where ticks are more common.

## **Zac Stritch-Hoddle** Host

## 11:09

Why is it then that only 30% of alpha-gal sensitised people experience an allergic reaction to alpha-gal containing products?

## **Professor Antony Basten** Guest

## 11:14

Well, this figure applies to the Sydney Basin, where the incidence of mammalian meat allergy is around 1%. The reason why only 30% of sensitised people experience an allergic reaction at any point in time is due to the influence of a number of cofactors, as mentioned previously by Professor van Nunen. These include amount of alpha-gal ingested, co-ingestion of alcohol or spicy foods, exercise and prior administration of anti-inflammatory drugs. In addition, there is a rare condition known as mastocytosis, characterised by a large increase in mast cells that can amplify the chances of a severe allergic reaction. This condition can be detected by what is called a trypsin assay.

## **Zac Stritch-Hoddle** Host

## 12:10

I understand there's also research suggesting an increased risk of coronary heart disease in those who are alpha-gal sensitised. Could you explain what this research could mean?

## **Professor Antony Basten** Guest

## 12:20

The case of coronary heart disease, there are two studies, one from the USA and one from here in Sydney, Australia, showing that alpha-gal sensitization is independently associated with an increased risk of developing plaques that cause narrowing of the coronary arteries and predispose to heart attacks. In other words, alpha-gal sensitization is an additional risk factor for coronary heart disease, over and above high blood pressure, cigarettes, alcohol consumption and obesity. The important thing, though, to

remember is that people can be sensitised by a tick bite without necessarily developing a clinical allergic reaction.

## **Zac Stritch-Hoddle** Host

## 13:08

If alpha-gal sensitivity doesn't always result in an allergic reaction, should people still take precautions to avoid developing it?

## **Professor Antony Basten** Guest

## 13:17

Yes, they certainly should, and there are several reasons for that. First, some people can develop allergic reactions to other substances in tick saliva, including the proteins there. Secondly, as just mentioned, it may reduce the risk of coronary heart disease. And thirdly, tick bites can become infected, although here in Australia there is no evidence whatsoever for the presence of the germs causing Lyme disease in our particular tick populations. What's more, measures to avoid sensitization are simple, for example, wearing protective clothing when you go on a bushwalk and removing ticks correctly. For those already sensitised, being aware of all the cofactors predisposing to these reactions is vitally important.

## **Zac Stritch-Hoddle** Host

## 14:10

Up until now, our discussion is primarily focused on the allergic responses to alpha-gal. However, there seems to be a flip side to this. Could you tell us about the potential benefits of the alpha-gal immune response, particularly in relation to its implications for disease control and prevention?

## **Professor Antony Basten** Guest

## 14:28

Regular internal exposure to alpha-gal on microbes normally present in the intestine can lead to another type of antibody called IgG, which is non-sensitizing but is important for protection against certain common infections. For example, non-sensitizing IgG antibodies have been shown to decrease transmission of malaria by mosquitoes. Presumably this is due to the fact that the malarial parasites carry alpha-gal on their surface, which can be targeted by the antibodies. The same may apply to transmission of another common infection in the developing world, namely tuberculosis.

## **Zac Stritch-Hoddle** Host

## 15:15

Another common infection in the developing world, namely tuberculosis. Considering the recent impact that COVID-19 has had on the world, I have to ask, is there any connection between alpha-gal research and COVID-19?

## **Professor Antony Basten** Guest

15:38

That's an interesting question. An association has been reported between the severity of COVID infection and the level of antibodies to alpha-gal Surprising and very interesting. In other words, more severe disease is associated with lower levels of anti-alpha-gal antibodies. Well, what does this mean? Is it of any value? This raises the prospect of boosting alpha-gal antibody levels through the use of probiotics, which, in turn, could benefit patients with severe COVID disease.

## **Zac Stritch-Hoddle** Host

16:04

And finally, Professor Bastin, if you could resolve one outstanding question about alpha-gal, what would it be?

## **Professor Antony Basten** Guest

16:12

As far as we're concerned, in the laboratory where I work, we really want to know why it is that 10% of people are allergic and make sensitising IgE antibodies to alpha-gal and lots of other allergens, and 90% of the population make different protective but not harmful antibodies.

## **Zac Stritch-Hoddle** Host

16:37

For our last guest, I'd like to introduce Professor Daniel Christ, who unfortunately only had limited time to speak with me. Daniel joined the Garvan Institute in 2007 as head of antibody therapeutics to translate structural and genomic advances into drug candidates and treatments for cancer and inflammatory conditions. He's also the director of the Center of Targeted Therapy at the Garvan Institute. Professor Christ, if you could resolve one outstanding question about AlphaGo, what would that be?

## **Professor Daniel Christ** Guest

17:18

Is what we call this IgG to IgE switch, where the immune system switches from producing antibodies that neutralise and protect to those that mediate allergy and autoimmunity, and I think it's fair enough to say that we don't fully understand that question at the moment, but it's certainly a matter of intense research and tests. It's probably related to the fact on how the alpha-galactose antigen is presented in the context of the tick bites or in the skin, and then also in the context of tick proteins and then also other tick components such as alpha-galactose linked lipids and the like.

## **Zac Stritch-Hoddle** Host

### 17:54

Professor Christ, have there been any particularly surprising or eye-opening moments in your research on alpha-gal?

## **Professor Daniel Christ** Guest

### 18:01

So I think one of the things that surprised us is the discovery of what we call sort of public antibodies.

### 18:07

So if you look at the human immune system, we have different germlines or sort of different flavours of antibodies, and in fact humans have over 70 of those right, and what you get in many cases is, you know, what we call a polyclonal response, so that all of the 70 or most of the 70s are used, and that's also the case in the alpha-galactose response.

### 18:30

However, we did find certain preferences and in particular we found one germline which is called 3-7, which didn't dominate the response but was sort of overrepresented in the response. And then we also did a lot of structural and biophysical characterization of the response and we found that this germline so antibody germline 3-7, had sort of basically genetically encoded properties that seems to make it particularly suited for binding alpha-galactose, and I mean that to one extent provides an interesting biology insight. So we do seem to have some genetic predisposition of making antibodies that can recognize, structurally recognize alpha-galactose. And then also its early days. I think this might also open up some pathways towards the development of therapeutic antibodies, because it's basically nature telling us what is an optimal solution to this recognition problem. So how do we bind alpha-galactose with maximum affinity and maximum specificity?

## **Zac Stritch-Hoddle** Host

### 19:27

On behalf of TiARA.org.au, we'd like to thank everyone at the Garvan Institute for speaking with me today. Everyone at the Garvan Institute for speaking with me today and, as always, a reminder if you're ever bitten by a tick, please freeze, don't squeeze. For more on preventing alpha-gal allergy, visit tiARA.org.au, where you'll find research papers and show notes. Make sure you stay tuned for our next episode with Stephen Doggett, a renowned Entomologist from Sydney, Australia, discussing all things tick related. Thanks for tuning in to Tick, Tick, Boom!.