HEART DISEASE IS THE PRIMARY CAUSE OF DEATH WORLDWIDE, PRINCIPALLY BECAUSE THE HEART HAS MINIMAL ABILITY TO REGENERATE MUSCLE TISSUE. MYOCARDIAL INFARCTION (HEART ATTACK) CAUSED BY CORONARY ARTERY DISEASE LEADS TO HEART MUSCLE LOSS AND REPLACEMENT WITH SCAR TISSUE, AND THE HEART’S PUMPING ABILITY IS PERMANENTLY REDUCED. BREAKTHROUGHS IN STEM CELL BIOLOGY IN THE 1990S AND 2000S LED TO THE HOPE THAT HEART MUSCLE CELLS (CARDIOMYOCYTES) COULD BE REGENERATED BY TRANSPLANTING STEM CELLS OR THEIR DERIVATIVES. IT HAS BEEN ~18 YEARS SINCE THE FIRST CLINICAL TRIALS OF STEM CELL THERAPY FOR HEART REPAIR WERE INITIATED (1), MOSTLY USING ADULT CELLS. ALTHOUGH CELL THERAPY IS FEASIBLE AND LARGELY SAFE, RANDOMIZED, CONTROLLED TRIALS IN PATIENTS SHOW LITTLE CONSISTENT BENEFIT FROM ANY OF THE TREATMENTS WITH ADULT-DERIVED CELLS (2). IN THE MEANTIME, PLURIPOTENT STEM CELLS HAVE PRODUCED BONA FIDE HEART MUSCLE REGENERATION IN ANIMAL STUDIES AND ARE EMERGING AS LEADING CANDIDATES FOR HUMAN HEART REGENERATION.


ADDITIONALLY, INVESTIGATORS HAVE STRUGGLED TO EXPLAIN THE BENEFICIAL EFFECTS OF ADULT CELL THERAPY IN PRECLINICAL ANIMAL MODELS. BECAUSE NONE OF THESE INJECTED CELL TYPES SURVIVE AND ENGRAFT IN MEANINGFUL NUMBERS OR DIRECTLY GENERATE NEW MYOCARDIUM, THE MECHANISM HAS ALWAYS BEEN SOMEWHAT MYSTERIOUS. MOST RESEARCH HAS FOCUSED ON PARACRINE-MEDIATED ACTIVATION OF ENDOGENOUS REPAIR MECHANISMS OR PREVENTING ADDITIONAL DEATH OF CARDIomyocytes. MULTIPLE PROTEIN FACTORS, EXOSOMES (SMALL EXTRACELLULAR VESICLES), AND MICRO RNAs HAVE BEEN PROPOSED AS THE PARACRINE EFFECTORS, AND AN ACUTE IMMUNOMODULATORY EFFECT HAS RECENTLY BEEN SUGGESTED TO UNDERLIE THE BENEFITS OF ADULT CELL THERAPY (4). REGARDLESS, IF CELL ENGRAFTMENT OR SURVIVAL IS NOT REQUIRED, THE DURABILITY OF THE THERAPY AND NEED FOR ACTUAL CELLS VERSUS THEIR PARACRINE EFFECTORS IS UNCLEAR.

OF PARTICULAR IMPORTANCE TO CLINICAL TRANSLATION IS WHETHER CELL THERAPY IS ADDITIVE TO OPTIMAL MEDICAL THERAPY. THIS REMAINS UNCLEAR BECAUSE ALMOST ALL PRECLINICAL STUDIES DO NOT USE STANDARD MEDICAL TREATMENT FOR MYOCARDIAL INFARCTION. GIVEN THE UNCERTAINTIES ABOUT EFFICACY AND CONCERNS OVER THE VERSATILITY OF THE MANY UNDERLYING DATA, WHETHER AGENCIES SHOULD CONTINUE FUNDING CLINICAL TRIALS USING ADULT CELLS TO TREAT HEART DISEASE SHOULD BE ASSESSED. PERHAPS IT IS TIME FOR PROONENTS OF ADULT CARDIAC CELL THERAPY TO RECONSIDER THE APPROACH.

PLURIPOTENT STEM CELLS (PSCS) INCLUDE EMBRYONIC STEM CELLS (ESCs) AND THEIR REPROGRAMMED COUSINS, INDUCED PLURIPOTENT STEM CELLS (iPSCs). IN CONTRAST TO ADULT CELLS, iPSCs CAN DIVIDE INDEFINITELY AND DIFFERENTIATE INTO VIRTUALLY EVERY CELL TYPE IN THE HUMAN BODY, INCLUDING CARDIOMYOCYTES. THESE REMARKABLE ATTRIBUTES ALSO MAKE ESCs AND iPSCs MORE CHALLENGING TO CONTROL. THROUGH PAINSTAKING DEVELOPMENT, CELL EXPANSION AND DIFFERENTIATION PROTOCOLS HAVE ADVANCED SUCH THAT BATCHES OF 1 BILLION TO 10 BILLION PHARMACEUTICAL-Grade CARDIomyocytes, AT ~>90% PURITY, CAN BE GENERATED.

PRECLINICAL STUDIES INDICATE THAT iPSC-CARDIOMYOCYTES CAN REMUSCULARIZE INFARCTED REGIONS OF THE HEART (SEE THE FIGURE). THE NEW MYOCARDIUM EXISTS FOR AT LEAST 3 MONTHS (THE LONGEST TIME STUDIED), AND PHYSIOLOGICAL STUDIES INDICATE THAT IT BEATS IN SYNCHRONY WITH HOST MYOCARDIUM. THE NEW MYOCARDIUM RESULTS IN SUBSTANTIAL IMPROVEMENT IN CARDIAC FUNCTION IN MULTIPLE ANIMAL MODELS, INCLUDING NONHUMAN PRIMATES (5). ALTHOUGH THE MECHANISM OF ACTION IS STILL UNDER STUDY, THERE IS EVIDENCE THAT THESE CELLS DIRECTLY SUPPORT THE HEART’S PUMPING FUNCTION, IN ADDITION TO PROVIDING PARACRINE FACTORS. THESE FINDINGS ARE IN LINE WITH THE ORIGINAL HOPE FOR STEM CELL THERAPY— TO REGENERATE LOST TISSUE AND RESTORE ORGAN FUNCTION. ADDITIONAL EFFECTS, SUCH AS MECHANICALLY BUTTRESSING THE INJURED HEART WALL, MAY ALSO CONTRIBUTE.

BREAKTHROUGHS IN CANCER IMMUNOTHERAPY HAVE LED TO THE ADOPTION OF Cell therapies USING patient-derived (autologous) T cells that are genetically modified to express chimeric antigen receptors (CARs) that recognize cancer cell antigens. CAR T cells are the first U.S. Food and Drug Administration (FDA)–approved, gene-modified cellular pharmaceutical (6). THE CLINICAL AND COMMERCIAL SUCCESS OF AUTLOGOUS CAR T CELL TRANSPLANT TO TREAT B CELL MALIGNANCIES HAS OPENED DOORS FOR OTHER COMPLEX CELL THERAPIES, INCLUDING PSC DERIVATIVES. THERE IS NOW A REGULATORY PATH TO THE CLINIC, PRIVATE-SECTOR FUNDING IS ATTRACTED TO THIS FIELD, AND CLINICAL INVESTIGATORS IN OTHER AREAS ARE ENCOURAGED TO EMBRACE THIS TECHNOLOGY. INDEED, THE FIRST TRANSPLANTS OF HUMAN ESC-DERIVED CARDIAC PROGENITORS, SURGICALLY DELIVERED AS A PATCH ONTO THE HEART’S SURFACE, HAVE BEEN CARRIED OUT (7). IN THE COMING YEARS, MULTIPLE ATTEMPTS TO USE PSC-DERIVED CARDIOMYOCYTES TO REPAIR THE HUMAN HEART ARE LIKELY.

WHAT MIGHT THE FIRST HUMAN TRIALS LOOK LIKE? THESE STUDIES WILL PROBABLY EMPLOY AN ALLOGENIC (NON-Self), OFF-THE-Shelf, CRYOPRESERVED CELL PRODUCT. ALTHOUGH THE DISCOVERY OF iPSCs RAISED HOPE FOR WIDESPREAD USE OF AUTLOGOUS STEM CELL THERAPIES, THE CURRENT TECHNOLOGY AND REGULATORY REQUIREMENTS LIKELY MAKE THIS APPROACH TOO COSTLY FOR SOMETHING AS COMMON AS HEART DISEASE, ALTHOUGH THIS COULD CHANGE AS TECHNOLOGY AND REGULATIONS EVOLVE. GIVEN THAT IT WOULD TAKE AT LEAST 6 MONTHS TO GENERATE A THERAPEUTIC DOSE OF iPSC-DERIVED CARDIOMYOCYTES, SUCH CELLS COULD ONLY BE APPLIED TO PATIENTS WHOSE INFARCTS ARE IN THE CHRONIC PHASE WHERE SCARRING (FIBROSIS) AND VENTRICULAR REMODELING ARE COMPLETE. PRECLINICAL DATA INDICATE THAT CHRONIC INFARCTS BENEFIT LESS FROM CARDIOMYOCYTE TRANSPLANTATION THAN DO THOSE WITH ACTIVE WOUND-HEALING PROCESSES.

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The need for allogeneic cells raises the question of how to prevent immune rejection, both from innate immune responses in the acute phase of transplantation or from adaptive immune responses that develop more slowly through the detection of non-self antigens presented by major histocompatibility complexes (MHCs). A current strategy is the collection of iPSCs from patients who have homozygous MHC loci, which results in exponentially more MHC matches with the general population. However, studies in macaque monkeys suggest that MHC matching will be insufficient. In a macaque model of brain injury, immunosuppression was required to prevent rejection of MHC-matched iPSC-derived neurons (8). Similarly, MHC matching reduced the immunogenicity of iPSC-derived cardiomyocytes transplanted subcutaneously or into the hearts of rhesus macaques, but immunosuppressive drugs were still required to prevent rejection (9).

Numerous immune gene editing approaches have been proposed to circumvent rejection, including preventing MHC class I and II molecule expression, overexpressing immunomodulatory cell-surface factors, such as CD47 and human leukocyte antigen E (HLA-E) and HLA-G (two human MHC molecules that promote maternal-fetal immune tolerance), or engineering cells to produce immunosuppressants such as programmed cell death ligand 1 (PD-L1) and cytotoxic T lymphocyte–associated antigen 4 (CTLA4) (10). These approaches singly or in combination seem to reduce adaptive immune responses in vitro and in mouse models. Overexpressing HLA-G or CD47 also blunts the innate natural killer cell–mediated response that results from deleting MHC class I genes (11). However, these manipulations are not without theoretical risks. It could be difficult to clear viral infections from an immunologically stealthy “patch” of tissue, and possible tumors resulting from engraftment of PSCs might be difficult to clear immunologically.

Ventricular arrhythmias have emerged as the major toxicity of cardiomyocyte cell therapy. Initial studies in small animals showed no arrhythmic complications (probably because their heart rates are too fast), but in large animals with human-like heart rates, arrhythmias were consistently observed (5, 12). Stereotypically, these arrhythmias arise a few days after transplantation, peak within a few weeks, and subside after 4 to 6 weeks. The arrhythmias were well tolerated in macaques (5) but were lethal in a subset of pigs (12). Electrophysiological studies indicate that these arrhythmias originate in graft regions from a source that behaves like an ectopic pacemaker. Understanding the mechanism of these arrhythmias and developing solutions are major areas of research. There is particular interest in the hypothesis that the immaturity of PSC-cardiomyocytes contributes to these arrhythmias, and that their maturation in situ causes arrhythmias to subside.

A successful therapy for heart regeneration also requires understanding the host side of the equation. PSC-derived cardiomyocytes engraft despite transplantation into injured myocardium that is ischemic with poor blood flow. Although vessels eventually grow in from the host tissue, normal perfusion is not restored. Achieving a robust arterial input will be key to restoring function, which may require cotransplanting other cell populations or tissue engineering approaches (13, 14). Most PSC-mediated cardiomyocyte cell therapy studies have been performed in the subcutaneous window, equivalent to 2 to 4 weeks after myocardial infarction in humans. At this point, there has been insufficient time for a substantial fibrotic response. Fibrosis has multiple deleterious features, including mechanically stiffening the tissue and creating zones of electrical insulation that can cause arrhythmias. Extending this therapy to other clinical situations, such as chronic heart failure, will require additional approaches that address the preexisting fibrosis. Cell therapy may again provide an answer because CAR T cells targeted to cardiac fibroblasts reduced fibrosis (15).

Developing a human cardiomyocyte therapy for heart regeneration will push the limits of cell manufacturing. Each patient will likely require a dose of 1 billion to 10 billion cells. Given the widespread nature of ischemic heart disease, 106 to 108 patients a year are likely to need treatment, which translates to 1014 to 1016 cardiomyocytes per year. Growing cells at this scale will require introduction of next generation bioreactors, development of lower-cost media, construction of large-scale cryopreservation and banking systems, and establishment of a robust supply chain compatible with clinical-grade manufacturing practices.

Beyond PSC-cardiomyocytes, other promising approaches include reactivating cardiomyocyte division and reprogramming fibroblasts to form new cardiomyocytes. However, these approaches are at an earlier stage of development, and currently, PSC-derived cardiomyocyte therapy is the only approach that results in large and lasting new muscle grafts. The hurdles to this treatment are known, and likely addressable, thus multiple clinical trials are anticipated.

REFERENCES AND NOTES

ACKNOWLEDGMENTS
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