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Perspective

Stem Cells as Vehicles for Youthful Regeneration of Aged Tissues

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Stem cells hold great promise for regenerative therapies for a wide spectrum of diseases and disorders of aging by virtue of their ability to regenerate tissues and contribute to their homeostasis. Aging is associated with a marked decline in these functionalities of adult stem cells. As such, regeneration of aged tissues is both less efficient and less effective than that of young tissues. Recent studies have revealed the remarkably dynamic responses of stem cells to systemic signals, including the ability of "youthful" factors in the blood of young animals to enhance the functionality of aged stem cells. Thus, there is much hope that even aged stem cells retain a remarkable regenerative potential if provided with the correct cues and environment to engage in tissue repair. The overall focus of the presentations of this session is to address the determinants of changes in stem cell functionality with age, the key characteristics of stem cells in aged tissues, the extent to which those characteristics are capable of being rejuvenated and by what signals, and the potential for stem cell therapeutics for chronic diseases and acute injuries in aged individuals.

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AGING is associated with a marked decline in functionalities of adult stem cells, namely tissue homeostasis, repair, and regeneration (1–3). The remarkable advances in the biology of stem cells has shed new light on the cellular and molecular mechanisms by which somatic stem cells generate new differentiated cells throughout the life of an organism and, simultaneously, maintain a pool of stem cells through self-renewal. However, many questions within this field remain to be addressed particularly with respect to the relationship and contribution of the stem cell compartment to the structure and function of adult tissue. Because there is vast diversity among mammalian tissue types, the role of the stem cell compartment within specific tissue types is highly variable (1).

Some mammalian tissues, such as cells of the blood and epithelia of the skin and intestines, engage in constant turnover throughout life, attesting to the essential role of stem and progenitor cells in those tissues for the maintenance of organismal viability. Other tissues, such as the vasculature and skeletal muscle, exhibit far less turnover in the absence of injury or disease but have remarkable regenerative potential when the tissue is damaged. In those tissues, the stem cells are more like reserve cells. The liver represents an organ of endodermal origin with high regenerative potential that appears to be stem cell-independent, whereas pancreas also is derived from endoderm and has little regenerative potential and appears to be essentially devoid of stem cells. The central nervous system is highly stable with little regenerative capacity, properties that contribute to the devastating consequences of strokes and spinal cord injury, but has restricted areas of active stem cell function throughout life giving hope for the potential for stem cell therapeutics. Finally, there are tissues such as kidney and heart which also have extremely limited regenerative potential and in which the very existence of somatic stem cells in those tissues remains a matter of scientific debate. In the face of this diversity, questions arising from the roles of stem cells in tissue homeostasis and repair, directly intersect with the critical issues of the biology of aging (4).

In this session of the Advances of Geroscience Summit held at the National Institutes of Health on October 30th and 31st, 2013, the fundamental relationships between stem cell function and tissue aging were explored. The basic biological questions focused on several aspects of the stem

cell/aging interface. Among the most important questions is whether or not stem cells themselves experience "aging" (2,3). In this scenario, stem cells would be seen to be a "cause" (or at least an origin) of tissue aging, in the sense that they would give rise to differentiated cells within the tissue that are dysfunctional because the intrinsic, detrimental changes in the stem cells themselves would be passed on to their progeny. At the other end of the spectrum is the notion that somatic stem cells retain special biochemical features that protect them from age-related decline. Such features that have been examined include the enhanced DNA repair mechanisms, the expression of telomerase, and the ability to undergo asymmetric cell division to segregate either damaged DNA or damaged protein to the more differentiated daughter thereby protecting the youthfulness of the stem cell compartment (5–10). In this scenario, stem cells would be viewed as preventing tissue aging by serving as a reservoir of "young" cells that could continue to replace the differentiated cells that may be more susceptible to the effects of aging. Beyond these issues of intrinsic changes of the stem cells themselves are the issues of how local ("niche") and systemic factors change with age and influence stem cell function (11–13). Finally, all of these issues are brought into sharp relief in the consideration of diseases of aging. Like age-related changes themselves, it is paramount to understand how stem cells either contribute to or fend off age-related diseases. Likewise, the promise of stem cell therapeutics, already a fundamental basis for the efficacy of bone marrow transplantation and skin grafting, has entered into the discussion of the future of virtually every age-related disease. In this session, this important topic was explored from many angles, including the nature of the diseases that are most amenable to stem cell therapies, approaches that range from enhancing endogenous stem cells to the transplantation of exogenous stem and progenitor cells (including those derived from embryonic stem cells or induced pluripotent stem cells), and the importance of understanding the environment in which endogenous stem cells would be expected to expand or exogenous stem cells would be expected to engraft in order to combat any particular disease. Throughout the discussion, the importance of stem cell biology for healthspan was a central focus, relating to each of the questions, as presented below, that formed the starting point of this critical topic.

What Are the Key Genetic and Metabolic Determinants of Stem Cell Aging and What Are the Relationships to Age-Related Diseases and Organismal Longevity?

This topic addresses the fundamental question as to whether stem cells actually mediate tissue aging as they, themselves, undergo age-related changes thereby generating progeny that are "less youthful" than those in younger individuals, or whether stem cells are the guardians of youthfulness as source of progeny that replace aged differentiated cells with more youthful cells. Clearly, understanding where in fact stem cells exist on this spectrum is critical for understanding tissue aging, age-related diseases, and the potential of stem cell therapies for diseases of aging. This topic was addressed by Dr. Danica Chen (University of California, Berkeley). A major focus was on the role of sirtuins in the regulation of stem cell function (14), one of many genes that are among determinants of organismal longevity and also regulators of stem cell self-renewal and differentiation (15–17). This places stem cells squarely in the middle of the consideration of how metabolic changes may be both a cause and a consequence of tissue aging. In addition, much recent research has examined the role of tumor suppressor genes in age-related changes in stem cell function (18). The implications are that stem cells may be specialized to respond to genotoxic stress and that the development of cancer in tissues may be integrally related to the balance between genomic damage and various tumor suppressor mechanisms, ranging from the molecular to the cellular, along stem cell lineages. These concepts were discussed also in relation to their implications for stem cell therapies for age-related tissue dysfunction and disease.

Do Stem Cells Accumulate Genomic Damage With Age and What Are the Implications for Stem Cell–Based Therapeutics?

As stem cells are responsible for the homeostatic turnover and repair of tissues across the life span, the ability of somatic stem cells to resist the accumulation of genetic mutations and other forms of macromolecular damage is critical to tissue health with age. However, increasing indirect evidence suggests that somatic stem cells are susceptible to various environmental stresses, including oxidative stress and the consequent genotoxic stresses, exhibiting markers of DNA damage that increase with age (19,20). Genetic mutations are likely to occur in quiescent stem cells due to processes similar to those that lead to mutations in postmitotic cells, including the generation of reactive oxygen species from cellular metabolism and the exposure to xenobiotics that may reach stem cell compartments from the systemic milieu. In addition, stem cells are susceptible to replicative stress and the accumulation of replication-induced mutations that accompany DNA replication (21). Dr. Emmanuelle Passegué (University of California, San Francisco) presented evidence of age-related DNA damage in stem cell compartments, and in particular the hematopoietic stem cell compartment, examining the direct and indirect evidence of such genomic instability. Particular attention was paid to cellular activities that sense and repair DNA damage during replication and the importance of replication stress in driving stem cell aging. These processes are clearly of importance with regard to the potential for endogenous stem cells to sustain normal tissue homeostasis

as opposed to giving rise to less functional progeny with age in various tissues (4).

How Does Aging Alter the Local Stem Cell Niche and, if so, Does This Contribute to Chronic Disease Progression

Stem cells respond to and depend upon other cells for support and decision making, either via cell–cell contact or paracrine signaling. This cellular and noncellular environment influencing stem cells is referred to as the niche. Novel research demonstrates that niche aging contributes to the age-related declines in stem cell function (22,23). Interestingly, a more common feature of aged niches seems to be misactivation of stem cells, rather than losing their activation potential toward stem cells (24,25). Without a doubt, local and systemic inflammation is both a cause of age-related changes in stem cell niches, and in the context of hematopoietic stem cells and their niche, likely a cause. Dr. Hartmut Geiger (Cincinnati Children's Hospital Medical Center) discussed recent advances in the biology of niche aging and how this might contribute to the progression of age-related changes or progression of age-related chronic diseases, with particular emphasis on the hematopoietic system.

Is It Possible to Repair Whole Organs, Including Those With Limited Stem Cell Compartments and Limited Regenerative Potential, in an Aging System?

Among the tissues with limited regenerative potential, the approach to combatting age-related changes and age-related diseases are perhaps more formidable (26). The cerebral cortex, the myocardium, and the renal cortex are prime examples. In these tissues, age-related changes can be detected in the postmitotic cells that are their primary constituents. For scientists and clinicians, it therefore poses a unique challenge to the approach to delaying, preventing, or reversing the pathological processes since cellular replacement is not part of the normal biology. In that case, perhaps one approach is to target the local or systemic environment as a way of restoring function to postmitotic cells where age-related changes in proteostasis may be central (27). Building upon the conceptual and technical advances that have resulted from the study of the stem cell/aging interface, particularly in relation to understanding the stem cell/stem cell niche dynamics during aging, experimental results that could potentially lead to novel approaches to tackling important clinical problems in postmitotic tissues have emerged (28,29). At the same time, the importance of even very limited stem cell potential in such tissues warrants attention. Perhaps there is the opportunity to take advantage of endogenous, albeit limited, activities to pursue stem cell therapeutics via transplantation of stem or progenitor cells from diverse sources, and in particular from induced pluripotent stem cells. Dr. Richard Lee (Harvard University) led the discussion on these topics, focusing particularly on the bioengineering challenges of organ repair and on the importance of the interface between stem cell biology and the endogenous physiological and pathophysiological processes that impact stem cell function.

Is Stem Cell Rejuvenation a Likely Candidate for Therapeutics Against Chronic Disease?

To the extent that chronic diseases could be treated by replacement of dysfunctional cells with functional cells of the same tissue, stem cell therapeutics hold promise for age-related diseases. As outlined in previous sections, there is much interest in the age-related changes both of the stem cells themselves as well as the local (niche) and distant (systemic) environments that may negatively influence stem cell function. Recent studies using both physiological (eg, heterochronic parabiosis, ref. 11,30) and pharmacological (eg, rapamycin treatment, ref. 31) approaches suggest that aged stem cell functionality can be rejuvenated, restoring more youthful regenerative potential to aged tissues. These changes speak directly to epigenetic changes in stem cells in the setting of aging and rejuvenation (32). Dr. Irina Conboy (University of California, Berkeley) reviewed studies that bring together these basic findings with important translational and clinical implications. Depending on the method of rejuvenation, enhancement of aged stem cell functionality can, apparently, either be due to restoration of cell intrinsic functions or restoration of niche activity that subsequently enhances stem cell function (33–36). In either case, signal transduction pathways in aged stem cells transition from patterns seen in aged cells to those seen in youthful cells. In the search for systemic factors that may influence stem cell function, proteomic screens have revealed candidates that have been validated in functional assays (12,28). In addition, pluripotent stem cells, perhaps as part of their "youthful" characteristics, appear to produce factors that have beneficial effects on aged tissues and their stem cells (37,38). The scientific advances in this area are among the most important bridges between the study of stem cells and the study of the biology of aging and age-related diseases. Dr. Irina Conboy focused particularly on the notion that cell states (especially that state of "youthfulness") that may be modulated by endogenous, systemic proteins may also be set by small molecules, and that this notion leads directly to therapeutics.

Conclusions

The topics that are encompassed by the framework and questions for this session are of course only a subset of the important scientific and clinical issues that relate to "Stem

Cells and Regeneration" in the context of aging, age-related diseases, and human healthspan. Nevertheless, the breadth of topics overlap with all other sessions in this Summit. In fact, in this summary, the consideration of how stem cell biology pertains to the healthspan and chronic disease touched on the major topic of every other session of the Summit ("Inflammation", "Stress", "Epigenetics", "Metabolism", "Macromolecular Damage", and "Proteostasis"), highlighting the fact that stem cells sit at one nexus of the biology of aging, perhaps representing a critical evolutionary development for somatic maintenance in metazoans. It is clear that understanding basic stem cell biology and how that biology changes with age has great potential in terms of therapeutic dividends from the investment of basic research.

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