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The next frontier in cardiovascular developmental biology—an integrated approach to adult disease?

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For centuries, clinicians and basic scientists worked side by side in a common quest to identify, understand, and heal the sicknesses of the day. Since the mid-twentieth century, however, a wedge has been slowly driven between these disciplines. Clinicians, constrained by demands incumbent on newly survivable conditions and an ever increasing knowledge base, have become inhibited from participating in basic scientific inquiry, while basic scientists have increasingly pursued more focused studies that result in less clinical translation. These trends are disturbing because of the fundamental need each profession has for the other. One clear example of the interdependent nature of these disciplines is developmental biology. Before the advent of powerful imaging techniques, such as ultrasound and MRI, clinicians were completely dependent on basic scientists to understand embryonic and fetal development.

Cardiac disease remains the number one cause of death from congenital malformations in infancy and accounts for 45–50% of postneonatal deaths due to congenital anomalies in the US.[1] Over the last 30 years, developmental biologists have worked to identify the morphogenic events and mechanisms of cardiovascular development. They have, however, placed much effort on the generation of mutant mice, with the hope of creating embryonically lethal phenotypes that highlight the importance of a particular gene, but which have little clinical utility. Developmental biologists are only now beginning to understand that it is a balance between signaling proteins, transcription factors, and a host of metabolic events, which determines the formation (and homeostasis) of structure. Most importantly, the complexity and balance of signals that affect processes controlling the formation of the heart also come into play in adult life, where their re-expression can be associated with the ability of heart cells to adapt to physiological or pathological stimuli.

Among the key discoveries in embryonic heart development with relevance to adult cardiovascular disease is the process of epithelial-to-mesenchymal transformation (EMT). In this process, epicardial and endocardial (endothelial) cells transform into mesenchymal cells, which then invade the underlying matrix and generate fundamentally different tissues. EMT is a critical event in numerous steps of cardiovascular morphogenesis, including heart valve development, coronary artery formation, and inflow and outflow tract septation.[2] The study of EMT has revealed numerous growth factors, transcription factors, and extracellular matrix proteins, which are vital mediators of cardiovascular morphogenesis, deficiencies in which have been identified as causal of several congenital heart defects. EMT, however, is just the first step in a complex series of morphogenic events that 'remodel' the newly formed mesenchyme into functional, mechanically competent structures (such as septa and valves) while continuously maintaining a unidirectional pumping action in an increasingly demanding hemodynamic environment.

Embryonic development, however, is not the only time that EMT occurs. A number of adult cardiovascular diseases contain aspects of this event, which contribute to pathological tissue remodeling. We recently showed that periostin is a key extracellular matrix protein involved in EMT and subsequent mesenchymal maturation in embryonic heart valves, and that deficiencies in this protein result in the persistence of undifferentiated mesenchyme and uncondensed matrix in adult valve leaflets, resulting in valve insufficiency.[3] Interestingly, the expression of this protein is markedly upregulated in a variety of adult cardiovascular diseases, including vascular injury, dilative cardiomyopathy, and myocardial infarction.[4] Atherosclerosis also bears hallmark signs of EMT, and recent evidence suggests that some of the intimal thickening observed in atherosclerotic plaques is endothelial in origin.[5,6] These plaques contain proliferative and secretory phenotypes typically seen during normal embryonic development, but not in quiescent adulthood. The results of these studies suggest that

the maintenance of the delicate balance of matrix proteins and cell phenotypes in cardiovascular structures is a lifelong labor, and that some of the adult pathology can, therefore, be construed as a recapitulation of developmental phenotypes. Thus, developmental biologists can support clinicians in the diagnosis and understanding of adult cardiovascular disease by connecting pathogenic events to similar embryological events in heart development.

Developmental biologists are also playing an emerging leadership role with major clinical relevance in the discovery of new solutions for the repair or replacement of diseased adult cardiovascular tissues. For example, the application of fundamental developmental biological principles to stem-cell biology could hold enormous potential for regenerative medicine, and is a future direction for clinical cardiology. Bone marrow stem cells—mesenchymal stem cells and hematopoietic stem cells (HSCs)—are attractive targets for regenerative medicine and have been used to populate biodegradable matrices *in vitro*, which were then implanted *in vivo*. After 20 weeks, the explanted tissues not only closely mimicked the complex, trilaminar matrix structure of adult valves, but also differentiated into both endothelial and interstitial-like cells.[7] We recently showed that clonal populations of HSCs injected into lethally irradiated mice populated valve leaflets *in vivo* and differentiated *de novo* into mature phenotypes.[8] These studies suggest that bone-marrow-derived cells can not only be differentiated *in vitro* to recapitulate mature native phenotypes, but also mobilized and recruited *in vivo* to restore function. By definition, the process of stem-cell differentiation is far more a developmental-like than adult-like event, yet bone-marrow-derived HSCs are routinely used in clinical (and basic science) settings with little understanding of what motivates their functional responses.

Besides the studies of EMT and HSCs, developmental biologists have painstakingly mapped out a number of other morphogenic processes and identified their mechanisms of action. So why aren't these 'road maps' being used as a template for tissue regeneration? Again, we believe the answer lies in the divisions that have gradually led physicians to question the applicability of basic science to clinical practice and scientists to criticize clinicians' understanding of biological mechanisms. For example, many developmental biologists tend to look for a single cause of a pathological problem, whereas clinicians know from experience that it is rarely so simple. Heart disease will affect at least half of the US population during their lifetime, and many of these pathologies are likely to have their origins in embryonic development or postnatal adaptation. We propose that understanding the real pathology of an organ, with hope for treatment or repair, will require more than the identification and deletion of simple targets, such as a single gene knockout or knockdown. As developmental biologists, we believe the crux of the issue lies in shifting our focus towards understanding later embryological cardiovascular development—where many malformations have their morphological origins—and the interactions between multiple signaling networks, rather than continue to investigate early events or single genes. We hope that the information gained from these types of studies will be beneficial to clinicians, and that they might be encouraged to collaborate to solve these critical issues and develop regenerative strategies.

Competing interests
The authors declared they have no competing interests.

References

1 Thom T *et al*. (2006) Heart disease and stroke statistics—2006 update: a report from the American Heart Association Statistics Committee and Stroke Statistics Subcommittee. *Circulation* **113:** e85–e151
2 Eisenberg LM and Markwald RR (1995) Molecular regulation of atrioventricular valvuloseptal morphogenesis. *Circ Res* **77:** 1–6
3 Rios H *et al*. (2005) *periostin* null mice exhibit dwarfism, incisor enamel defects, and an early-onset periodontal disease-like phenotype. *Mol Cell Biol* **25:** 11131–11144
4 Katsuragi N *et al*. (2004) Periostin as a novel factor responsible for ventricular dilation. *Circulation* **110:** 1806–1813
5 Xu Q *et al*. (2003) Circulating progenitor cells regenerate endothelium of vein graft atherosclerosis, which is diminished in ApoE-deficient mice. *Circ Res* **93:** e76–e86
6 Ikari Y *et al.* (1999) Neonatal intima formation in the human coronary artery. *Arterioscler Thromb Vasc Biol* **19:** 2036–2040
7 Sutherland FW *et al*. (2005) From stem cells to viable autologous semilunar heart valve. *Circulation* **111:** 2783–2791
8 Visconti RP *et al*. (2006) An *in vivo* analysis of hematopoietic stem cell potential: hematopoietic origin of cardiac valve interstitial cells. *Circ Res* **98:** 690–696