Commentary

Genetically Linked Scientists: The One-Two Punch For NFATp Knockout

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The comprehensive report in this issue by Ranger et al. (1) is a landmark contribution for several reasons: first, it identifies an important regulatory agent that functions in several diverse mesenchymal pathways; second, it shows that transcription factors that affect differentiation pathways in adults are not necessarily prominent in embryonic events; third, it documents that alert and inquisitive minds are most effective when their field of vision is not restricted to the narrow limit of their specialized fields; and finally, it demonstrates that collegial and scholarly networks are important in scientific inquiry. In the context of this last point, it is important to point out that although the physician–basic scientist phenotype is being squeezed by fiscal and organizational constraints and is, thus, endangered, the physician-scientists are well positioned as bridging components and are uniquely contributory to the progress of scientific inquiry. As an unusual human interest aside, a behind-the-scenes situation developed in which the molecular immunologist daughter went to her orthopedic surgeon father to help unravel the key features of an experimental system.
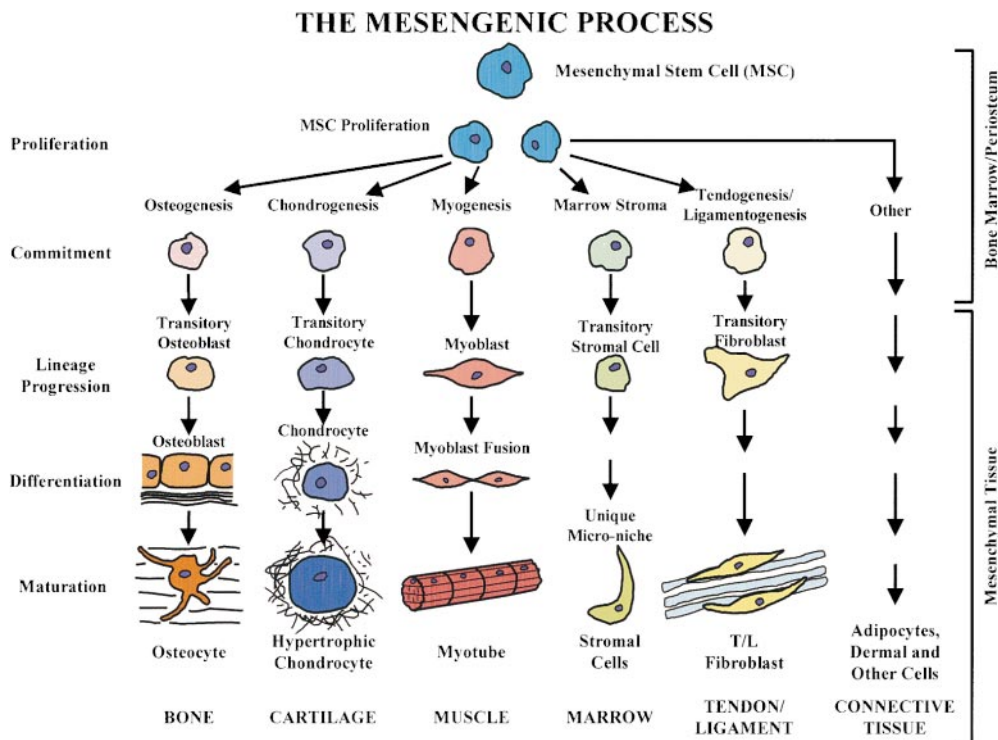
The primary focus of these studies is the nuclear factor of activated T cells (NFAT) family of transcription factors that are regulatory components in T lymphocyte activation in adults (2, 3). Interestingly, one member of this family, NFATc, is active in the morphogenesis of cardiac valves and septum (4, 5), indicating that other members of the NFAT family could be regulators of other mesenchymal tissues. In the 1990s, one powerful way to study a family of regulatory factors is to "knock out" each gene of that family and to study the developmental and physiological consequences. Laurie H. Glimcher's laboratory and others used this strategy (4-10) and, as might be expected, observed that the immune system of the mice was affected. Importantly, new insight into the control of the genesis of a specific class of immune cells was obtained that may have implications for organ transplantation or immune deficiency diseases. In the tedious and often unrewarding process of raising, back-crossing, and breeding these knockout mice, the researchers in Glimcher's lab noticed that the older, breeding, NFATp-deficient female mice had distinctive limps and eventually developed severe ambulation problems. For a laboratory focusing on the details of immune regulation, this side-observation was bothersome but certainly not profound. This ambulation difficulty may have led to naught had Laurie Glimcher, an established T cell molecular biologist (6, 7, 11–14), not been the daughter of Melvin J. Glimcher, a prominent orthopedic surgeon and scientist who has worked on aspects of bone formation and biomineralization for over 40 years (15–19). This fortuitous meeting of two genetically related minds led to an in-depth analysis of the NFATp knockouts as they relate to cartilage and bone turnover and differentiation from adult cells. Upon review of the histological slides of various joints of NFATp-deficient adult mice by the Glimchers, the members of their laboratories, and other orthopedic colleagues in Boston, they concluded that the ambulatory problem arose from an overabundance of cartilage in the joint. Melvin Glimcher hypothesized that this exuberant cartilage growth arose as a result of uncontrolled mesenchymal stem cell (MSC) differentiation and suggested that they contact our laboratory. We provided human (20–22) and mouse (23, 24) MSC preparations in various stages of differentiation to be used for the analysis of NFATp expression. These collegial networks and scholarly interactions led to the identification of NFATp as the first repressing transcription factor regulating adult MSC differentiation into the chondrogenic pathway. Had Laurie Glimcher's lab not been diligent in its pursuit of the cause of the limping in the knockout mice and had these multiinstitutional, multigenerational, and scholarly networks not been available and active, it is possible that this important observation would have been missed.

This late onset, adult cartilage overexpression in the NFATp knockouts reinforces the thesis that this and other laboratories put forth many years ago (25): namely, that a genetically regulated developmental program that controls embryonic events does not cease at birth but continues throughout life, albeit at much slower rates. Thus, postfetal changes in cells, tissues, and organs occur throughout the entire life span of an organism, and are small and occur relatively slowly compared with embryologic events. The fact that 20-yr-old, 50-yr-old, and 80-yr-old organisms are different in tissue morphologies and functions implies that the sequential changes are programmatically controlled. The fact that newborn and neonatal NFATp knockout mice have totally normal skeletal and cartilage structures indi-

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THE MESENGENIC PROCESS

cates that NFATp is not a prominent control element during the preceding developmental period. Thus, the report in this issue by Ranger et al. (1) establishes not only that specific transcription factors tightly control the morphology and function of certain tissues, but also that the regulatory program itself changes with postnatal age, such that NFATp plays a dominant role in adult MSC activities even though it has no apparent influence in embryonic or neonatal mesenchymal events. Thus, transcription factors, in this case one with repressor activity, join the list of molecules that are cassetted into action during distinctive transitions that occur in adult life. For example, the adult skeletal muscle myosin isoform is inserted into functioning contraction arrays long after the embryonic and neonatal isoforms have sequentially appeared (26, 27). Likewise, the posttranslational fabrication of the chondroitin sulfate chains of the cartilage proteoglycan, aggrecan, continually changes throughout life and probably accounts for the decreased resiliency of cartilage and, thus, increased susceptibility to damage with age (25, 28). In this latter case, cartilage doesn't "age," but rather the molecules that are slowly replacing their predecessors (i.e., turnover) are themselves different and less able to effectively structure water, resulting in a progression leading to mechanically compromised tissue.

The factors that regulate turnover and repair in adult tissues may be different from those observed during development. That these events are under genetic control is obvious from observing families that have problems with joint cartilage with age compared with those that rarely have problems. Some of the molecules responsible for these heritable characteristics may be adult-dominant transcription factors like NFATp. In sum, developmental changes continue throughout the life span of the organism, and aging, by itself, is the readout of a slower adult genetic program. Thus, it is clear that the medical complexity of aging will be multifactorial and that cell- and organ-specific cures will require interfaces at many loci.

In retrospect, it also seems logical that the NFAT family of transcription factors would target MSCs, their individual differentiation pathways, and/or their differentiated progeny. Several years ago, we proposed (21) the existence of the multipathway scheme referred to as the Mesengenic Lineage in which adult MSCs could differentiate down a number of phenotypic pathways (Fig. 1). Both hematopoietic stem cells and MSCs are derived embryologically from a common mesodermal progenitor (29). Now it seems that the NFAT family of transcription factors, which has been shown to affect components of hematopoietic pathways, might also influence the other mesenchymally derived tissues, such as cardiac and skeletal muscle, adipose tissue, and now, cartilage. Whether NFATp only targets MSCs for all of these mesenchymal tissues is still an open question; in contrast, the report by Ranger et al. (1) clearly establishes its effect on the adult chondrogenic pathway. The issue now is to translate this molecular discovery into treatment protocols that address aging-related cartilage failure.

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