**Intro: [00:00:00]** Hello and welcome to the first episode of Otoscope. This podcast is produced by a group of PhD students at the London Institute of Medical Sciences, where we interview experts in different areas of science about their interest in bringing new, exciting science directly to your ears. This first episode was recorded before the COVID-19 pandemic, so about what seems now like 84 years ago. And we thought a good way to start this series was to talk about the thing that got us here in the first place, which is evolution, and how this relatively new field called epigenetics may help us understand evolution with a fresh perspective. For this interview, Radina Georgieva and Saul Moore met with Dr. Peter Sarkies in The Pod at White City Place to talk about his work on these things called transposable elements, his take on epigenetics, and the ethics of genome editing in humans.

**Radina: [00:01:17]** So today with us to talk about these things is Peter Sarkies. So we're going to discuss why and how the study of evolution and epigenetics can shed light on exciting, new mechanisms of gene regulation. So a bit about Peter, he completed both his PhD and postdoc in Cambridge. And in 2014, he formed his own group here at the LMS. His group studies epigenetic inheritance and evolution, and also since 2015, he’s been a junior research fellow at Imperial College London. And also, recently, he was one of the 26 life science researchers selected as EMBO Young Investigators, which is quite a thing. So welcome, Peter, we are delighted to have you here. And thanks, again, for agreeing to participate.

**Peter: [00:02:15]** Thank you for having me.

**Radina: [00:02:17]** All right. So before we move on to the science, basically, we wanted to ask you a bit about your PhD and postdoc, and how it was for you to make such a quick transition to leading your own group.

**Peter: [00:02:34]** Well, I was very lucky. I had an opportunity quite early on to start my own group. So I did my PhD at the LMB in Cambridge, which is quite a famous place. It was very daunting to be there initially because you'd sort of bump into Nobel laureates in the corridor, often quite old, and people who weren't actually in active science anymore, but they would still be sort of coming in and around the lab to sort of see, “Yeah, well, that’s--” Yeah. So that was also a very sort of competitive place, somewhat difficult to be a PhD student, actually, and at times, quite stressful, but a fantastic environment in which to do science. I was very lucky to start off there. And then I moved from there to do a postdoc also in Cambridge, and that was with Eric Miska. And Eric's lab is a complete contrast to the LMB, very exciting, very fun place, lots of parties. And so the contrast to the LMB, the lab I was in there was very much sort of focused and drilled down in one particular area. But Eric was really expansive, and anything to do with RNA, and he was interested.

**Radina: [00:04:06]** Yeah, I was going to ask, what sort of stuff were you doing when you were in his lab?

**Peter: [00:04:10]** Exactly. So for my PhD, I was working on how epigenetic marks, and maybe we'll talk about this in a bit more detail later, but how epigenetic marks are propagated when cells divide. So essentially, in the same way that a DNA sequence has to be copied exactly when a cell divides. In particular, I was interested in what happens when DNA gets damaged. Because DNA damage, I thought anyway, during my PhD, it might have a particular problem for replicating epigenetic marks faithfully through cell division. And actually, I showed that that is indeed the case, that DNA damage and also certain types of regions in the genome that are very difficult to replicate, under some conditions, they can actually give rise to a loss of epigenetic information. So in a way, that allows us to test the models for how epigenetic information is transmitted because we can use that as a system to study it by looking at what happens when that goes wrong. So that was quite interesting.

 **[00:05:24]** And all of that work really was done using a system that probably not many people have heard of, which is quite an interesting one, which is a chicken cell line called DT40. And the reason we used this was that this was before CRISPR and it was very difficult to knock out genes in cultured cell lines. But this particular cell line, for some reason, was very efficient for using homologous recombination in order to target and delete genes. So basically, we could delete genes that we were interested in, in this cell line, and then do experiments with that, which as I said, at the time, in other mammalian invertebrate cells was very, very difficult. So that's why we used that.

**Radina: [00:06:05]** That’s quite interesting.

**Peter: [00:06:06]** Yeah. So I mean, it used to be a bit of a joke in the lab that we work on chickens, but actually, we weren't really, we were using it as a model.

**Radina: [00:06:15]** There was a good reason for that.

**Peter: [00:06:16]** Exactly. Well, chickens are very cool, but--

**Radina: [00:06:19]** There’s nothing wrong with chickens.

**Peter: [00:06:20]** Nothing wrong with chickens. But yeah, we were using this system. So then, for my postdoc, I was told by someone very eminent at the LMB, when I was sort of thinking about what I would do, they said that you should never change both the question and the model system that you use when you switch from doing your PhD into doing your postdoc because he said, “Either you keep the theory, and then you learn new techniques, or you keep the system and you learn new theory, but doing both is something that is perhaps too much.” And so I thought, “That's very good advice,” so I ignored it completely. And so I went to Eric's lab, and there I was working, I suppose it was on epigenetics, but it was a very different type of epigenetics because we’re working with small RNA molecules. And it turns out actually that small RNA molecules are a type, in a way, of epigenetic information. And I also was working in a completely different system, because instead of using cells, I was using C. elegans, which is a nematode worm - very small, microscopic animal. And it was actually while I was in Eric's lab that I got interested in evolution. And it was really because I had to learn so much background in a very short space of time. I had to learn about this type of RNA molecule that Eric was interested in called piRNAs. So piRNAs are a type of small RNAs, I say, that are used to control transposable elements. So they're a silencing RNA.

**Radina: [00:08:07]** Is that the reason why it's considered an epigenetic mechanism? Just to make it clear to our listeners, basically, because it's another layer on top of the DNA sequence. Can you explain that a little bit?

**Peter: [00:08:17]** Right, exactly. I think that different people have different definitions of what epigenetics means. But for me, epigenetic means a type of information, which is not in the DNA sequence, which nevertheless can be inherited when cells divide, and crucially, that inheritance has to be independent of the initial signal that established it. So to give a concrete example, imagine that you had a signaling pathway, which led to a particular gene being switched off. That signaling pathway will be active in a cell, and then that would lead to the gene being switched off. But for that switching off to really be classified as epigenetic, you could take the signaling pathway away, but the gene would still be off.

**Radina: [00:09:09]** Oh, right. Yeah.

**Peter: [00:09:10]** And crucially, when the cell divided, that gene would stay off in all of its descendants.

**Radina: [00:09:16]** So it's a bit long term in its effect.

**Peter: [00:09:18]** So long term inheritance of a gene expression state.

**Radina: [00:09:22]** Okay.

**Peter: [00:09:23]** Now, piRNAs, initially, you might think, there's no reason to imagine them being epigenetic because what they are is they are small RNAs which silence particular elements in the genome. These elements are called transposable elements. We can talk about them a bit more, but they're essentially, you can imagine them being a bit similar to genes, but they are genes that cells want to keep off all the time because they're nasty. So piRNAs when they're present, silence these elements, but the crucial point, and this is actually what Eric's lab sort of in the first year when I was there really, I was involved in some work, which showed that surprisingly, the piRNAs can initiate an epigenetic type of silencing. And we could show that because you could make a sort of model gene, which was targeted by piRNA. And that model gene could then be turned off when the piRNAs were active. But then we could take a nematode worm with piRNAs, and we could cross it. So mate it with a nematode worm where the piRNA pathway was mutant, and the gene would stay off. So that shows that actually there’s an epigenetic silencing, which doesn't require the initial signal present at all. And actually, it turns out that that mechanistically is caused by another type of small RNA. So the piRNAs initiate the formation of another type of small RNA, which is known as a 22G siRNA. And these actually don't exist in mammals, but they do exist in worms. And it's actually quite an easy name to understand because these RNAs are 22 nucleotides long and the first one is always a G. So it's not particularly clever.

**Radina: [00:11:17]** That’s a good name.

**Peter: [00:11:18]** But these small RNAs, they have an epigenetic ability kind of built into them. Because once you start making them, they can then cause more to be made. So it's like a sort of positive feedback if you like. And that means that they can be maintained even when you get rid of the initiating signal…

**Radina: [00:11:38]** When the signal is not there anymore.

**Peter: [00:11:39]** When the signal is completely gone.

**Radina: [00:11:40]** Okay.

**Peter: [00:11:40]** So that means that the piRNA pathway actually turned out to be an epigenetic silencing pathway. And so it was something that I was very interested to work on when I was in Eric’s group.

**Radina: [00:11:53]** Okay. I think that's a very good example to keep in mind until the rest of the episode.

**Peter: [00:11:59]** Well, I think it is important because a lot of people would immediately when they think of epigenetics, think of something which is directly attached to the DNA, like, for example, DNA methylation, where a particular base in the DNA becomes chemically modified. But actually, epigenetics is a lot more broad than that. It can also mean molecules that are regulating genes, but which are able to propagate themselves, that could be epigenetic, just like DNA methylation can be epigenetic.

**Radina: [00:12:29]** And I guess we're still learning about those mechanisms.

**Peter: [00:12:33]** Exactly.

**Radina: [00:12:33]** It's quite a new field. Okay. And just one thing we were interested [in] was how was it when you started your own research group? Did you find it very challenging in the beginning? Was it the people management side that you found most challenging, or the research side or grants and all these things?

**Peter: [00:12:56]** Well, so I was very lucky in that at the LMS, when I started, I didn't have to write grants, because you start with a kind of package which allows you to do your science, so that was great. And initially, for the first sort of four or five months, I was sat in a room on my own, supposed to be doing research, but not having anyone in the lab because I had to wait until people came in. And being faced with all these decisions that I had to make, that I knew nothing about, and also, actually, to be honest, I'm pretty hopeless that even now. So things like for example, ordering consumables, kitting out the lab, that kind of thing. And sort of literally, like a blank sheet of paper. And I got a lot of help, actually, because, for example, there’s somebody who is… you may have come across Dipti Shah, who is sort of working with them, keeping all the equipment and so she was able to-- I remember, for example, going through into some strange room in the hospital where she told me that I could get a centrifuge for free following this path, and we still have it in the lab, actually. So that kind of thing allowed me to kind of kit the lab out because I didn't know where you would get these things.

**Radina: [00:14:29]** Yeah, maybe you have to smuggle a centrifuge in there.

**Peter: [00:14:32]** Exactly, because it's very expensive, and you've got a limited budget and you can't necessarily go over that. You get some startup money to kit it out but then you want to save as much as you can. So it was quite hard actually doing that sort of thing. But then once people started to come into the lab, it got a bit easier, and essentially for the first two or three years or so I basically worked in the lab as a postdoc, and because I didn't have to write grants it allowed me to do that and to do computational analysis. And so that allowed us to get some projects started. And once we had some results in, we started to get some interesting things. That's when I got a little bit more relaxed. But initially, I was really stressed all the time, because I was thinking, where are the results going to come from? Is any of this going to work? And also, if somebody was working for me, you know, the postdocs that we had starting in the lab, when things didn't work, I blame myself for that, because I had suggested that they did it.

**Radina: [00:15:33]** Of course. All the responsibility is on your shoulders.

**Peter: [00:15:34]** Yeah, exactly. So it gets a little bit easier as you sort of get used to it.

**Radina: [00:15:40]** Okay, well, it's always great that it's worked out.

**Peter: [00:15:42]** Yeah, in the end, I was just lucky, and I got a lot of help. And it was good to be supported and surrounded by people who were in a similar position to me. And that kind of made it a bit easier.

**Radina: [00:15:52]** Yeah, the environment is nice.

**Peter: [00:15:53]** Exactly.

**Saul: [00:15:57]** Great. So we've read your blog, and you've previously compared evolution to running on a treadmill. Could you just explain that a little bit?

**Peter: [00:16:09]** Well, so the treadmill analogy refers to a specific type of evolution, which is where you have a host and a parasite. So in the example that I'm interested in, the parasite is actually something that we mentioned a bit earlier, which is a transposable element. So a transposable element, you can think of it as being a bit like a virus which cannot spread between cells. So it's essentially in a way, it's kind of the ultimate parasite, in my view, because it's extremely lazy. That's all parasites want to be - lazy. They want to do as little as possible. They don't want to make stuff, they want to use everything inside the cell. And this is like the ultimate in that because it doesn't actually make anything except for a copy of itself. And then that copy of itself somehow can be reintegrated into the genome. And it uses the host RNA polymerase, it doesn't really need anything else. It uses the host DNA repair mechanism. The only thing that it might encode, for example, some transposons encode a molecule called a reverse transcriptase, which would copy an RNA that's made from the gene into a DNA. And then that DNA gets back into the genome and that's how it gets copied.

**Saul: [00:17:40]** I see.

**Peter: [00:17:41]** And there are other types which act solely at the DNA level. So they make a gene called a transposase, which is a molecule which acts on DNA, and cuts the transposon out of the DNA, and then moves it somewhere else. And that can allow it to copy itself through the genome if it's done in a particular way. So these are basically as I say, they're lazy viruses. Now, what hosts have to do is they don't really want huge levels of this, because it's a burden, it's a waste of resources, you have to keep making these things. And also, every time they copy themselves as well, there's a risk because they could put themselves into a gene and that might disrupt the gene. And also, the very act of making damage to the DNA could potentially be nasty for the cell. So they have to evolve mechanisms in order to silence them. But at the same time, the transposable element is wriggling to try and get away from these.

**Saul: [00:18:46]** I see. It's like an arms race.

**Peter: [00:18:48]** Exactly. It's an arms race, so it is a bit like running on a treadmill because you are always having to innovate as both the parasite and the host, but you never really get anywhere at the end of the day. So it's like sort of wasting energy and in a way a treadmill, although you might feel good after running on it, it's not as if you've really achieved very much in terms of your distance.

**Radina: [00:19:06]** You didn’t get anywhere.

**Saul: [00:19:07]** You haven’t got anywhere.

**Peter: [00:19:08]** You're still in the same place unless there's something very seriously wrong with the treadmill. So yeah, so that's I suppose, where that analogy comes from.

**Saul: [00:19:18]** Right. Is there any particular interesting examples of that that you've been working on?

**Peter: [00:19:28]** The problem is, it’s quite difficult. It's a theory, but when you look at a particular time, you don't know what stage in the cycle you're looking at, you don't know whether the transposon is winning or whether the host is winning. So I suppose that you can infer it because if you look at transposable elements in the genome, there are a lot of transposable elements that are kind of dead. In other words, they are actually not able to spread anymore. And what you can think of is that this is one way it's been silenced. And because it's been silenced, it hasn't been able to copy itself. And then that means that eventually over time, it will just degrade. So that's in a way, why we can sort of infer that over time, transposable elements get shut down, and less active. So initially, they'd be silenced solely at the epigenetic level, if you like, but then over time, they accumulate mutations, which make them not able to work anymore.

**Saul: [00:20:35]** That’s very interesting.

**Peter: [00:20:35]** So they're kind of the skeletons, if you like, of an active transposable element. So from that, we can infer that evolution is kind of working all the time. But to follow it over real-time is rather difficult because these things take a very long time. And any species that we're looking at right now is kind of at the endpoint of that process and we don't know what point along the line it is, if that makes sense.

**Saul: [00:21:02]** Right, yes. Interesting.

**Radina: [00:21:06]** So going back to the DNA sequence, thinking about DNA and the various elements present in it, we think that your research on transposable elements is really interesting. But sort of to give people an overview of what those things are, so we know that they're very prevalent in our genomes, correct me if I'm wrong, like maybe 40%?

**Peter: [00:21:31]** Around that.

**Radina: [00:21:32]** Quite a substantial proportion. So can you tell us a bit more about how they insert themselves in our genomes, so to speak, in the first place? And yeah, a bit more on how they propagate themselves. And also, you also mentioned that they can have sort of deleterious effects when they disrupt important genes. So I was also curious, why are they there, then? So maybe there is an advantage of actually keeping them? Maybe they can do some sort of useful stuff as well?

**Peter: [00:22:06]** Right. So I think it's lots of very interesting points there. So let's start with where transposable elements come from in the first place. I think that largely, we don't really know about that. To push it back one level is quite straightforward, you can say that a transposon can very rarely cross between different species. And so a transposon in one species can go into another species. And then usually, that will lead to that particular transposon copying itself quite a lot very quickly because there won't be any control mechanisms in place in that naive species if you like. So that's one way, but of course, that doesn't tell you ultimately where the transposable element comes from, because it's just saying it comes from a species previously. So I suppose that an attractive hypothesis, which might explain where at least some transposable elements come from, is they are kind of broken viruses. Actually, broken is the wrong word. They're kind of, as I said before, lazy viruses. And so in a way, they're actually more clever than a virus.

**Radina: [00:23:11]** So it's an actual virus that has infected the host, and then it sort of remained.

**Peter: [00:23:17]** It infected the host, but then lost the ability to spread. So it no longer makes an outer coat, no longer makes the kind of machinery that allows it to get out. And so that is one very simple way whereby a transposable element could essentially evolve if you like. So we know that there are so-called endogenous retroviral particles which are at different points along this time scale. So some of them even still make capsid protein, for example. And some of them can very occasionally spread between cells but at low rates. So that gives you some evidence to think that quite a lot might have arisen in that way. But I think we don't know for certain about all of the transposable elements. Some of them may have actually evolved specifically as transposable elements because if you think about it, that's not that difficult to do. All you would need to do is to evolve a protein which could recognize itself in the DNA and then cut that DNA out. And then that can then jump and insert somewhere else in the DNA and that could actually sort of evolve almost de novo. It wouldn't have to come--

**Radina: [00:24:29]** From the actual genome.

**Peter: [00:24:30]** You could imagine it happening. Yeah.

**Radina: [00:24:32]** Yeah. So what would be the incentive for this to happen? We don't know, obviously.

**Peter: [00:24:37]** We don't know, but my argument would be that the strong null hypothesis if you like, is that transpose elements exist for the sake of transposable elements.

**Radina: [00:24:49]** Selfish.

**Peter: [00:24:50]** They are selfish. So *The Selfish Gene*, which is a very famous book, written by Richard Dawkins, imagines the gene not really caring too much about the organism, what's important for the gene is-- So the gene only cares in a way about the organism because the organism is a vehicle for the gene to copy itself. So the transposable element is kind of the ultimate example of a selfish gene. And all it needs to do is just lay sufficiently low that it doesn't cause enough harm to kill the cell and then it can kind of hitch a free ride. And just because it can do that, it will then remain and propagate.

 **[00:25:35]** Now, an interesting question which you raise is whether this is actually something that-- Because you imagine from a sort of point of view of efficiency, the cells would like to shut down and transposable elements completely. So why are transposable elements still active?

**Saul: [00:25:54]** Yes, and what are the advantages of keeping them?

**Peter: [00:25:55]** Right, but do there need to be any advantages? Because if we talked, as we remember about this treadmill situation, we're looking at a situation which is not necessarily the endpoint of any situation. There's a constant battle going on between the host and the transposable element. So the transposable element could be, in a way, at least partially winning some of that battle, and that means it could still be spreading. So it might be that if you asked… I’m going to be hopelessly anthropomorphic, if you asked the cell, “Do you want the transposable element?” it would say, “Absolutely not. Get rid of them all.” But it's not able to do that, because the transposable element is fighting to keep itself alive. So that's one possibility. And I think that that is still a very strong null hypothesis. And my guess would be that that probably explains the majority of the prevalence of transposable elements.

 **[00:26:51]** However, there's another possibility, which is that there is some benefit to transposable elements. And therefore, there's a sweet spot whereby you can silence transposable elements just about enough to make sure that they're still doing good, but what cell wants, but they're not doing sufficient harm to be really deleterious.

**Radina: [00:27:18]** Yeah. Super interesting.

**Peter: [00:27:20]** That's a very interesting idea. And there is some evidence that, in some cases, that there might be some benefits to transposable elements. So one idea that's quite simple, but it's controversial and complicated, in a way, but the idea itself is simple, but when you look into the details of it, it's a bit complicated. But if you imagine that organisms have to adapt to their environment, a transposable element could be a way in which you could get quite a large change in the genome quite quickly. For example, transposable elements, when they copy themselves could lead to the copying of another gene nearby, and that might be beneficial in some way. Alternatively, transposable elements normally have a promoter which makes the transposable element be made. So it allows the transposable element to produce RNA and that's what obviously genes do as well. So if the transposable element were to insert itself in such a way that its promoter actually drove one of the host’s genes, then that could change the pattern of expression of the host’s genes, and that might be somehow beneficial. So maybe, because these things could happen, we keep the transposable elements there because they provide a reservoir of possibility for evolutionary change.

**Radina: [00:28:51]** Exactly. And I guess if an element that is sort of useful for regulating genes needs to spread across the genome, then sort of hitchhiking on a transposon would be an efficient way to do it. But yeah, that’s just a hypothesis.

**Peter: [00:29:06]** Yeah, that's also interesting. So what happens is, if you look across genomes that exist now, you find quite a few of these examples whereby elements from transposable elements, parts of transposable elements are actually used in ways that contribute to how the host cell works. Now, that is interesting, but it doesn't tell you really that the reason that we still have transposable elements is because cells want that to happen. Because it could be that the vast majority of things that happen as a result of transposable elements are very negative, but all of those things will be eliminated by natural selection. And then the few things that remain are the very rare cases where it has been to some advantage. So again, I would come back to this idea of asking the cell what it wants. And the cell might say, “Well, even though yes, okay, very rarely it gives rise to this, still, I would be better off without them altogether.” And we don't know whether that's really the case or not. And I think that there are some clever experiments that people are doing now I know about with long term evolution experiments, which might get at actually asking those questions in real-time.

**Radina: [00:30:37]** All right, that would be interesting to follow up on. All right, so now let's switch a little bit to the other main topic which is a bit more about epigenetics. So, Peter, can you explain a bit more about the main epigenetic mechanisms? You've touched upon small RNAs a little bit, but sort of what are the main mechanisms? And do all of the marks that are imprinted by those mechanisms get passed on to the next generation? Or maybe only some of them? And how does it all work?

**Peter: [00:31:16]** Yeah. So I think that the way that I like to think about it is that an epigenetic mechanism is something where there exists the possibility for it to template its own formation after the cell has divided, but it doesn't necessarily mean that that particular pathway is always epigenetic under all circumstances. In other words, it doesn't necessarily mean that it is always acting in that way, but it has the potential to do it. So let's consider DNA methylation. That's in some ways, the simplest way of imagining it because DNA methylation, you add a methyl group to a cytosine. And then that methyl group can be propagated when the cell divides independently of the initial addition of the methylation because there's an enzyme that exists in the cell that can recognize DNA that is methylated on one strand and not methylated on the other, which would happen when the DNA is replicated, and then copy, if you like, the methylation from one strand on to the other to allow it to be propagated.

**Radina: [00:32:35]** So does that always happen, or it happens in specific regions of the genome?

**Peter: [00:32:38]** Well, so I think it depends probably on the organism, but largely in a mammalian cell, yes, that would happen. Because the enzyme in question is called DNA Methyltransferase 1 (DNMT1), and this, which can recognize the hemimethylated, half methylated sequence is active all of the time when cells divide. So that means that by default, the methylation will be copied. But there are some organisms that don't have that enzyme, they have only enzymes that introduce methylation onto unmodified species that exist.

**Radina: [00:33:22]** Sort of de novo.

**Peter: [00:33:23]** Yeah, exactly de novo if you like. And, in those organisms, you would imagine that DNA methylation cannot be epigenetic in the same way because of the enzyme.

**Radina: [00:33:32]** All right. So the same mark can be epigenetic in some organisms or not. That’s pretty cool.

**Peter: [00:33:36]** Exactly. So I think that that also applies in a much more widespread sense to histone modifications. So histone modifications can be inherited. And the way that they can be inherited is actually conceptually similar to the way that DNA methylation is inherited. You can imagine a protein that recognizes a histone modification, and then it will introduce it to other histones in the vicinity. And so that means when you divide the cell, the old histones are mixed with new histones. And then the old histone can then template, if you like, the modifications of the new histones, so the new histones mature, if you like, into the histones that are characterizing the surrounding region. So that would allow a histone modification to be propagated. And that definitely happens under some circumstances, but it also definitely doesn't happen on all circumstances.

**Radina: [00:34:33]** It’s not so black and white. Yeah.

**Peter: [00:34:34]** And it certainly doesn't happen with all histone modifications because they don't all have the machinery present to recognize themselves and then to template their own sort of copying, if you like. And then the RNA is a third opportunity to have this sort of epigenetic change. And there as I said before, the crucial point is again, these trends 22G RNAs, which silence genes downstream of piRNAs, but they have the ability to template their own formation. So once they've been made, they can make more of themselves and that can allow them to keep their levels high when cells divide. So otherwise, they would just get diluted all the time.

 **[00:35:23]** So there are even examples of epigenetic changes, which I would call epigenetic, which involve solely proteins. So an example of this would be a prion. So a prion in a way, is an epigenetic process because it's information, which is outside of the DNA sequence, and it's encoded in the shape of the prion protein. And the crucial point is that prion proteins can exist in two different shapes and there's one shape that can bind to the other shape and cause it to change into itself.

**Radina: [00:36:04]** Wow. That’s super **[strange 36:06]**

**Peter: [00:36:07]** And then that means that you only need a very small number of the, let's call them shape A, which is the prion shape. And shape B, which has exactly the same amino acid sequence can be much more abundant, but you just need a few of shape A, to convert shape Bs into shape A. And then because more shape As can then be made, it's a chain reaction, if you like, which can lead to all of the shape Bs being converted into shape As.

**Radina: [00:36:41]** But do they affect the expression of genes, of how genes work?

**Peter: [00:36:43]** And for example, you could imagine that a prion could be a transcription factor, which could **[inaudible 36:50]** genes--

**Radina: [00:36:50]** Oh, so it's more like a type of protein.

**Peter: [00:36:55]** So in shape A, it could turn on genes, and in shape B, maybe it can't. And then that would mean that the cells would behave very differently depending on whether they had all shape Bs or all shape As. And so that would be a perfectly valid way for epigenetic change to occur. So what I would say is that I would not be restrictive about calling different mechanisms epigenetic or not; many can be epigenetic. But the crucial point is that the process of epigenetics for me requires evidence that you can get propagation of the mark independently of the thing that introduces it, and that can be inherited through the division of the cell.

**Radina: [00:37:48]** Yeah. So it's more about the way rather than the specific mechanism.

**Peter: [00:37:39]** The way rather than the specific mechanism.

**Radina: [00:37:52]** All right.

**Saul: [00:37:52]** Right. Just as a follow up to that, so what could happen if epigenetic mechanism malfunctions for the next generation?

**Peter: [00:38:04]** Well, you could imagine a situation where an epigenetic pathway was too exuberant. And let's say it's a histone modification, and the histone modification is silencing, and it silences a particular gene. So if you imagine that that has a really, really active way of propagating itself, it could spread and spread and spread, and it could just silence the whole genome. So actually, we think that there are particular elements inside the genome called insulators, which can prevent this from happening. They confine the region of epigenetic modifications to a particular region of the genome, and they stop it from spreading outside of that. And that's very important because it could poison the regions around it. So that's, I suppose an example where you could imagine an epigenetic pathway malfunctioning. I don't actually know whether that happens particularly commonly, but I could imagine that. Yeah.

**Saul: [00:39:07]** Very interesting.

**Radina: [00:39:10]** All right. Another interesting question is about the recent sort of surge in those biotech startups that claim to be able to suggest lifestyle changes to a particular person by sort of measuring or looking at their genome-wide methylation profiles. I think most of them use this technique called bisulphite sequencing. So they look at your profile, and that's based on a concept called…

**Saul: [00:39:46]** The epigenetic clock.

**Radina: [00:39:47]** Epigenetic clock. Exactly. Saul, do you want to brief us into what's the epigenetic clock?

**Saul: [00:39:55]** It suggests that DNA methylation in certain genomic sites could be correlated with age. Based on this, one’s methylation levels could be compared to the expected norm for their age. What are your thoughts on that?

**Peter: [00:40:17]** So I’d say the science behind the epigenetic clock is very sound. There's lots of evidence that there are sites where the methylation changes in a predictable way over age. And there's also some evidence, which suggests that it's better correlated with your biological age than with your chronological age. In other words, if you are ill in some way, then you accumulate some of these changes more rapidly than you would expect, given the rate at which the changes arise in most people.

**Saul: [00:41:02]** I see.

**Peter: [00:41:03]** Now, as I say, I think the science behind it is sound, but what is probably less sound is the idea of extrapolating that into a kind of test that is used to predict outcomes in people. So I think that that is likely to be more challenging. So I think that what I would say is that getting it done, and having your epigenetic clock measured might be of interest to you but I don't think it should be something that you should really take seriously in terms of any kind of predictive outcome for yourself. And I think that a lot more evidence will be needed before it could actually be used to say, “You are likely to get some sort of disease in the next few years,” or something. I think that that will be my take on it.

 **[00:42:08]** The problem is you see that I think, as a bit of fun, it's great. It's like 23andMe. I did 23andMe.

**Radina: [00:42:22]** Yeah, same.

**Peter: [00:42:23]** It's quite interesting, it’s quite fun. But I think that the problem is that it's easy for me to say that because I know what it is not. And I know that if I see something that tells me that I have a five percent elevated risk of a blood clot, I know how that is calculated. It’s calculated on the basis of thousands and thousands of people. It doesn't really mean anything causative for me. It doesn't necessarily mean that I actually will have a blood clot, etc. I think the problem is that the education level is probably not high enough that we can reliably say that people will interpret it in that kind of way.

 **[00:42:58]** So I had actually an email from somebody in the general public, just after we published our paper on the evolution of DNA methylation. See, they'd found this paper, or I think it may even have been the Imperial blog that I wrote about it afterwards, and they thought, “Ah, this is somebody who knows about methylation.” They'd had one of these tests done and they said, “I was told that I had hypomethylation. And hypomethylation when I looked it up on the web is something that happens in cancer cells. So do I have cancer?” And obviously, I didn’t really know how to respond to that. But the answer is if you think you might have cancer then I would go to a doctor rather than trying to ask me. But I suppose that there is a risk associated with these kinds of tests in that people might misinterpret or rather not be sufficiently able to understand the limitations to know that-- Because probably what happened was that actually, the reason that it was grossly hypomethylated is because in that case, the test went wrong in some way or he gave them a sample of the wrong thing or something like that. It’s unlikely that it was because he had leukemia or something, but he didn't really realize that. Yeah.

**Radina: [00:44:23]** Poor them, they must have been totally freaked out **[inaudible 44:26]** hypomethylation.

**Peter: [00:44:25]** Well, yes. Exactly. I think it's a very complicated thing. I don't really know enough about the legal or ethical considerations that are going on to make much more of a comment on it. But what worries me is that I think that the companies rush these things into general use, but I think the reason that they do that is because they are hoping that people will take them up and they will not tick various boxes, saying, “I don't want my data to be shared, used, etc.” And that will allow the company to get a lot more information from the general public, where they can then use correlative data to then improve the output of what they get. And actually, you can see that with 23andMe, because I looked on it first and it had some estimates of various ancestry composition of mine, which actually wasn't particularly in line with what I had known from my family history. And then, actually, a few months later, I got an email from 23andMe, saying, “Log in to your 23andMe, it's changed. There's more information changed.” I looked at it. And actually, now, there's a lot more information about it and it reflects much better.

**Radina: [00:45:45]** So they've improved their database.

**Peter: [00:45:46]** So their database has improved in the meantime because other people had-- And I hadn't given them any of… so it wasn't like they were using my own family history, because I hadn't actually given that information. But I think that the risk is that, for me, I'm a bit worried about what that information… how much rights we have over that if we give it to 23andMe. Because they're using it for themselves, but in the future, they might use it--

**Radina: [00:46:11]** Yeah, you don't know how they're going to use it after they've given you your results.

**Peter: [00:46:14]** And I guess that this epigenetic clock thing, it's based on sound science, as I say that there are sites in the genome that change in a predictable way with age. But I think that what they're hoping is that people will go onto it, and then they'll fill in all their medical records. And then that will allow them actually to get much more information about what the epigenetic clock might actually mean, which might be good but on the other hand, there's a risk associated with that, which is that people's data is being used in a way that they might not be very happy with if they really knew about it.

**Radina: [00:46:43]** Yeah, it’s like they are monopolizing.

**Saul: [00:46:44]** Or perhaps not yet being used in that way but it could be in the future.

**Peter: [00:46:46]** Right, because it’s not going to be deleted, it's going to be stored somewhere.

**Saul: [00:46:51]** Indeed.

**Peter: [00:46:52]** Maybe even after they're dead.

**Saul: [00:46:54]** Yeah.

**Peter: [00:46:55]** Anyway, that's perhaps a bit of a digression but yeah.

**Saul: [00:46:59]** Controversial topic…

**Radina: [00:47:00]** Which leads us--

**Saul: [00:47:01]** …Which leads us perfectly on to our-- Yes. So we'd now like to bring even more controversy into our conversation as I bring you to our Otoclave segment. So in this segment, we'd like to bring some hot and pressing issues to discussion, in order to clear up some false information and to discuss controversies. Earlier this year at the Second International Summit on Human Genome Editing in Hong Kong, Dr. Jiankui He presented data on how he successfully edited the genome of two embryos, two twins born in February 2019. By removing the gene CCR5, he developed a procedure that he claimed could immunize an individual against HIV and AIDS virus. This has inevitably led to enormous ethical or moral debate among the scientific community. By altering the genome of human embryos, he has opened the door to a scientific era in which scientists could potentially rewrite the genetic pool of future generations of our species and alter our own evolutionary trajectories. What do you think about the emergence of designer babies and a possible Gattaca type dystopian future?

**Peter: [00:48:14]** Yeah, so this is actually a very interesting area. So I mean, I love Gattaca. It's one of my favourite films.

**Radina: [00:48:22]** Saul actually hasn't seen it. Let me disclose.

**Peter: [00:48:23]** You haven’t seen it? Well, you better watch it. It's a good film. But I don’t know, it's this is such a difficult area because, on the one hand, I think you could argue that editing the genome of a human is something that inevitably will happen. And, in a way, I mean, it may already have happened in this particular case, although I think that it hasn't been peer reviewed, and so on. I'm not sure whether it's been released in a conventional way. So it seems to me that any kind of technique of this kind, there's going to be so much interest in it that somebody will do it at some point. And maybe it is better for it to be done in a way that is regulated. In other words, for a country which has a good regulatory system to actually open it up and say, “We're going to allow this in a limited way,” because then you would stop practitioners doing things which are really very unsafe, potentially.

 **[00:49:36]** So an example of where this could relate, I mean, is the topic of abortion. I think that a lot of people even who are not happy about abortion, accept that it's better to have it in a regulated system, rather than have it in a situation where people will go underground and then do it at great risk to themselves. So you could argue that a regulated system of modification of human embryos may be something that we should be working towards, but I don't think we're at a point where we could do that, for many reasons. I think that the crucial thing that we do not understand is, we don't really understand epistasis enough. So take a very simple genetic disease like cystic fibrosis. So there are a few different alleles of cystic fibrosis but they result with quite high penetrance in a disease where people cannot clear mucus properly from their lungs and from other places in the body. And it gives rise to a lot of different phenotypes, which usually lead to an early death.

 **[00:51:01]** So you could imagine that that would be a good example where you could go into the embryo, and you could edit the embryo to make sure that it doesn't have that gene. But if you look, even within people who have cystic fibrosis, there's a massive range, even within the same allele, of what happens to them. And a lot of that I would suspect is actually not due to differences in environment, it’s actually difference in their genome. Because human genomes are very complex and very diverse and they will have other alleles in there which will affect how that cystic fibrosis disease manifests itself, even if they have the same allele for cystic fibrosis. So I think that the risk with CRISPR editing, even if you take something that's a very simple case, you do it in a particular background, and it might give rise to a lot of things that you're not expecting. And I think we just don't know enough, as I say, about how unpredictable some genetic interactions can be, to be able to be very safe about doing anything, even in very straightforward cases like that.

 **[00:52:02]** Certainly, when we come to thinking of things like modifying eye color and desirable traits of various kinds like that, or what happens in Gattaca, there, I think we're so far away from understanding anything about how those traits - polygenic traits - are determined and how they interact with other traits, that it would be very risky to introduce it. So my thought would be that maybe it might be a good idea for there to be a body which is actually set up with the stated aim of doing corrective gene therapy on embryos. And it reviews all the evidence and it's funded properly and it makes progress towards that. And maybe it will actually give the possibility of curing some or correcting some genetic diseases. But I think that that's my concern. I mean, I'm not saying that that is definitely a good thing but in some ways, it seems to me that that might be better than various crackpot clinics in China, but it could also happen in the US I could imagine, [that] would just be able to set themselves up and offer this as a service to people and manipulate people into doing things that will lead to production of babies who might be very unhealthy as a result of interactions, as I say, that we don't understand.

**Radina: [00:53:33]** Definitely, and since science is already getting there anyway, it seems like a --

**Peter: [00:53:36]** That's the problem. It's sort of like a bit like this, I mean, it's not a very nice phrase, but the cat being out of the bag. Once these techniques exist, then it's very difficult to sort of pretend that they don't exist.

**Radina: [00:53:49]** Exactly. I think so too.

**Peter: [00:53:50]** And that would be my feeling about it. But I think that it's got to be handled very carefully. And I'm not volunteering to do but this but that would be my take on it.

**Saul: [00:54:01]** Right. Absolutely. Now we move into the last stage of our podcast, with our Publication of the Month.

**Radina: [00:54:13]** All right. Hi, Vass.

**Vassili: [00:54:15]** Hello.

**Radina: [00:54:16]** So just to introduce Vass’s paper, it's entitled “Cytosine methylation affects the mutability of neighbouring nucleotides in human, Arabidopsis, and rice.” And just to translate this to rather simpler language, so from what I understand, Vass was trying to understand the shorter and longer-term effects of methylation on genome fragility and evolution in general. But yeah, Vass, maybe you can summarize your paper for us and the main findings.

**Vassili: [00:54:46]** Sure. So what the paper, or my work, was looking at was at this type of epigenetics called DNA methylation that Peter talked about earlier. Specifically, methylation, where in genetic code you have these four letters the A, T, C, and G, and the C, the cytosine, can get modified by adding this methyl chemical group to it. And now this changes the DNA a little bit for this purpose of epigenetics of regulation and stuff. So physically, it makes the DNA more rigid, it makes it harder to separate. And from an epigenetics standpoint, it’s normally used to silence parts of the genome, or for various other things, whichever is more species-specific. And for quite a long time, people have known that when you methylate the cytosine, the cytosine itself mutates more frequently. So in evolution, there are two main driving forces, you have this mutation that consistently happens, that Peter said before how you can kind of date an organism based on how many changes, how many mutations you get over time, and then you have selection on these changes. So I was looking specifically at these changes.

 **[00:55:55]** So the cytosine itself, when it's methylated, can mutate more frequently due to two reasons; A, chemically, it just mutates more often. And B, it’s repaired less efficiently once it does mutate. But no one has really looked at how this methylation affects the mutation rate nearby. And you can kind of envisage that since it does change how the DNA itself physically behaves, maybe there are more widespread effects. So what our work did was we used rare polymorphisms, rare mutations, and rare changes in populations of humans, rice, and Arabidopsis, which is a watercress, to try and infer any correlation between the methylation and the changes nearby. And we find that in human, it actually reduces the incidence of mutations, whereas, in plants, it seems to increase it.

**Radina: [00:56:48]** All right, so a logical follow-up question on that would be why do you think you are observing these opposite effects? Or maybe is there a simple explanation for that, or it's rather complicated? Can you elaborate?

**Vassili: [00:56:59]** So it becomes quite hard to look into exactly what's driving it because to correlate these things, you need quite a large sample size, and mutations are generally quite rare over a long period of time because it's a quite slow effect. But what we think it might be due to is the differences in how these different species use the methylation as a mark. So different species might use different epigenetic ways of regulating themselves. So in humans, around 80% of these CpGs - these Cs **[followed by 57:33]** Gs - are methylated, whereas, in plants, it's much lower. So in our Arabidopsis, it's only 23%. So you can kind of envisage that in human, methylation is like the default state of the cytosine in this context, whereas in plants, it's a bit more specifically designated. Like either targeted to the transposable elements that Peter mentioned before, or for whatever reason. So maybe, because it's more widespread and human, you don't want it to be mutating as much because it might be covering a lot of genes that you're trying to keep preserved from one generation to the next and so on, so forth, whereas in plants, because it's a bit more specifically placed, like since--

**Radina: [00:58:19]** So remind me, in humans, it was like the methylated cytosine was surrounded by bases that would mutate more or less frequently?

**Vassili: [00:58:28]** In humans, less frequently,

**Radina: [00:58:29]** Less frequently. Okay.

**Vassili: [00:58:30]** Yeah, the adjacent bases were particularly--

**Radina: [00:58:31]** So you want kind of want to restrict that effect because these methylated Cs occur so often in the genome that maybe it will make sense to not have that many mutations around.

**Vassili: [00:58:41]** Yeah, exactly.

**Radina: [00:58:42]** It's hard to say, I guess.

**Vassili: [00:58:43]** Yeah, exactly. And you could also maybe imagine that in plants, it’s going back to this arms race or this “on the treadmill” evolution that Peter was mentioning, that if you're more specific with where you put the methylation in plants, i.e., only to silence transposable elements, then you’re kind of wanting them to mutate because you're trying to disable them. Whereas in humans, if you’re methylating more of a genome, you're also hitting genes, which are important, and you don't want those to mutate. So you maybe have evolved a better repair mechanism in humans, whereas in plants, they’re like, “We don't care. Let's break those bits,” you know?

**Radina: [00:59:17]** Okay, so that's a potential explanation of this.

**Vassili: [00:59:19]** That's a potential explanation, but to quantitatively test that becomes a lot more hard.

**Radina: [00:59:24]** Yeah, I guess so.

**Saul: [00:59:25]** Could I ask a question?

**Radina: [00:59:26]** Yeah, sure.

**Saul: [00:59:27]** So did you look in invertebrates in which there's methylation? So in honeybees, for example, there's very sparse methylation, which is only in genes that are expressed at quite high levels. So I wonder whether do you see the similar or different effects in those species or is it just in humans and Arabidopsis that you looked?

**Vassili: [00:59:51]** So we really wanted to look at invertebrates, and honeybee was actually one we did try to look at, but the issue was that we not only need very good methylation maps of the genome at the base-specific level, but we also need to have a large enough population sequenced for the mutations themselves. And our data set for honeybee wasn't powerful enough, so the error bars were just too large to say anything, which was unfortunate because that would have been a really interesting comparison to make. But I guess as we sequence more and more different organisms, that question can only be answered in a better way, right?

**Saul: [01:00:27]** Yes. And did you look in other species of mammal?

**Vassili: [01:00:32]** We looked at mouse, but again, a lot of the sequenced mutations for mice are done in lab strains and they're not a very good representation of what happens in the wild. So that also becomes a bit different. But in mice, just from our preliminary data, it looked a lot more like human, so it seemed to reduce mutation rates nearby. So yeah.

**Radina: [01:00:55]** Okay, congrats on your preprint.

**Vassili: [01:00:57]** Thank you.

**Saul: [01:00:59]** So that's it for today. We would like to thank our guests, Peter Sarkies, and Vass, and the White City Place for allowing us to record this session in The Pod. As related to all things inherited, if you like this episode, please pass this podcast on to your children and grandchildren, and great-grandchildren. See you next month.