Text: Louise Hagbard, Ph.D.

STEM CELL INNOVATORS

PETER ANDREWS

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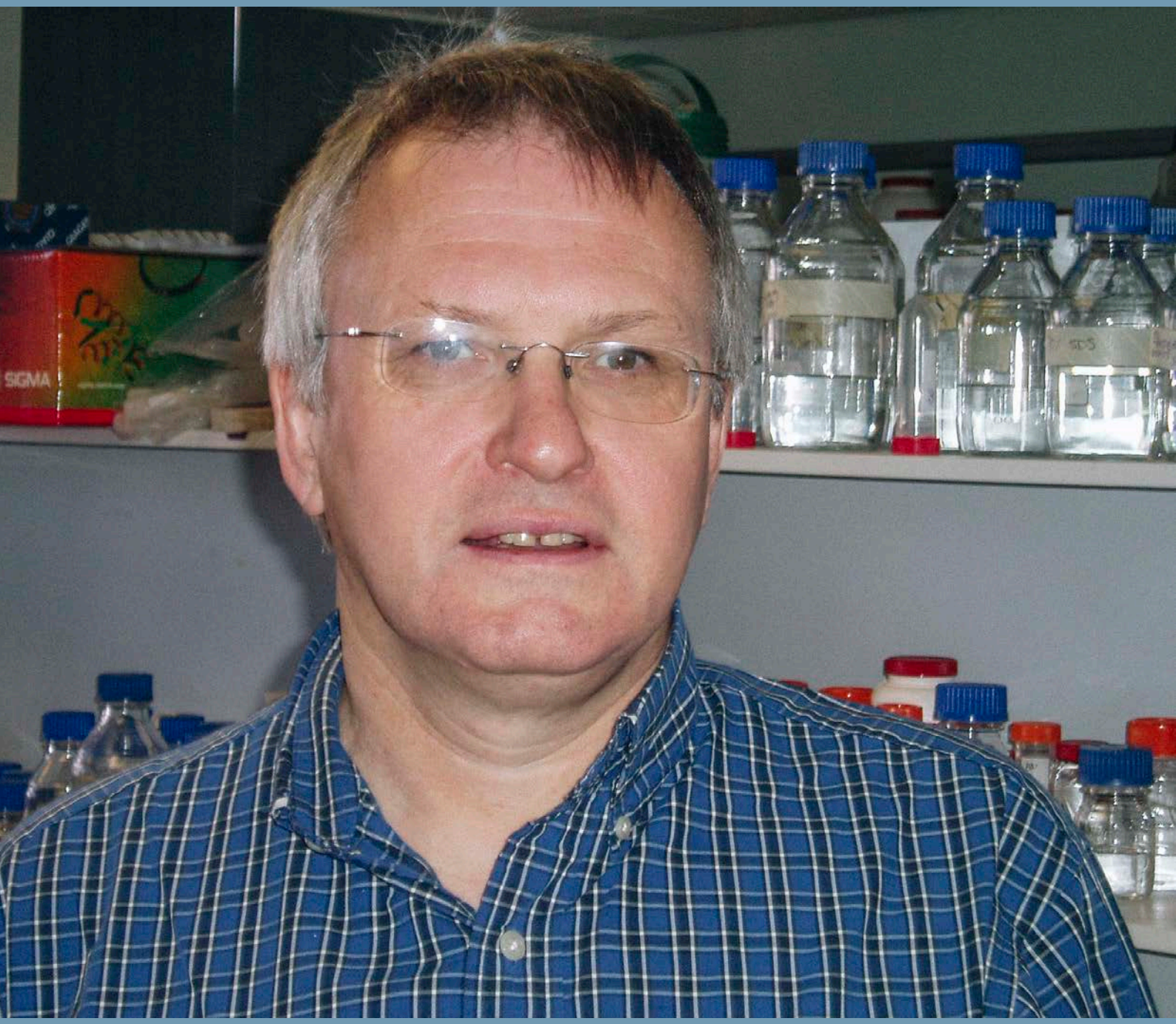

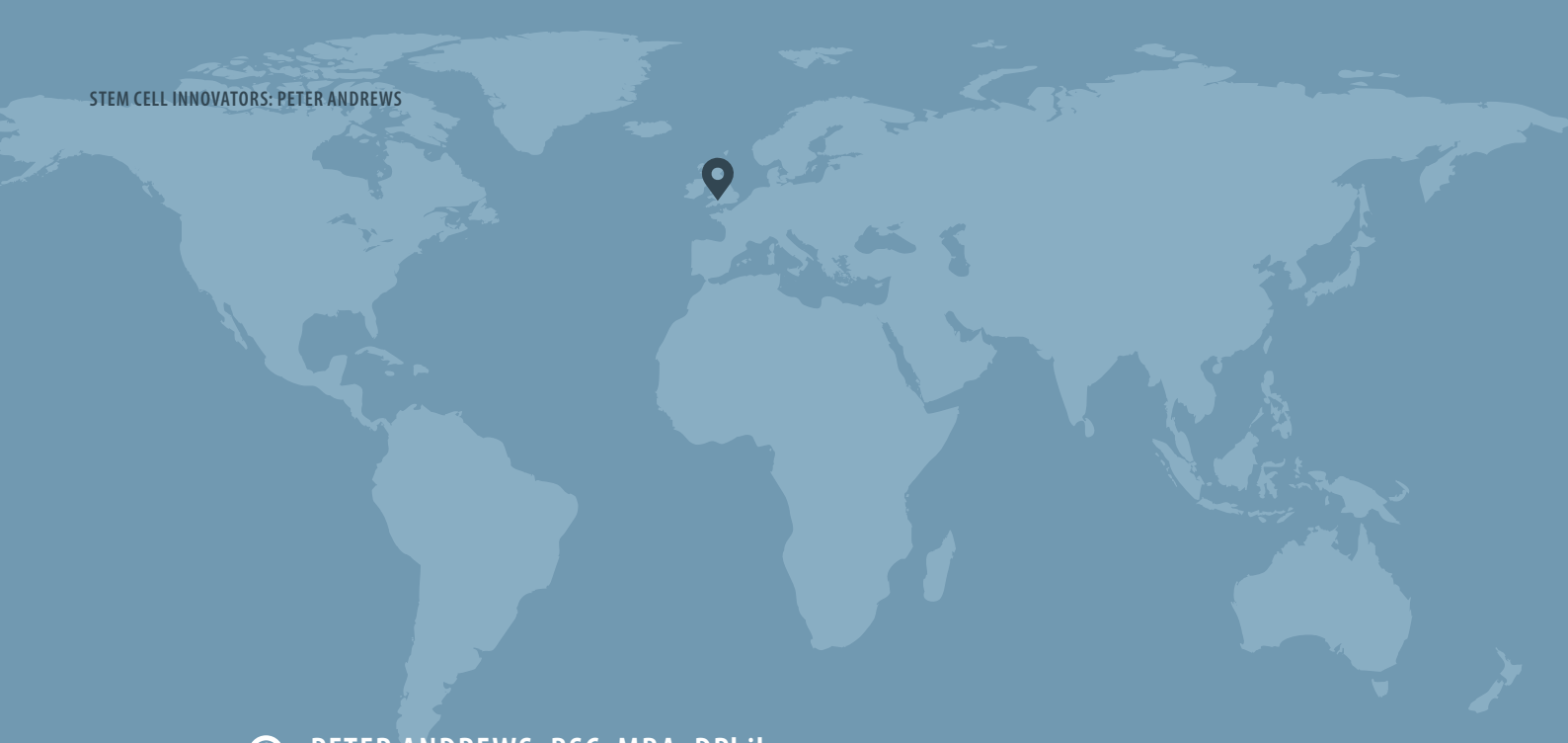

PETER ANDREWS, BSC, MBA, DPhil

ARTHUR JACKSON PROFESSOR OF BIOMEDICAL SCIENCE,
THE UNIVERSITY OF SHEFFIELD, UNITED KINGDOM

ABOUT: **Professor Peter Andrews has devoted his research career to studying the biology of human embryonic stem (ES) cells and their malignant counterparts, embryonal carcinoma (EC) cells. Prof. Andrews was the first scientist in the UK to work with human ES cells, following their derivation in 1998. Prof. Andrews' laboratory studies the causes and consequences of the non-random genetic abnormalities observed in human ES cells after prolonged culture, as well as the progression of stem cell-based cancers. Further work is focused on using induced pluripotent stem (iPS) cell techniques to establish models to study pediatric cancers. Prof. Andrews coordinated the International Stem Cell Initiative and was the director of the Pluripotent Stem Cell Platform, a hub under the UKRMP. He is also on the editorial board of several stem cell journals.**

Prof. Andrews holds a D.Phil. from the University of Oxford, and after postdoctoral periods at the Institute Pasteur and Memorial Sloan Kettering Cancer Center, he joined the Wistar Institute in 1978 where he studied the biology of embryonal carcinoma (EC) cells. In 1992, he moved to Sheffield as an Arthur Jackson Professor of Biomedical Science. Prof. Andrews was a co-founder and director of Axordia Ltd., one of the UK's leading hESC companies (now a subsidiary of Pfizer). Axordia is focused on developing and commercializing technologies and materials for the hESC industry, hESC-based products for drug discovery, and hESC-based therapeutic treatments for cardiovascular disease, diabetes, and Parkinson's. Prof. Andrews has been involved in the derivation of several clinical grade hESC lines (the Sheffield lines), deposited in the UK Stem Cell Bank.

THE UNIVERSITY OF SHEFFIELD

The University of Sheffield is a public research university in Sheffield, South Yorkshire, England. It received its royal charter in 1905 as successor to the University College of Sheffield, which was established in 1897 by the merger of Sheffield Medical School, Firth College and Sheffield Technical School.

Please tell me your story. How did you get into the stem cell research field?

"I think, for me, it started back in late '60s early '70s following Dr. Stevens' work on the 129 mouse and the teratomas. Stevens noticed that the primordial germ cells that gave rise to teratomas looked a lot like the cells of considerably earlier embryos and that they could differentiate. Because these cells could give rise to cancerous as well as normal cells, they became known as embryonal carcinoma, or EC cells. A lot of developmental biologists got interested in teratomas as a route of looking into the mechanisms of embryonic development. There were various groups around that got hold of tumors and were growing out cell lines, trying to define what mouse Embryonal carcinoma (EC) cells were and trying to define pluripotency. There was Chris Graham in Oxford, François Jacob's group in Paris, and of course Martin Evans with Gail Martin in the UK."

"I did my Ph.D. in Oxford and then I went to the Pasteur for my first postdoc in François Jacob's group. I joined them because they were one of the main groups at the time working with teratocarcinoma cell lines. So, this was all before human ES cells, even before mouse ES cells. From there, I went to Memorial Sloan Kettering

EMBRYONAL CARCINOMA (EC) CELLS

Embryonal Carcinoma (EC) cells are the malignant stem cells of teratocarcinomas, a subset of germ cell tumors. EC cells are malignant counterparts of embryonic stem (ES) cells and have been used as simple surrogate models for the study of pluripotent human ES cells, to define marker antigens that characterize the undifferentiated EC phenotype, and for monitoring differentiation. EC and ES cells share some common features, such as expression of stemness markers Oct4 and Nanog. They proliferate fast and can be maintained in culture. Human EC cells are typically highly aneuploidy and include some karyotypic changes that also occur as non-random genetic abnormalities during long term culture of human ES cells.

“I think the problem we still have is understanding the basic biology of the cells.”

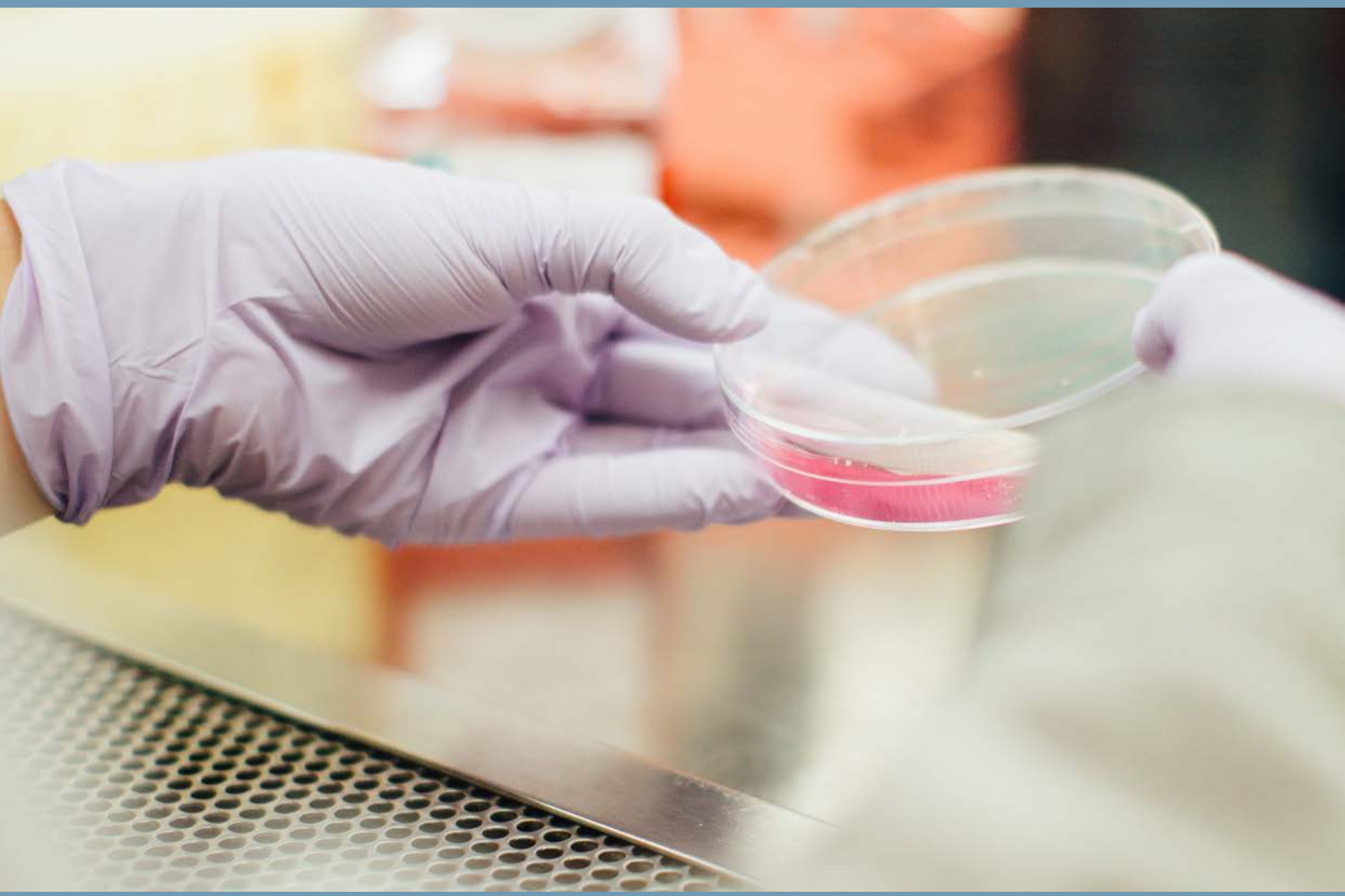

Photo: Drew Hays/Unsplash

Photo: Viktor Kjellberg

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Cancer Center and then moved to the Wistar institute in ‘78 to work with Barbara Knowles and Davor Solter. That is really when I got hooked. Barbara and Davor had made one of the first monoclonal antibodies to recognize the key cell surface antigen, SSEA-1, which is expressed by both mouse ES and EC cells. People were starting to think that if you could get information on mouse embryos from looking at mouse teratocarcinomas, maybe you could get information on human development by looking at human teratocarcinoma cell lines. There had also been a couple of papers published describing cell lines derived from human teratocarcinomas. These expressed an antigen known as the F9-antigen, related to SSEA-1, so everyone thought they had found human EC cells.”

“When I moved to the Wistar Institute Davor had established a collaboration with a clinical group in Minneapolis, working with cell lines from human testicular germ-cell tumors. So I started working on these cells, with the idea to see if we could find a human EC cell. Indeed, in some cell lines there were SSEA-1 positive cells but it took us a couple of years to suddenly realize that we were looking at the wrong cells, and that actually the human EC cells didn’t express SSEA-1. There was another monoclonal antibody that Davor and Barbara had made which identified another cell surface antigen they termed SSEA-3. This antigen is expressed by cleavage stage mouse embryos but not by mouse inner cell mass cells or mouse EC cells. It turned out that many of these human cells we were looking at were positive for SSEA-3, and suddenly it twigged with us that the human EC cells were actually SSEA-3 positive and SSEA-1 negative, completely different to the mouse.

So, I really got involved in the human pluripotent stem cell activity in the late ‘70s then. I was really trying to identify human EC cells, what they were and if we could get a pluripotent one which would differentiate. I think it was Martin Pera and I who were the two people who made their career on it. This was just after Louise Brown was born and IVF clinics were started, and we and others were wondering, “What’s the possibility of getting human embryos and deriving human ES cell lines.” At the time, there was very limited access to human embryos, it was just logistically difficult. Jamie Thomson was a PhD student with Davor in Wistar when I was there and he later ended up in the primate center in Wisconsin, with access to primates and an interest in early development. That access to the monkey embryos allowed him to derive the monkey ES cells, which was then the key to him getting access to some human embryos. So, he tried what he had done with the monkey embryos on the human ones and derived his human ES lines. And it was quite comforting when Jamie derived his human ES cell lines that actually they expressed a pattern of antigens that we had described in human EC cells, being different from mouse EC and ES cells. So, a lot of the markers that we had originally characterized, for characterizing human EC cells, turned out to be the ones that everyone uses today for characterizing human ES cells.”

Back then, were you thinking about the potential for regenerative medicine.

"It never really crossed my mind, I was focused purely on developmental biology. There was a pluripotent human EC cell line that I characterized, NTERA-2, which I published in 1984. It turned out that this line made neurons and group at the University of Pennsylvania who got hold of the cells developed further methods for purifying neurons from those cells, and actually did some trials with stroke patients. I don't think they were particularly sensible trials. Nothing good came out of it, but fortunately, nothing terrible happened either. They published that but I wasn't involved. That was probably the first clinical trial, for what it's worth, of derivatives of pluripotent cells. It hadn't really crossed my mind, until that point actually, that there was a potential in regenerative medicine.

When I moved to Sheffield in '92, Harry Moore, joined the university at the same time. He came from an IVF background with interests in reproduction. And so, we started talking and trying to work with local clinicians here to see whether we could access human embryos, with the idea of deriving ES cells. Just, we couldn't get it together. No-one had managed to do it until Jamie did, and that really set the ball rolling."

What about the ethical view on embryo research back then?

"I think that the regulatory environment has always been good in the UK, much because of the IVF invention. It had provoked a lot of discussion in the early 1980's about the ethics of working with human embryos in Britain, and that led to a parliamentary commission and the so-called Warnock Report. I think really a key person on that was Anne McLaren, a developmental biologist, and that led to the law being passed in parliament in Britain which was the Human Fertilization and Embryo Act of 1990. That really provided the legal framework in the UK for working with human embryos. I'd say it was all done without thinking about ES cells. It was more in the context of understanding human development."

What have we learned from the EC cells that can be used in the development of pluripotent stem cell-based therapies?

"Everyone looks back at EC cells and the clinical pathology of germ-cell tumors in the pursuit of trying to understand whether ES cells can be dangerous or not. When you make a teratoma from a human ES cell, there are some cell types and tissues that sometimes appear that you might want to look at in the context of germ-cell tumors, since they are regarded as clinically undesirable, a bad prognostic value. The other thing is that we've done quite a lot on the issue of genetic variants cropping up in human ES cell lines when they are grown for a while in cultures. We keep referring back to EC cells to try to understand whether particular genetic changes may be significance in terms of tumorigenicity, and whether they might be good, bad or indifferent. So, I think the two cell types have always gone hand-in-hand, but it's probably how I grew up and how my own career developed that I've always had this considered the close link between the two."

What are your thoughts around the future for pluripotent stem cell therapies?

"I think it has the potential to be big, but I think it's going to take a long time. First, we have to see whether or not there really are clinical benefits, and then work out how to produce the cells in a routine way, to the right standards, and how to deliver them. And of course, what is the business model for this and who is going to pay. So there's a lot of problems to resolve but who knows? In 20 years' time, I'm pretty sure that in some areas, for some diseases, it will be important. There's clearly a number of things on the horizon. Parkinson's is obviously the next one, where I believe Jun Takahashi in Japan has already started in patients."

"We've been loosely linked with is Pete Coffey's clinical trial for treatment of age-related blindness. He's actually using the first cell line that my colleague Harry Moore derived in Sheffield, the SHEF-1 cell line. There

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"Trying to understand why cells do things motivates me: What are the signaling mechanisms, what are the cues, what are the rules that make cells do one thing rather than another? That's fundamentally what has driven me ever since I started doing science."

have been a lot of issues along the way, but it's good to see that he now has two patients who have shown some clinical benefit."

What do you think will be the biggest hurdle to overcome to reach the clinic with pluripotent cells?

"I think the problem we still have is understanding the basic biology of the cells. That's always my hang-up, as I am a basic biologist. I have a problem dealing with the push for translation that comes particularly from funders and everyone who wants to make money out of it or cure everyone, but don't actually understand a lot of the basic biology of the cells. How cells make decisions, how do you get the right sorts of cells out? And one of the big problems there is the relationship of these cells to the human embryo. Of course, we know an awful lot about mouse embryos at this point, but we don't know nearly as much about human embryos except that there's a lot of differences. Still, people try and squash the data about human ES cells into information that is from the mouse embryo and not the human embryo."

"There are now a few groups that are actually doing real embryology on early human embryos, which I think is really important to give us some better insight into what human ES cells correspond to and the mechanisms that control their differentiation."

"One of the big issues is how derivatives of these cells mature. I mean, a common observation is that the cells people were getting out of human ES cells tended to have immature properties as opposed to mature properties of whatever cells people were trying to make."

"Another issue is getting the correct cells from ES cells. I think Malin Parmar's work on getting the right sort of dopaminergic neuron is interesting. The discovery of two regions in the brain from which dopaminergic neurons behaved differently has allowed them to develop ways of

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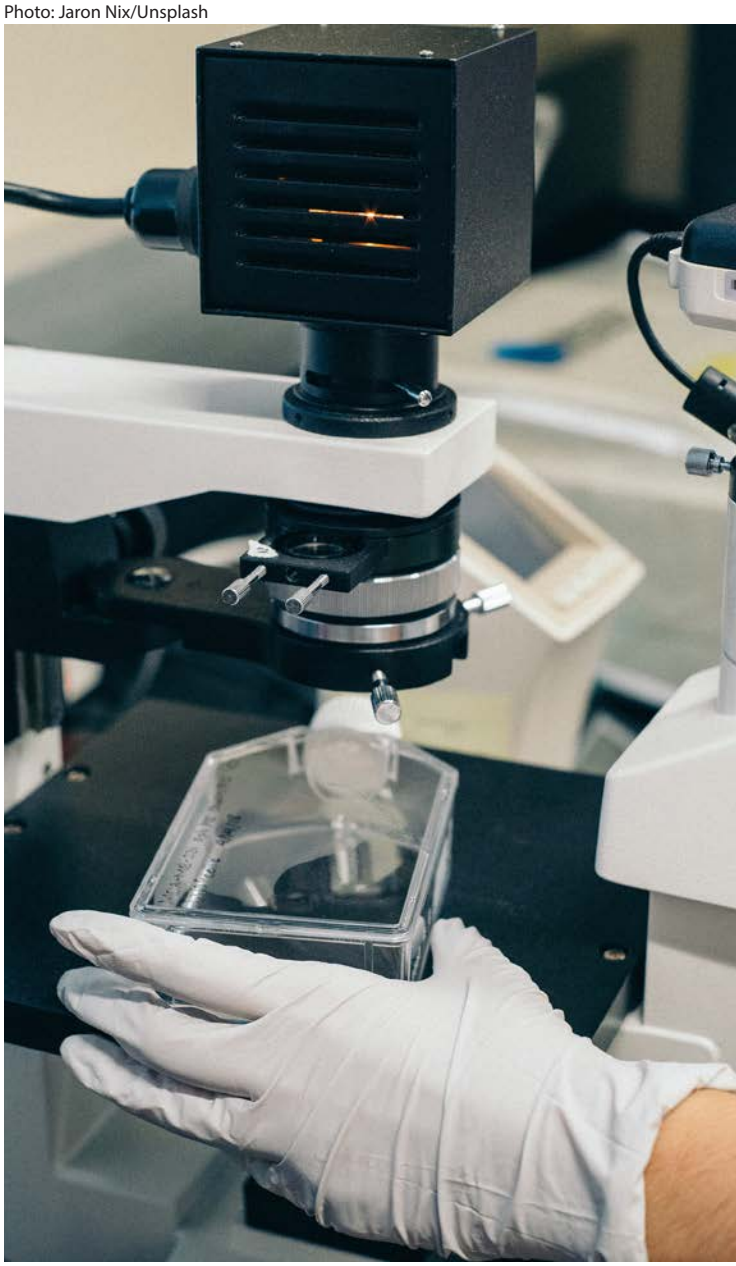


treating the ES cells to get the right sort of dopaminergic neurons out so that they get better engraftment. That's really quite interesting and probably going to be reflected in what people do elsewhere later in other systems."

What advances do you hope to see in the coming years?

"As I mentioned, I think that there is the whole area of developmental biology to visit. Disease processes, how embryos develop, how cells develop throughout life, and then what can go wrong. With regards to all the differentiation protocols currently around, there is a kind of bucket level chemistry approach. To make dopaminergic neurons, for example, there are recipes in which you pour onto cells signaling factors, cytokines, in ways that in the embryo, in life, cells never see.. Rather, they get exposed to particular concentrations of factors, for particular periods of time, in cyclical ways which are not being reproduced in many *in vitro* protocols. There's a huge area of trying to understand how the signaling pathways really work. So I think it is going to be really interesting to see what happens when people seriously start looking at single cells and looking at how they respond, over time to particular signals, and combinations of signals."

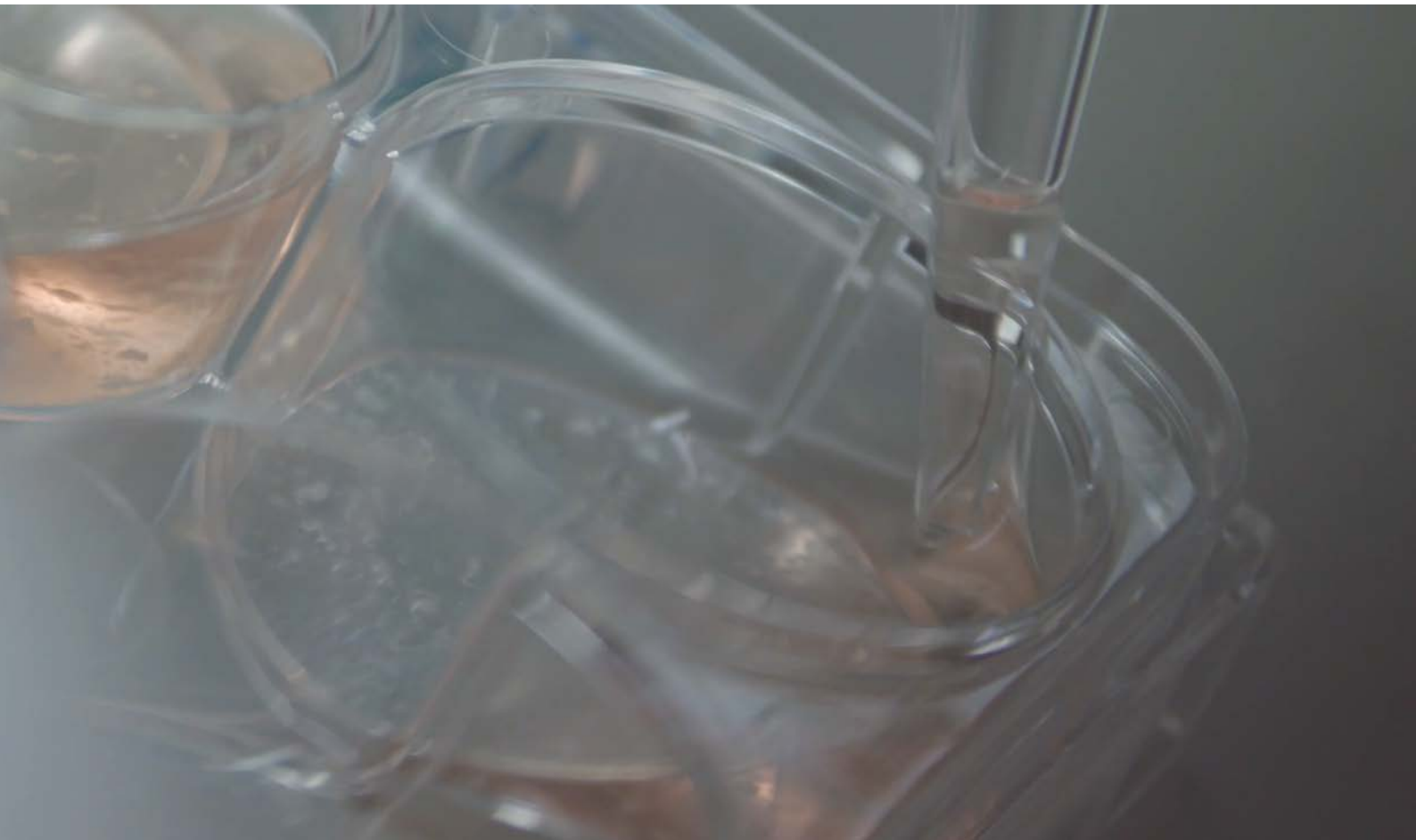
What is your biggest inspiration in your everyday work? What motivates you?

"Trying to understand why cells do things. What are the signaling mechanisms, what are the cues, what are the rules that make cells do one thing rather than another? What's the logic systems behind cell decisions, fate decisions? That's fundamentally what has driven me ever since I started doing science."

If you had a hundred million pounds for any kind of research area, what would you do?

"I would answer it in the context of what I think is wrong with the current funding preoccupation with translation. I think what is really crucial is the underlying blue skies, curiosity-led research,. I would favor spending this money on curiosity-led basic research and not top down directed projects. It's fundamental to enable people to enter unknown territory, explore it, and pick up on ideas." •

Photo: Ritualen



PETER ANDREWS ON ACHIEVEMENTS

? If you look back on these 20 years since Thomson derived his first line, what do you think is the biggest achievement?

! Well, of course, the development of IPS cells is a major change. It changes the landscape a lot, from many points of view. There's always the logistics issue of getting access to human embryos. For a lot of the early cell lines, ES cell lines were only derived by groups who had easy access to human embryos. When the iPS cells came along it suddenly meant that almost anyone could do it, and it opened up the field to a lot more people. This of course caused some problems, I think, because these cells suddenly became an off-the-shelf tool resulting in, in some cases, less good quality control, and a focus on potential applications without considering some of the underlying basic biology. Another major thing is that it overcomes ethical issues with the ES cells. And, of course, there's the basic biology of it, which is, I think it blew everyone's mind that we could reprogram a somatic cell to an embryonic state just by over expressing four genes. No one expected that. There's a lot of things which have surprised people along the way, and one thing is simply how quickly the whole field has moved. That after 20 years, there are clinical trials taking place. That is actually quite remarkable. It's still very early and I think we still have to be very cautious, but it's still remarkable.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