Speaker 1:

Welcome to the Eye on the Cure Podcast, the podcast about winning the fight against retinal disease from the Foundation Fighting Blindness.

Ben Shaberman:

Welcome everyone to the Eye on the Cure Podcast. I am your host Ben Shaberman with the Foundation Fighting Blindness and I'm very pleased today to have as my guest John Flannery, Ph.D. at UC Berkeley, where he's a professor of neurobiology in the Department of Molecular and Cell Biology and he's also associate director at the Helen Wills Neuroscience Institute. And he's also a scientific co-founder, at Vedere which is a gene therapy company that's in the foundations RD fund portfolio. And the RD fund, for those of you that don't know, is our venture philanthropy fund for helping move companies into clinical trials. And I'd be remiss if I didn't add that John has been a member of the Foundation's Scientific Advisory Board for many years, for decades, and he's received funding from the foundation over the years for a lot of different great research efforts including gene therapy, some excellent work in gene therapy and optogenetics, which is going to be the focus of our discussion today. And John, welcome to the Eye on the Cure. It's great to have you.

John Flannery:

Thanks, Ben.

Ben Shaberman:

So I've known you for many years and I've heard you talk, I've read your papers, but I don't know how you got started in this field or just in science in general. Can you go back to when... I don't know, perhaps you were a little guy at five years old and you were playing with test tubes. Can you tell us when you realized that you had a penchant for science and decided to get into research and then what got you into focusing on the retina?

John Flannery:

Yeah, I think this is a Woody Allen joke, right? I started out as a child, my father... I come from a very blue-collar family. My father always thought, who was an engineer when he started his career, always thought that we should be able to fix everything in the house. So I would go with him to fix the car, the washing machine, the dishwasher, everything. So I'm used to taking things apart and hopefully putting them back together again. Actually, that's the only thing that students let me do in the lab anymore. So yeah, I mean, I've always had an interest in discovering how things work by disassembling them. And so when I was an undergrad, I volunteered to work in Steven Fisher's lab who was a retina researcher when he came to UC Santa Barbara, where I was an undergrad. So I volunteered because I thought it would be nice to do stuff with my hands and more than listen to lecture. So I've been working on retina since I was undergrad. Actually, I don't think I... Actually, I haven't ever worked on anything other than in retina.

Ben Shaberman:

That's really interesting. So while you've been in research for all these years, you've had a lab, your own lab for many years. And can you give us just sort of a sense of what happens in a lab and what it's like to run a lab?

John Flannery:

Yeah. I mean, different faculty members have different styles. One of the faculty members upstairs who has a great sense of humor says what he does is he raises money, buys Ferraris, and throws the car keys out the office window. At Berkeley, the student, the graduate students, and the postdoctoral fellows are generally incredibly excellent at what they do. Yeah and to some extent, my job is to give them the resources for them to be creative and excel. They need good equipment supplies, animal models. So a lot of what professors do is try to keep the grad students focused and I don't think I ever have to tell them to work harder and to give them the resources to do excellent work and creative work, so.

Ben Shaberman:

That's great. And I presume you're a fun mentor but a very inspiring and educational person. I presume you do a lot of mentoring in your role.

John Flannery:

Well, I don't like to micromanage. My office is actually right in the lab. So my style is sort of... I try to keep the door to my office open and not look too intimidating. So some of the students like to talk to me when things are working. Other ones only want to talk to me when it's not working. Some of them if... They all have their own style. So I try to not look intimidating.

Ben Shaberman:

Okay. Well, again, I'm sure you're a great mentor. So let's talk about optogenetics and in your own words, of course, I'm asking you a question so you should be in your own words, how would you describe optogenetics and what are the opportunities it provides for patients, but what are the challenges in developing and implementing an optogenetic therapy?

John Flannery:

Yeah, the term comes from mixing two terms together. It's optical, so it's triggered by light and the thing that's triggered by light is protein, but it's delivered to the tissue, in our case the retina by a gene, by putting the gene in, not the protein. So that's where optogenetics come from. The first time I heard about it, a colleague of ours at Stanford who is a neurologist that works on narcolepsy, patients that have narcolepsy have a small center deep in their brain that has a defect that causes them to fall asleep randomly. And previous to his work, the therapy was to put a electrode wire deep to that center in their brain and stimulate it electrically. The trouble with that is that the wire gets covered with cells called glia over time and it insulates the wire. So you have to keep turning up the electric current to make it work and eventually it stops working.

So their idea was to make the cells in that region of the brain in the narcoleptic patient light sensitive and use a fiber optic light to turn on an off cells and put an optogenetic sensor switch in those cells that we respond to light. So what I thought is that the retina is a natural place to do this because you don't have to drill a pit hole in a person's head to get the fiber optic in there. There's all naturally corny in the lens focus all the light on the retina. Another part of the literature that we had seen for a long time is that almost all the genes that... People that identify genes from patients have discovered, which they discovered without any particular bias, almost every single one of those genes causes a defect in rod photo receptor cells.

Like 95% of the retinal degeneration genes kill rods, a few of them kill corns. But there's many other kinds of neurons in the retina ones that aren't light sensitive that seem to survive for decades in the patient, but they don't allow the patient to see. So if you lose the photo receptor cells, the other cells

can survive but they're not responding to light. So the way those two marry together is that if you can take the second or the third neurons in the signaling chain and put in a new gene that makes them light sensitive, perhaps that would restore vision. So that's where the optogenetics concept came from. Several labs, including ours, had the same idea. It's a natural fit for optogenetics. So it's also a natural fit for many people who have heard of the spark LCA2, RPE65 gene therapy that uses a virus to transfer a naturally occurring normal copy of the defective gene in LCA2 to the retinal cell where it's missing.

So in optogenetics you use the same sort of delivery mechanism, AAV vector, but in the case of this, you don't put in replacement gene for a defective gene. You put in a light sensing gene into a cell that wasn't normally like sensitive. So it's gene therapy but it's not gene therapy to replace something, it's to put in a new thing. And in that way, in some ways it has an advantage because the number of patients could be much bigger because you don't have to put back the exact gene that the patient has a defect in because there's at least 300 different defects that are known for RP. In fact, you don't even have to know what the patient's defect is because you're not replacing it.

Ben Shaberman:

Yeah, that's I think one of the most exciting things for people with retinal diseases is it is gene agnostic and it has the opportunity to help people regardless of the disease and it has the potential to restore vision to people who have no vision left essentially. And so I know a lot of the trials now are focused on helping people have lost most or all of their vision with RP. But do you think it'll work for conditions like AMD and Stargardt disease which are more of essentially a central vision loss as opposed to losing outer retinal cells?

John Flannery:

Yeah, that's still an open question. Clinical imaging has gotten to be so sophisticated in the last couple of years, maybe a decade. Ophthalmologists get a very good idea of what cells are remaining in the patient's retina and where they are. So in areas where in Stargardt are in macular degeneration where the patient has a vision loss, we call it scotoma, they could have a big island in the center of their visual field, it can be randomly shaped. Some AMD patients can't see in the very center, but they can see in the edges. Some people have different areas of loss between the right and the left eye. If you do careful imaging, you'll be able to ascertain whether or not they have surviving second and third order neurons and those areas where their photoreceptors are lost. So if they still have ganglion cells in the regions where they can't see and those are intact and they're still connected to the patient's brain, optogenetics has the potential to fill in those areas because the cells that are targeted by the optogenetic gene therapy is the ganglion cells.

So in many patients, it looks like by imaging that they still have a inner retina in the areas of the vision loss that will still remain to be seen because no animal models that we work on to test the therapy have that kind of vision loss. All the mouse models lose vision uniformly across the whole eye. Most of the large animal dog and pig models have that and there's very few primate models of RP. So it's still an open question that'll be resolved only when people start testing these therapies in patients, so whether or not it can fill in a scotoma.

Ben Shaberman:

Right. So there are obviously some excellent opportunities for optogenetics again in restoring vision to people who have lost really all their vision. And again, it's gene agnostic, but what are some of the challenges in implementing or really designing an optogenetic therapy?

John Flannery:

The challenges are basically the same as all the other gene therapies that are clinically being tested in patients for other photoreceptor diseases, there's ones for Ushers and Stargardt, LCA2, et cetera. So you want to deliver the gene efficiently. So the optimizing how well the virus carrier works, getting it to as many cells as possible, you'd like to use as low a dose as possible because you don't want to trigger the immune system by the optogenetic protein or the virus. So efficiency is important so you could inject less as minimal as possible. There's some questions on optogenetics of what are the best cell to use to put the optogenetic protein into. So most of the companies, including ours are currently looking at ganglion cells because they're the last part of the chain of signaling in the retina. So in the fovea, for example, there's foveal photoreceptor cones and there's a bipolar cell which is a relay cell and there's ganglion cells and it goes to your optic nerve.

In other parts of the retina, there's cells that do some computation and image processing called amacrine cells and horizontal cells that process some of the image signaling before it goes to the retina. So some of the students in the lab are doing optogenetics where they put the sensor in bipolar cells, which are one step up in the chain from ganglion cells. One of the other students has put it in amacrine cells. And amacrine cells are very broad, they run horizontally through the retina and they input many, many ganglion cells can be a hundred. So there may be advantages, you may get a better percept for the patient by not putting it in the third cell of the chain by putting it up a little bit in the signaling and maybe you can retain some of the intrinsic properties that the retina does. So it may work better that way. So none of the clinical trials are currently using those other cell types, but we testing them in mice right now.

Ben Shaberman:

Right. And I think that brings up an important point. We're always excited about what's in a clinical trial, but behind those clinical trials a little further back in the pipeline are often better approaches that are moving toward the clinic. So I appreciate that you and other lab researchers are always coming up with better approaches to these different treatment modalities. So I want to move into the work that you and your colleagues have done for Vedere. So the original Vedere, as we say it's Vedere I, that particular approach was acquired by Novartis. Actually, the company if I understand correctly was acquired by Novartis and then there's a second incarnation which is being termed Vedere II. Can you talk about what the two different approaches, the Vedere I and Vedere II approaches do and what some of the advantages of each are?

John Flannery:

Certainly, the Vedere initial program was to take the photosensitive protein from middle wavelength that are called green cone photoreceptors and put that into the viral AAV vector and put the cone green pigment in ganglion cells. So it's what we call a one-component therapy. You put in the cone opsin and the light-sensitive molecule that the cone opsin normally used in the cone photoreceptors, it still uses. So it's still available, it's delivered the normal way, but now the cone opsin is in ganglion cells, and in the case the patients loses their rod and cone photoreceptors, it will be functioning in this other neuron down in the signaling pathway.

So the initial acquisition of the dairy by Novartis was to buy that intellectual property to do that program. And in other parts of the lab, we had identified by screening optimized viral vectors to deliver the cone opsin and so they licensed two or three AAV vectors and the cone opsin technology. But as I said earlier, we have other approaches in the lab at the time and Novartis could have bought those as well, but they didn't. So [inaudible 00:15:58] iteration now is looking at the other approaches and other

cell types and with other types of sensors for optogenetics. Novartis was pretty happy with keeping the name Novartis. They have all kinds of tote bags and hats and pens. So they didn't need our names, we're still Vedere, they didn't acquire the name.

Ben Shaberman:

Right. And Vedere is using, again... Or now Novartis, but also the existing Vedere is using this. The cone opsins that you mentioned and cone opsins have some advantages over the light-sensitive proteins that are being used by some other companies. Can you talk about the advantages of using cone opsin versus some of these other proteins that other companies are using?

John Flannery:

Yeah, initially we tried rod opsin with the same AAV delivery system to ganglion cells and rod photoreceptors using rod opsin are incredibly fast at seeing changes in the light and they're really, really sensitive. But when you move the rod option to another cell, this case the ganglion cell, it turns out that there's a transduction machinery in that cell that it plugs into. That machinery and ganglion cells is much slower than the machinery it was using in photoreceptor. So we found that the rod opsin, and even though the animals could sense light very sensitively, they didn't need any goggles or anything. They could only do tasks... The mice could only do tasks that didn't require the vision to be quick. So they could move. If you had a box with a light and a dark side, they prefer to be in the dim light.

They feel safer so they could move to the dim part of the chamber. But if you did a task that required them to see quickly, a vision to refresh, like if you put an iPad at either end of the cage with stripes on it, that moved a little bit, they couldn't do that. But we found with the cone opsin, it's also really, really fast in cones when you put the cone opsin in ganglion cells, it's much, much faster than the rod opsin for biophysical reasons and it's just as sensitive. So we felt that that was a good system for providing vision. And in animals, in mice, even though they don't have a vision that's as good as people, in some cases the mice can perform vision tasks as well with cone opsin as wild type mice, normally cited mice.

But in measuring the response, the cone opsin may not be as fast as you ideally would like for people moving around in the world where your eyes are moving, your head's moving, the objects moving, et cetera. So the current Vedere programs are using photo switch molecules that are not the naturally occurring ones. And the thought is that they will respond more quickly than the cone opsin. Both those programs, the cone opsin, and the new Vedere programs don't require any light-intensifying goggles. So for example, the gen site molecule is called Crimson R. It's incredibly fast. There's no issue that it's going to not be fast enough for moving objects or moving around, but it's not very light-sensitive. So that's what the goggles provide is they make the light brighter.

Ben Shaberman:

So the advantage if I understand of the Vedere approach is that people will be able to see more in natural light without the need for these light-intensifying glasses. And also that they should be able to perceive movement pretty accurately without blurring. That's something I don't think we think about with vision is that healthy eyes that don't have retinal issues can see objects moving very well. Our eyes can respond quickly to the changing environment in front of us. I don't think we appreciate how well our retinas and the rest of our visual system enable us to do that. So that's something that you have to account for in these optogenetic therapies. And it sounds like Vedere, the two approaches do that pretty well so that you can see movement without things being blurry.

John Flannery:

And one of the important things for the students and myself in the lab is to keep trying to understand how the retina works when it's healthy. And there's still many, many things to understand that aren't understood about how the retina works. But one thing that's clear is that if you stop the retina from moving, you'd have to anesthetize the person or the animal... Actually, the image goes away. So the way the retina is normally wired, it requires the image to be moving across the retina all the time. If you stop the eye from moving, the image actually disappears. So movement is intrinsic to the vision process.

Ben Shaberman:

Interesting, I was not aware of that. That is very fascinating. So for my last question, with some of these more advanced optogenetic approaches, like what Vedere is working on right now, what do you think people will be able to see if these therapies work as planned?

John Flannery:

That's a great question that's really difficult to answer. So far all of our programs have been testing in animals and you can only get the animal vision back to where it was wild type and mouse vision is nowhere near close to what human vision is. Studies we've done in dog and pig models that has the same issue. With the dog models, you can design behavioral tests that the dogs will run a Y-Maze or things like that. Trying to get the pig models to do any behavior is like a comedy show. They just won't do it. And so I think from the patients will be the first people to really tell you what they can see. We can make some calculations what they can see. I think also patients' brains are very plastic as we say. And so I think the patient's vision will get better the longer they use the device, the optogenetic sensor or the headset goggles, et cetera.

In the LCA2 trials, the patients get better with time. And from what we know about how the gene therapy works, the gene therapy doesn't really change much after about a couple of weeks with stabilizers, but the patient's vision changes and that's their brain getting used to the new input. So I think in the few patients from gen site that have been reported so far, it's very encouraging, but those patients have only used the goggles for a couple of weeks, they seem to be seeing remarkably well. And so I think they'll just get better. Hopefully, for the other programs from the other groups, including ours, that the patients will get some vision restoration in a couple weeks, they should get a big gain of function. And then I think their understanding of what they're seeing will get better. In the electronic prosthetics, that was definitely the case. The patients learned how to use the second site device with time, they got better and better.

Ben Shaberman:

And I'm sure our listeners, many of whom our patients would agree that even a modest amount of vision improvement for somebody that has no vision or very little vision would be huge. So this approach, the work you're doing is really-

John Flannery:

At a recent meeting, one of the patients said, If you could just get me back to where I was five years ago, which tells you something about what people's expectations are, I don't think there's much opportunity for optogenetics to ever get back to 2020 vision because just the way the photoreceptor is so amazingly good at that. Will it provide vision for people to move around the room? I think so. It's an open question if you'll ever be driving or reading your phone or something. I think it just remains to be found from the patient's reports.

Ben Shaberman:

Right. Well, John, this has been, no pun intended, very enlightening. I appreciate you reflecting on the work you've done and what's coming down the pike. We're really excited to see the Vedere approaches move toward clinical trials. It provides a lot of hope for, again, people that have very little vision left or no vision left. So again, thank you for taking time out of your day to talk about this great work and we wish you continued success, you and your lab as you move forward. So again, thanks for joining us.

John Flannery:

Happy to talk to you. Thanks very much.

Ben Shaberman:

Sure. And listeners, thank you as always for joining Eye on the Cure and we look forward to having you back for our next episode. Stay well.

Speaker 1:

This has been Eye on the Cure. To help us win the fight, please donate at foundationfightingblindness.org.