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Translational Development of Mesenchymal Stem Cell Therapy for Cardiovascular Diseases

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Currently, a growing number of small clinical trials are testing the impact of cell-based therapies for heart disease.[1] In order to guide future approaches, it is important in the context of these trials to consider issues pertaining to mechanism of action.[2] To date, the bone marrow[3] has been the most widely studied source of cell-based therapy for heart disease, and this has been followed by skeletal myoblasts.[4] With regard to the bone marrow, both whole mononuclear bone marrow and cultured bone-marrow–derived mesenchymal stem cells (MSCs) have entered clinical trials. There have been attempts to study the impact of cell-based therapy for patients who have had an acute myocardial infarction or who have ischemic heart failure, and in the latter case both ventricular remodeling and hibernating myocardium have been targeted.

At present, the University of Miami has 2 trials in active enrollment and a 3rd in the planning stage. The goal of the current trials is to test the hypothesis that injected MSCs can produce reverse remodeling in the chronically infarcted heart by reducing the scar burden and improving cardiac structure and function. In parallel studies performed in pigs, we have tested the fate of injected cells and have documented engraftment, the formation of new myocytes (unpublished observations), and vessel growth.[5] Thus, our program currently targets the treatment of chronic ischemic cardiomyopathy. This disorder, in comparison with acute myocardial infarction, requires special consideration of the delivery method, and we have chosen to focus on direct delivery in the operating room or via a percutaneous injection system.

The development of cell-based therapy is still at a stage in which the primary goal is to establish safety, which is to say that most studies are at Phase I. Nevertheless, sophisticated imaging methods, such as cardiac magnetic resonance imaging (MRI), provide the opportunity to obtain surrogate efficacy markers as a secondary endpoint.

Preclinical Studies. In order to convincingly establish safety and efficacy, we developed a relevant preclinical model for ischemic cardiomyopathy in adult mini-pigs. The results of these studies were instrumental in providing the Food and Drug Administration with preclinical safety information as part of an IND application, and were also useful in performing studies regarding the mechanism of action of the cell therapy.

A porcine model was chosen due to the anatomic similarity between the porcine and human hearts, particularly with regard to coronary anatomy. A significant problem with pigs is the steep growth rate of immature animals, rendering them too large to fit in an MRI magnet, which is the primary imaging device used in these studies. In addition, the rapid rate of growth could confound interpretation of cardiac anatomy. To solve this problem, we created a model of ischemic cardiomyopathy in adult Göttingen mini-pigs.[6] The animals are studied at approximately 18 months of age. At this point, they weigh 35 to 40 kilograms, and for the next 6 months they will gain only another 5 kilograms. Reproducible transmural infarctions, affecting 20% to 25% of the left ventricle, can be created in the animals by occluding for 2.5 hours the left-anterior descending coronary artery with an angioplasty balloon. The scars, generally localized on the anteroseptal wall, were allowed to heal for 3 months. During the subsequent 3-month interim, autologous mesenchymal stem cells were amplified from a bone-marrow aspirate and then surgically delivered in the operating room under di-

rect visualization via injections administered throughout the scar and its border zone. The scar was the direct target of the therapy, the goal being to replace it with functional tissue.

In regard to the safety evaluation, animals were monitored for arrhythmias, laboratory blood tests were performed serially after injection, and whole-body autopsies were performed 3 months after the injections. Overall, the approach was very safe: no adverse events, anticipated or unanticipated, occurred. Of note, the whole-body autopsies found no ectopic tissue in the heart or elsewhere.

These preclinical studies demonstrated the very substantial repair potential of the therapy.[7] We used cardiac MRI to monitor the animals over the course of the studies, and—by administering cells at doses comparable to those used in current trials (20 million and 200 million cells per heart)—we reduced infarct size by approximately 25% and increased ejection fraction significantly, in comparison with pre-injection values. These effects were not present in the control group.

Clinical Trials for Ischemic Cardiomyopathy. With these data in hand, we received approval from the Food and Drug Administration for the PROMETHEUS study (Prospective Randomized assessment Of MEsenchymal stem cell THErapy in patients Undergoing Surgery). The study, funded through the National Heart, Lung, and Blood Institute's Specialized Center for Cell Therapeutics (SCCT), enrolls patients who require coronary artery bypass surgery and who have ejection fractions between 0.20 and 0.50 due to a previous myocardial infarction that caused segmental left ventricular akinesis or dyskinesis. The patient can enter the study if he or she meets enrollment criteria and consents to participate, and if the patient's surgeons and independent clinicians deem it safe to wait 4 to 5 weeks between bone-marrow harvest and treatment. The 4 to 5 weeks are necessary in order to amplify the autologous mesenchymal stem cells with which patients are treated at the time of surgery. The study groups are placebo, low-dose (20-million), and high-dose (200-million) MSC groups. The study has a rigorous design: double-blind, randomized, and placebo-controlled. All patients undergo bone-marrow aspiration and cardiac injection. An independent Data Safety and Monitoring Board monitors the study, under the auspices of the National Institutes of Health.

As mentioned above, the primary endpoint of PROMETHEUS is patient safety. Three specific components of the safety endpoint have been designated: sustained ventricular arrhythmias, ectopic tissue formation, or sudden unexpected death. The secondary endpoint is an efficacy endpoint that has its basis in detailed MRI phenotyping. Regular cardiac MRI serves as the primary method of monitoring the patients over the 18 months that follow surgery. Magnetic resonance imaging enables precise quantitative evaluation of major functional and morphometric values, including global wall motion, regional wall motion, tissue perfusion, and infarct size. The study is being performed at 3 hospitals at the University of Miami and at the Johns Hopkins Hospital, and it is fully funded by the National Institutes of Health through the SCCT mechanism.

The Transendocardial Autologous Cells in Heart Failure Trial (TAC-HFT) is another study that is currently enrolling patients in Miami. Eligible patients are similar to those in PROMETHEUS, with the primary exception being that these patients do not need revascularization, so cells are delivered percutaneously by means of the BioCardia® delivery system. In this study, mesenchymal stem cells are compared directly with autologous whole bone marrow. Although these are the most widely used preparations, they have never before been tested in a head-to-head comparison.

Mechanism of Action. As early-stage clinical studies accumulate, it is imperative to investigate the mechanism(s) of action of cell-based therapy. Although the initial intent of the therapy was to replace lost myocardium, an increasing number of studies in animal models have concluded that the dominant mechanism of action is the release of paracrine factors.[8] In addition, there is support for the potential of neovascularization and collagen metabolism as other mechanisms that might contribute to cardiac repair after cell-based therapy. Although the issue of cell engraftment and differentiation remains controversial, results from our laboratory continue to support a role for cardiomyogenesis and vasculogenesis following MSC injection.[9] Future studies are needed to determine the relative contributions of the various potential mechanisms toward cardiac repair.[10]

Immunoprivilege and Allogeneic Cell Therapy. Another intriguing aspect of mesenchymal stem cells is their immunoprivileged state. Due to the absence of major histocompatibility complex class II and co-stimulatory molecules, MSCs are immunoprivileged and suppress T-cell proliferation, as is shown by their ability to inhibit mixed lymphocyte reactions.[11] In addition, the cells secrete cytokines that are immunosuppressive.[12] However, recent publications[13,14] suggest that although MSCs are immunoprivileged, they are cleared immunologically over a long period of time. This issue has motivated design of the POSEIDON study (PercutaneOus StEm cell Injection Delivery effects on Neomyogenesis), which will use the BioCardia® system to compare an allogeneic preparation of mesenchymal stem cells with an autologous one.

In summary, data from the study of relevant preclinical animal models, together with the emerging safety profile, clearly support the ongoing conduct of trials that test clinical endpoints. The results of these studies will enable the further improvement and refinement of cellular therapy. It is now realistic to predict that pa-

tients could benefit from these therapies in the not-too-distant future, should these early-stage trials provide sufficient support for Phase III efficacy studies.

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