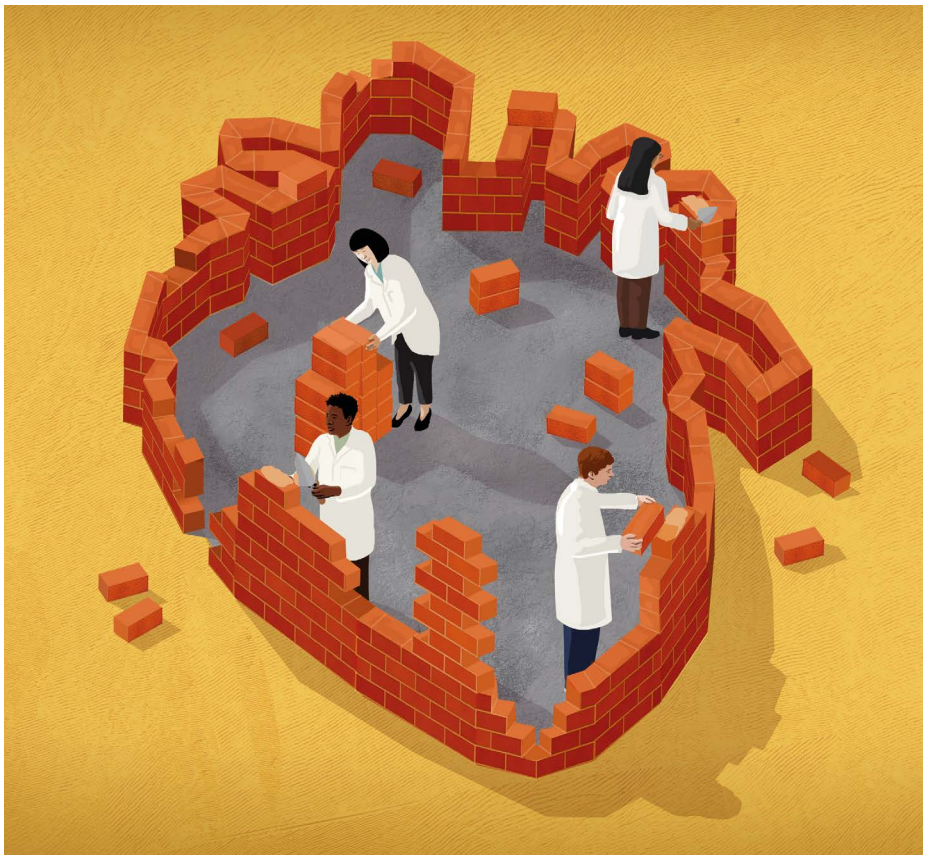

SAM FALCONER

Cells or drugs? The race to regenerate the heart

Researchers are working out how to get the heart to heal itself instead of laying down scar tissue after a heart attack. **By Benjamin Plackett**

Twenty years ago, cardiologist and stem-cell scientist Piero Anversa published an exciting paper. He was then a prominent researcher at New York Medical College in Valhalla, and his data in mice showed that injured hearts could regenerate with the help of stem cells taken from bone marrow[1] – contrary to prevailing wisdom.

Myocardial infarction, commonly known as a heart attack, deprives cardiac muscle cells of oxygen, causing them to perish. The human heart responds by laying scar tissue over lost muscle. But these reconstituted areas don't pump blood as competently as before. In time, this can lead to heart failure – particularly if other heart attacks follow. The implications of Anversa's work were clear: stem cells, through their growth and proliferation, had the potential to reverse the damage caused by heart attacks and thereby prevent heart failure.

But other researchers who attempted to replicate these mouse studies found themselves coming up short. Allegations of faked results eventually began to surface, and Anversa, who had since joined Harvard Medical School, and Brigham and Women's Hospital in Boston, Massachusetts, was forced to leave his posts in 2015. Two years later, Brigham and Women's Hospital paid the US government US$10 million to settle allegations that Anversa and his colleagues had used fraudulent data to apply for federal funding. And a 2018 investigation conducted by Harvard called for 31 of Anversa's papers to be retracted.

This saga has dampened the enthusiasm that once surrounded research into stem-cell therapy, says Michael Schneider, a research cardiologist at Imperial College London. "The controversy, overt scientific misconduct and evidence against Anversa's claims has cast aspersions on the field more generally," he admits. That's unfortunate, because many other stem-cell scientists are conducting legitimate research.

Meanwhile, another heart-healing strategy has emerged, drawing inspiration from species that, unlike humans, can regrow cardiac muscle after trauma. Researchers are seeking to learn more about the molecules produced by zebrafish (*Danio rerio*) hearts as they heal themselves – and are investigating whether injectable drugs containing the same substances could also yield reparative results.

The question is now whether it will be stem cells, small-molecule drugs or a combination of the two that achieve the goal of convincing the heart to heal instead of scar.

An evolution of thought

In the wake of the Anversa scandal, there has been an important evolution of thinking on the stem-cells front. A 2019 literature review pointed out that newer studies tend to show the most significant impact from stem-cell therapy comes from the substances the cells secrete, rather than their proliferation[2]. "After many years of work, we find that when we deliver cells into the heart, the benefit of replaced damaged cells is only minor," says the review's author Javaria Tehzeeb, an internal-medicine specialist at the Albany Medical Center in New York. The real work of regeneration happens, she explains, when the cells produce growth factors, which in turn affect heart repair by reducing inflammation and stimulating the development of new heart muscle.

That means stem-cell therapies share some similarities with the drug strategy – essentially it comes down to molecules secreted by the stem cells versus molecules that are directly injected. But they also have important differences.

First, part of the stem-cell therapy benefits might still come from the cells' proliferation, even if that bonus is relatively small. Second, there's little control over what substances the stem cells produce once they're injected, whereas specific molecules can be administered at known doses. And finally, the logistics of scaling up and delivering these two therapies will be very different.

A study published in 2020 showcased the importance of stem-cell-produced molecules by looking at the structural integrity of proteins found in infarcted mouse hearts[3]. The scientists artificially induced heart attacks in eight adult

mice. Four weeks later, they administered stem cells to half the rodents. After a further four weeks, their hearts were removed and washed with a series of buffer solutions and chemical reagents to extract the proteins, which were then analysed. "We essentially did a massive scan of every single protein in the heart," says Andre Terzic, lead author of the study. The authors were able to identify almost 4,000 proteins, and showed that heart attacks distorted the structure of 450 of them. But with stem-cell therapy, that number fell to 283.

"Proteins are the intimate components that make our hearts work properly, and when the heart is diseased, they become damaged," says Terzic, who is director of the Mayo Clinic Center for Regenerative Medicine in Rochester, Minnesota. "The ability of these stem cells to secrete healing signals is probably a key element to what we've observed."

All cells and tissues are constantly telling each other what they need and whether they're stressed through molecular signalling. "When you lose a chunk of cells in a heart attack, you lose part of that conversation," explains Charles Murry, an experimental pathologist and director of the Institute for Stem Cell and Regenerative Medicine at the University of Washington in Seattle. Injected stem cells could be filling in the missing dialogue by secreting signalling and rescue molecules, he explains.

"There's been an awful lot of time and money spent on stem-cell therapy, raising false hope."

Although this sounds encouraging, there are still parts of the stem-cell-therapy approach that need to be finessed. In a 2018 study, Murry and colleagues transplanted approximately 750 million cardiomyocytes into macaque monkeys that had experienced major heart attacks[4]. One month after the intervention, the amount of blood pumped by their hearts had increased by 10.6% compared with just 2.5% in the control group. This advantage persisted three months later, but one out of the five stem-cell-treated monkeys suffered arrhythmias. The onset of arrhythmia wasn't previously observed in small-animal studies, but it is a known complication of heart attacks. Nevertheless, the researchers thought it could be a potential side effect of the stem-cell infusion. "Obviously it isn't statistically significant, but common sense led us to classify this as a treatment complication," says Murry.

In addition to safety concerns, stem-cell therapies are also beset by questions of practicality. "Think of a lab with all these cell culture flasks where you have to grow millions of cells just to create a single dose," says Terzic. "Now imagine tens of thousands of patients. It's a formidable effort to be ready, especially if you want to intervene rapidly. You don't have the luxury of time to build up supplies."

SLOWMOTIONGLI/GETTY

Zebrafish *(Danio rerio)* can regenerate their cardiac tissue after injury.

Small-molecule drugs and fish

That's one reason why some people think the promise of cardiac rejuvenation lies elsewhere. "There's been an awful lot of time and money spent on stem-cell therapy, raising false hope in patients – and so far, the clinical outcomes have been largely disappointing," says Paul Riley, a cardiovascular scientist at the University of Oxford, UK. Riley is investigating whether inserting specific molecules into the heart might be more effective.

Human hearts can't regenerate on their own, but other animals do have such abilities. Zebrafish, for example, can regrow their hearts after as much as 20% is removed. Newborn mice can also regenerate heart tissue. Observing the molecular pathways in these animals might make similar results possible in humans.

Research has shown that following a myocardial infarction in zebrafish, the epicardium – a membrane surrounding the heart muscle – produces molecular signals that might kick-start muscle-cell regeneration[5]. The hope is that manipulating the human epicardium could elicit the same therapeutic results. "There are probably approaches we can take to target the cells that exist in the heart with small molecules or drugs, that could invoke repair and regeneration," says Riley.

Back in 2011, Riley and colleagues showed that this is theoretically possible[6]. They pre-treated adult mice with a daily injection of a protein called thymosin β4 for one week before inducing an infarction, and found that these mice were able to produce new cardiac muscle. This offers a road map to a pre-emptive therapy. "If an individual is at high risk of a heart attack", says Riley, "then it's conceivable they could be advised to take a priming or preventative therapeutic, which may counteract an event, but it's not quite the holy grail of restoring lost tissue after a heart attack that we're searching for." In other studies, Riley has since shown that other proteins besides thymosin β4 might also have a role in stimulating the epicardium to regenerate the heart[7].

It's easier to see how the drug route offers clearer prospects for scaling up – but the science behind this approach is newer, and there haven't been any clinical trials in humans yet. "What goes in stem cells' favour is the body of work behind them," says Tehzeeb.

It might be that stem-cell therapies achieve government approvals first, but then drugs overtake them once the science and research have had time to catch up. "When we get to the end of the line with molecules, then maybe we can say stem cells are a thing of the past," Tehzeeb says. "But until then, we should continue to pursue their potential."

Murry echoes that sentiment, arguing that findings from both camps could end up helping everyone's research. "We need an ecosystem with a competition of ideas, and as long as it's all openly published then we'll figure it out," he says. "That's the better approach, rather than saying my idea is better than your idea."

Benjamin Plackett is a science journalist based in London and the Middle East.

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