

Attempts to Recruit Stem Cells for Repair of Acute Myocardial Infarction

A Dose of Reality

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EXPERIMENTAL STUDIES SUGGEST THAT MOST OF THE myocardial cells destined to die following an acute coronary artery occlusion will do so within 3 to 6 hours.¹ Early coronary reperfusion with thrombolytic therapy, angioplasty, and/or stenting can certainly salvage ischemic myocardium and improve clinical outcome but late reperfusion will not salvage myocardium. However, there are therapies for which strong evidence has demonstrated benefit even when started beyond 24 hours. Angiotensin-converting enzyme inhibitors and angiotensin receptor blockers reduce the risk of heart failure and premature death, presumably by decreasing progressive left ventricular dilatation and remodeling.^{2,3} Other established and effective adjunctive therapies include antiplatelet agents and statins.⁴

A relatively new and exciting concept for late treatment of acute myocardial infarction (AMI) is that the thin, noncontractile collagenous scar that results after myocyte death could be replaced by viable, contracting myocardium (ie, the damaged heart can be rebuilt). Early work by several groups showed the feasibility of implanting immature cells such as neonatal or fetal cardiomyocytes or skeletal myoblasts into normal or infarcted myocardium.⁵⁻⁹ Using a rat model of myocardial infarction (MI), our research group observed that direct injection of either neonatal or fetal cardiomyocytes 1 week after coronary artery occlusion yielded well-delineated grafts of muscle, which thickened the thin wall of the infarct scar, improved left ventricular ejection fraction, developed a blood supply, and survived for a long period.¹⁰⁻¹²

Unfortunately, use of neonatal or fetal cardiomyocytes is limited by low availability, which has in part stimulated interest in the use of adult stem cells. There has been a virtual explosion of research into the use of stem cells to replace infarcted myocardium over the past 5 years. Adult stem cells derived from bone marrow,^{13,14} adipose tissue, and cardiac tissue have been studied; as have embryonic stem cells.¹⁵ Controversy continues regarding whether adult hematopoietic stem cells derived from bone marrow do indeed truly

transdifferentiate into cardiac tissue.¹⁶ Several small clinical trials have suggested that therapies such as peripheral blood-derived or bone marrow-derived stem cells improve cardiac function after MI.¹⁷

Techniques to deliver these cells include direct intramyocardial injection and intracoronary injection. Another approach to rebuilding myocardial scar tissue is to recruit stem cells to the site of injury (also referred to as homing) with various cytokines such as granulocyte colony-stimulating factor (G-CSF) and stem cell factor. The concept is that these proteins, usually administered intravenously, recruit bone marrow-derived progenitor cells that then target damaged tissue. Several clinical studies using G-CSF in AMI patients suggest improvement in left ventricular function.^{18,19} These studies as well as positive studies using adult stem cells¹⁷ have led to tremendous enthusiasm and hype.

While small trials are initially needed to establish safety and feasibility, many of the trials examining the use of stem cells for cardiovascular disease treatment have limitations that make interpretation of efficacy difficult. These include lack of controls, proper randomization, and blinding; lack of systematic assessment of infarct size or left ventricular function prior to administration of therapy; short-term duration; and small numbers of patients (usually between 5 and 50).^{18,19}

One common problem with some of the uncontrolled trials that have used either cells or protein^{17,18} is the assumption that an improvement in regional and global left ventricular function that occurred over time following coronary reperfusion for AMI was due to the therapy. Studies performed in the late 1970s and in the early 1980s showed that the return of function in the salvaged, viable, postischemic myocardium overlying the infarct could occur naturally following reperfusion of an occluded coronary artery but that this recovery required days to weeks. Recovery of this stunned myocardium²⁰ should not be interpreted as a treatment effect due to cell injection or pharmacological cell recruitment.

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In addition, the size of some infarcts decreases as dead myocardial cells are phagocytized, edema is reabsorbed, and collagen is laid down. The scar tissue may shrink as collagen contracts but this apparent reduction in infarct size is not necessarily due to cell therapy or stem cell recruitment.

In this issue of *JAMA*, Zohlnhöfer et al²¹ report the results of an important study to investigate the effect of stem cell mobilization using G-CSF in patients with AMI who underwent successful reperfusion with percutaneous coronary intervention. In this randomized, double-blind, placebo-controlled trial of 114 patients, G-CSF or placebo was administered subcutaneously for 5 days starting 5 days after AMI. The intended treatment activity of G-CSF was suggested by an increase in peripheral blood CD34+ cells and in white blood cells. Myocardial infarct size and left ventricular function were measured at baseline and at 4 to 6 months after randomization by standard and accepted techniques (technetium-99m sestamibi single photon emission computed tomography and magnetic resonance imaging).

There were no differences in myocardial infarct size between the 2 groups at baseline. This is critical because it suggests that patients with smaller infarcts were not preselected to receive therapy. The main results were that small reductions in infarct size occurred between baseline and 4 to 6 months after randomization in both groups but with no difference between the 2 groups. There were small improvements in left ventricular ejection fraction from baseline to follow-up but again with no group difference. Left ventricular angiography performed at follow-up showed no difference in global or regional left ventricular function between the G-CSF group and the placebo group. Therefore, the study by Zohlnhöfer et al,²¹ unlike many previously reported smaller, noncontrolled, nonrandomized, or unblinded studies or studies that did not assess baseline infarct size prior to therapy,¹⁸ showed no benefit (ie, negative results). On the other hand, the trial did not show any deleterious effects of G-CSF on restenosis, which was suggested by an earlier study.²²

These clinical results are somewhat similar to a preclinical study by Sesti et al²³ that showed no reduction of myocardial infarct size by G-CSF plus stem cell factor in a rat model of MI and no improvement in resting left ventricular ejection fraction. However, in this study contractile reserve of the left ventricle was improved by G-CSF plus stem cell factor in that ejection fraction improved to a greater extent when treated rats were exposed to dobutamine compared with control rats. This benefit occurred without the replacement of scar tissue by muscle tissue and could possibly involve an effect of therapy on noninfarcted tissue or other mechanisms. Zohlnhöfer et al did not specifically test whether contractile reserve might have been improved by G-CSF. In a previous smaller study by Ince et al,²⁴ G-CSF improved both resting left ventricular ejection fraction and ejection fraction during infusion with dobutamine in the

treatment group compared with the control group. Zohlnhöfer et al²¹ provide possible reasons for their negative results compared with previous studies.

Even 3 to 6 hours after AMI, certain therapies still may benefit the heart. However, therapies aimed at recruiting stem cells and regenerating new myocardium remain experimental and have yet to be proven effective in large, long-term multicenter trials in which therapies are administered in a randomized, placebo-controlled, double-blind fashion and the size of the initial MI and baseline cardiac function are taken into account. The study by Zohlnhöfer et al²¹ yielded negative results and some investigators may be disappointed with these results or may try to find fault with the study. However, this investigation is one of the first, controlled, larger, and more carefully designed studies to assess the effect of an attempt to recruit stem cells to an AMI. Additional large, carefully designed trials are needed to assess the true potential (or possibly lack of potential) of stem cell therapy to treat AMI, chronic ischemic heart disease (such as "hibernating" myocardium), and nonischemic dilated cardiomyopathy. Only with such trials will it be possible to differentiate between the hype often generated by smaller, less well-controlled trials and reality.

Financial Disclosures: None reported.

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Thyroid Disease 60 Years After Hiroshima and 20 Years After Chernobyl

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THYROID CANCER WAS THE FIRST SOLID TUMOR reported to be increased in frequency among atomic bomb survivors.¹ Subsequent surveys found a significant excess of papillary thyroid cancer but not of follicular, medullary, or anaplastic cancer.^{2,3} A straight line adequately describes the relationship between radiation dose and thyroid cancer incidence, relative risks are similar in males and females, and age at exposure substantively influences risk. Risk is highest for children exposed when younger than 10 years, and there is no significant increase in risk of thyroid cancer for those exposed after age 20 years. Radiation-induced thyroid cancers are rarely fatal, but the risk per unit dose following exposure in childhood is higher than for any other radiation-induced malignancy.⁴

Radiation-induced thyroid cancer has been extensively studied in human populations other than atomic bomb survivors. These populations include patients treated with radiation for malignant and nonmalignant conditions, populations exposed to radioactive fallout from nuclear weapons tests, and populations living in the vicinity of nuclear installations such as Chernobyl in the former Soviet Union and Hanford in Washington State.^{4,5}

The latest research from the Adult Health Survey (AHS) in Japan reported in this issue of *JAMA* by Imaizumi and colleagues⁶ derives from a cross-sectional study of 4091 survivors of the Hiroshima and Nagasaki atomic bombings in 1945 who were invited to a special thyroid examination more than 50 years later. The prevalences of thyroid cancer (n=87), benign nodules (n=207), and cysts (n=324) were signifi-

cantly increased and directly related to radiation dose, and the risk decreased with increasing age at exposure; however, autoimmune thyroid diseases (positive antithyroid antibodies, antithyroid antibody-positive hypothyroidism, or Graves disease) were not linked to radiation exposure.

There are several new findings of note. First, it is remarkable that a biological effect from a single brief environmental exposure nearly 60 years in the past is still present and can be detected. The radiation doses (mean, 45 cGy) were related to the distance from the hypocenters, and enormous effort was required to accurately estimate exposures for individuals.⁷ The existence of dose-response relationships strongly supports the authors' assertion of a radiation-related excess of thyroid neoplasms nearly 60 years after exposure. Second, using highly sensitive assays to detect serum levels of antithyroid antibodies and thyroid-stimulating hormone, this study was not able to confirm the findings of a smaller but similar investigation of Nagasaki atomic bomb survivors that purported to show an association between radiation and autoimmune thyroid disease.⁸ The absence of a dose-response relationship for any measure of autoimmune disease in the study by Imaizumi et al is consistent with earlier studies of atomic bomb survivors⁶ and a recent study of persons exposed as children to iodine 131 releases from the Hanford nuclear site.⁹

The study of atomic bomb survivors remains the single most important study of radiation effects in humans, but the exposure was brief, lasting less than a second, and thus provides no information on the effects from the prolonged

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