Brief bio (120 words)

1. **Why do you think research in to the gut microbiome has gathered so much interest recently in the field?**

   Microbiological testing has developed enormously over the last 10 years. It is now possible to obtain a complete genetic breakdown of a person's gut microbiota in one day, whereas in the past that would have been years' worth of work. The improvement in analyzing and identifying bacteria has resulted in an explosion in understanding what bacteria are natively present in the gut. The next step should be to continue to analyze the diversity of gut microbiota of patient groups with certain disease states. There is increasing evidence demonstrating that patients with certain disease states are deficient in or have an excess of certain bacteria.

2. **How is the gut microbiota affecting our immune system? How can we take advantage of this for our benefit?**

   It is difficult to know the exact role of gut microbiota in health and well-being. It is quite clear that certain bacteria produce metabolites and cause gut inflammation, resulting in an immune response. We are, however, yet to fully established what compounds are inflammatory or cause an immune response and how they do that. The role of bacteria in these responses is even more vague. While in some instances, such as *Clostridium difficile,* bacteria can produce compounds that have negative effects on the body, it is becoming increasingly clear that certain bacterial groups produce metabolites that the body needs, or the body reacts to. The first step to questioning what compounds are produced and how that influences disease, is identifying bacterial absence or presence with certain disease states.

3. In recent years, you have been studying the use of probiotics. Could you tell us a bit about your own work in this area?

A few years ago, our group started to ask the simple question: if a patient was to buy a probiotic, what sort of probiotic should they buy and how should they take it?

We decided the most important challenge for a probiotic would be stomach acid transit and thus, we added various off-the-shelf probiotics to hydrochloric acid in beakers (in line with each product's manufacturer recommendations) to attempt to recreate the process of a patient swallowing each product. Next, we questioned how long bacteria would reside in the stomach. We estimated around 30 minutes, after which bacterial samples were removed from the acid and viable bacteria were counted. We also determined bacterial re-growth, when added to a growth medium. We found that for any freeze-dried solid product such as a sachet, capsule or tablet; rehydration of those bacteria in acid resulted in poor viability. A key recommendation from that initial study was therefore, if a patient is going to buy a probiotic, they should opt for a liquid-based product instead of a solid-based product. Manufacturers will often argue that freeze-drying is a means of preserving bacteria and is how culture collections are maintained. While this is true, in a culture collection you only need one viable bacterium and ultimately, it will grow back. However, to exert a clinical effect in the body, it is generally necessary to swallow a reasonably high number of bacteria, so we recommend a liquid-based product.

This study also showed that liquid products were considerably better at providing a degree of protection to acid. Since this, we have internally started asking whether a probiotic can help to eradicate pathogens such as *C. difficile*, *Escherichia coli*, *Methicillin-resistant Staphylococcus aureus (MRSA)* and *Shigella sonai*? In all those cases, we found dosing with a probiotic eradicated infection.

Further to this, we have used the Simulator of the Human Intestinal Microbial Ecosystem (SHIME) (ProDigest, Belgium), an *in vitro* human gut modeling system, which simulates the human gut with an acid stage, two stages for the small intestine and two for the large intestine, which are filled with fecal slurry from human donors. The fecal sample is

suspended in a buffer allowing the bacteria to stabilize and colonize, providing a microbiota representative of those found in a human gut.

We used this system to analyze Symprove, a product containing three lactobacilli. Symprove was shown to be properly formulated such that it provides protection through stomach acid transit, allowing the lactic acid bacteria to arrive at the gut, where they proliferate and produce lactic acid. Since pathogens are not usually acid tolerant, one mechanism by which probiotics might exert an effect is by lowering the pH through the release of lactic acid, resulting in the eradication of pathogens. Furthermore, commensal bacteria in your gut interestingly, utilize lactic acid as a food substance which allows proliferation. As these proliferate, they produce short chain fatty acids implicated in many beneficial health effects, with the most important being butyrate. In our recent study, giving a probiotic over three weeks changed the proportion of the bacteria in the gut in such a way that the healthy bacteria increased, and the amount of butyrate being produced is increased.

Interestingly, our ongoing research is carrying out these 3-week probiotic treatments in studies of patient groups with disease states and finding complete changes of microbiota diversity, after probiotic treatment.

4. How do you think probiotics could benefit patients normally treated with antibiotics?

Commonly, patients with recurring *C. difficile* infections, for example, receive antibiotic treatments that are generally disastrous for commensal gut bacteria, only slightly affecting *C. difficile* and encourage resistance in *C. difficile.* In that case, the only treatment option is a fecal transplant. We are therefore trying to encourage the use of probiotics, such as Symprove, in patients with serious *C. difficile* infections and where antibiotic options have been exhausted. Our data suggests that the probiotic bacteria will arrive, lower the pH and create a toxic environment for the *C. difficile,* while providing nutrients for your commensal gut bacteria allowing recovery. There have been plenty of case examples where very ill patients have left hospital in a few days, using this approach, so we hope this will be adopted in the future. Thus, promoting probiotics as a therapy alongside antibiotic treatment could be important to avoid last resort options such as fecal transplants.

5. **What other important research is being carried out in the field and who could benefit? Are specific conditions being targeted?**

In my opinion, the key work involves attempting to identify key bacterial groups that are or are not related to certain disease states. With probiotics, I feel that they have fallen into disrepute as, for example, the ban in place on using the word on a product, in Europe. I believe this is the result of several badly formulated products which offer no actual patient benefit. There have also been a few small-scale clinical studies that have shown negative outcomes. I am personally not always convinced that this is down to the probiotics, but rather that the product hasn't delivered the bacteria successfully. There is a stigma that probiotics are never going to do any good but through research using these quite sophisticated *in vitro* tests, we are discovering that if the bacteria reach the site of action, they can have an effect. I hope new data on how probiotics influence microbiota will show the importance of using a properly formulated product and it leads to some joined up thinking. Patients with disease states should be studied to identify particular bacterial groups that either are present or absent, and then after administering a properly formulated probiotic, the influence it has on the gut microbiota should be evaluated. Our work also suggests that a formulated product may not need to contain the absent bacteria and instead, a product containing lactobacilli can allow commensal gut bacteria to recover by themselves.

An increasing number of links between the gut microbiota, probiotic therapy and disease states such as Parkinson's disease, multiple sclerosis, irritable bowel syndrome, inflammatory bowel disease, ulcerative colitis, Crohn's disease are being reported. There is therefore an increasing awareness of how bacteria affect some of these conditions and as that awareness becomes more widespread, people will then hopefully begin using probiotic therapy.

6. **What do you think are the key challenges facing the uptake of probiotic supplementation?**

The main challenge with probiotics in general is their classification as a food supplement. Under this category, they are not governed by regulatory authorities such as the Medicine and Healthcare products Regulatory Agency (UK) or the U.S Federal

Food and Drug Administration, which allows manufacturers to be less rigorous in the way they make their product and has led to many products on the market with no clarity in what they contain. With that said, I can't foresee an adverse effect from taking a probiotic. A badly performing product may have no effect at all but provided the product contains either lactobacilli, bifidobacteria or another bacterium that is generally accepted as not harmful or commensal, there should be no serious adverse effects. There is interesting data being published in the field but nevertheless, probiotics have had some bad press and there is a high degree of misinformation surrounding them. It would be helpful for the field to reset the public's perception and demonstrate that probiotics can have a benefit, if using properly formulated products. It would be helpful to introduce a degree of regulation to ensure products contain what they outline or to provide a degree of acid protection.

7. What do you think are the most important next steps for the field?

Initially, I think it is important to gain an understanding of bacterial groups, and in what ratio, should be natively present in healthy gut microbiomes. The next focus should be on whether disease states link to the degree of dysbiosis in the gut. The third question then becomes, is there a causative effect? Essentially, is dysbiosis a cause of the condition or a result of the condition? In other words, is it possible to identify bacterial species that do or do not cause disease? Once we know that, then it's possible to determine how a patient recovers from this degree of dysbiosis. The focus of the science should be on the causative relationship between bacteria and disease, and the focus from probiotic manufacturers should be in making sure they properly formulate products to efficiently deliver bacteria to the gut.