

Cardiology and Stem Cells

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Cardiac conditions like mitral valve disease, dilated cardiomyopathy (DCM), and congenital heart disease are widely prevalent in dogs, Dr. Mark Oyama said. However, diagnosis is fairly expensive, and treatments are generally limited to relieving symptoms rather than addressing the root causes of disease. For most acquired heart disease, the best veterinarians can do is try to alleviate symptoms and maintain quality of life.

Mitral valve disease and DCM are particularly frustrating for the breeding community, since they appear in the middle to later years of life, after dogs are past breeding age. Both can probably be traced back to a genetic cause, though nutritional, toxic, and infectious components cannot be ruled out.

“The majority of breeds have some liability for coronary disease,” and there have been advances in identifying heritability patterns and possible causative genes, he said. “But as we approach a day when we understand the gene or genes that cause these diseases, we’re looking at additional therapies, and that’s where stem cell therapy comes into play.”

Dr. Oyama described DCM as a “global weakening” of heart muscle cells, leading to poor contractility of the heart muscle and progressive dilation of the heart. The progressive loss of function leads to buildup of fluid, exercise intolerance, abdominal distension, and poor appetite. The final outcome of the disease varies by breed: while Boxers are more likely to collapse from arrhythmic sudden death, most Great Danes succumb to heart failure. Despite best efforts, the one-year survival rate after a diagnosis of DCM is 15% across affected breeds, and about 50% in humans.

Until very recently, researchers believed that certain adult organs like the heart and brain were “terminally differentiated,” so that cells could not be replaced once they died. More recently, studies have revealed populations of adult stem cells in the bone marrow, liver, and heart that might rebuild these tissues. Dr. Oyama cautioned that “the intensity of that response is relatively low,” and not robust enough to repair major damage. However, the existence of stem cells in heart muscle tissues is now well documented.

This suggests two possible avenues for stem cell therapy: harvesting cells from other parts of the body, or augmenting native populations in the heart. Cells from bone and skeletal muscles have very good regenerative properties, and bone marrow cells can differentiate into nervous tissue, bone, or heart muscle. “They’re really quite plastic, or flexible, in what they grow up to be,” he said. “It’s all about their environment, nature versus nurture.” Initial human trials have shown improved contractility, although the

study groups of 10 or 20 were far short of the thousands needed before a new drug can be approved.

So far, most successful human or animal trials have had to do with myocardial infarctions, Dr. Oyama said. In contrast to a chronic genetic condition in which all the heart cells eventually degenerate and die, the studies focused on a single event of poor blood flow in one part of the organ. Use of autologous stem cells sidesteps issues related to cell supply, transplant rejection, and broader ethics, and there is hope that skeletal cells will develop very similarly to heart muscle if they are placed in the right growth environment.

After extracting the cells and labeling them with fluorescent dye, the next step is to return them to the damaged heart in the least invasive manner possible, while covering the widest possible area of the organ. One challenge is that stem cell solutions contain a variety of different cell types, including fibroblasts that would grow into scar tissue. Some papers have suggested that up to 50% of injected stem cells are “garbage cells,” Dr. Oyama said, but researchers are still learning how to purify the solutions.

Once the cells are ready for implantation, the least invasive delivery method is a catheter from the femoral artery into the left ventricle of the heart. Stem cells are relatively large compared to the capillaries at the end of the coronary artery, which are only one cell thick. Researchers believe that the stem cells sometimes wiggle through to the surrounding heart muscle, into an environment “that will hopefully induce them to differentiate into functioning, working muscle tissue.”

Dr. Oyama said his laboratory injects between one and five million cells per treatment, since a larger volume will block the smaller coronary arteries. The most successful trials so far have involved up to three million cells, which is in itself a limitation, since the total number of injected cells is minuscule compared to the billions that make up the heart.

However, tests show that the cells do stay in the heart, rather than dying off or being flushed through the system, and the benefits in human trials seem disproportionate to the number of cells injected. Researchers are now considering that the treatment might benefit patients by drawing native stem cells to the damaged area, or by secreting some beneficial hormone that has yet to be identified. Follow-up studies have shown that granulocyte colony stimulating factor (G-CSF), a protein, produces considerable cell proliferation, Dr. Oyama said. It appears to mobilize native populations of stem cells, though the cells seem to have less ability to find the area where they are needed.

Beyond cytotherapeutics, Dr. Oyama identified gene transfer as an opportunity to correct primary and secondary deficiencies, rather than trying to cure them. Once a target has been identified, he said it might be possible to “infect” it with a virus that contains an active, healthy copy of the defective gene. The technique has been used to arrest mucopolysaccharidosis (MPS) in dogs, and it may be a useful treatment for muscular dystrophy in human children and Golden Retrievers, and in a rapidly

progressing form of DCM associated with Portuguese Water Dogs. "Here, the emphasis isn't so much on replacing the damaged cell. It's on replacing the gene in the cell that makes it damaged."

Discussion

Dr. Butherus asked whether new heart tissue grown from stem cells integrates automatically with the existing muscle. Dr. Oyama acknowledged that heart cells must act in close coordination to ensure a smooth, steady heart rhythm, but that myoblasts taken from skeletal muscle "aren't very good at making little plug-ins with their neighbors." As a result, human health researchers have concentrated their efforts on bone marrow stem cells.

A participant asked whether there are any differences between late-onset heart disease and the mitral valve disease that often occurs earlier in Cavalier King Charles Spaniels. "Unfortunately, we don't really know," Dr. Oyama said, but the evidence points to two different conditions.

A participant asked whether it would make sense to inject cells into Dobermans during the occult stage of the disease, before the heart fully dilates. Dr. Oyama said that would be unwise until more is known about the long-term safety, efficacy, and survival of stem cells. He agreed that it would be worthwhile to harvest bone marrow cells while a dog is still healthy, rather than compromising a sick animal with sedation or anesthesia, though it is still not known whether these cells can be frozen over long periods of time.