The Elusive Philosopher’s Stone in Young Blood

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According to legend, soon after the first Chinese Qin Emperor united China around 250 BC, he sent out a troop of young men and women to search for the elixir of life in the eastern seas to extend his life forever. With great expectation and fanfare, the searching party departed but never returned. However, our quests for the elusive life-renewing Philosopher’s Stone have never ceased either in Harry Potter’s wizard world or in biomedical research, and a sighting of the magical rejuvenating power continues to generate excitement and understandably high expectations.

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In 2005, a landmark study by Conboy et al first demonstrated the rejuvenating power of the blood of young animals using a heterochronous parabiosis approach where the circulation of a young and an old mouse was surgically joined together. This finding set off a race to find the putative systemic circulating factor(s) that can reverse aging. Since 2013, in a series of reports, researchers, including Harvard scientists Amy Wagers and Richard Lee, have found that blood from young mice could reverse aging-related pathological features in muscle and brain following a heterochronous parabiosis procedure. In particular, circulating growth differentiation factor 11 (GDF11) was identified as the serum factor responsible for the rejuvenating power in the young blood. These reports generated a wave of commentaries from leading scientific journals and sensational reports from mainstream news outlets, relating these observations to the discovery of the mythical elixir of life given the apparent therapeutic implications for aging-related illnesses. More directly, restoring serum levels of GDF11 with parabiosis procedure. First, GDF11 protein levels in plasma significantly with its homologous protein myostatin because of 90% sequence identity. In fact, the circulating GDF11, when injected intravenously into old mice (2 years of age) relative to the young mice (2 months of age). More directly, restoring serum levels of GDF11 with intravenous injection of a recombinant form of GDF11 protein can significantly reverse age-related cardiac hypertrophy and improve muscle and brain function. It is worth noting that GDF11 is not a panacea for all forms of pathologies. The original Cell report clearly indicated that GDF11 treatment had no effect on pressure-overload–induced pathological hypertrophy in the heart, highlighting the specific antiaging effect of GDF11. However, a recent study published in Cell Metabolism led by Novartis scientist David Glass raised significant questions on these 2 key conclusions with counterpointing evidences. First, they found that the proteomic and immunoblotting methods used by Loffredo et al in the Cell report actually not only detected GDF11 but also cross-reacted significantly with its homologous protein myostatin because of 90% sequence identity. In fact, the circulating GDF11, when measured using a new and more specific anti-GDF11 immunnoassay, could not be detected in the plasma from either old or young mice but showed a trend toward higher levels in aged rats or humans. Furthermore, treatment with GDF11 in vitro or injecting recombinant GDF11 in vivo inhibited myoblast differentiation and skeletal muscle regeneration, associated with similar signaling and molecular signatures as treatment with myostatin. On the basis of these observations, the Novartis group proposed that inhibiting GDF11, rather than enhancing its activity, is actually a potential strategy to promote skeletal muscle regeneration.

In this current issue of Circulation Research, researchers led by Steve Houser from Temple University also report a largely negative observation of the effect of GDF11 on age-related cardiac hypertrophy. Similar to the observations made by the laboratory of Glass, Smith et al found that the anti-GDF11 antibody from Abcam used in the initial Cell report can cross-react with both GDF11 and myostatin. Although another GDF11 antibody from R&D Systems seems to be more specific for GDF11, the detection sensitivity is too low to measure the serum level of GDF11 in mice from either young or old groups. Therefore, the premise of reduced circulating GDF11 as an underlying contributor to the aging process in older animal could not be validated in mice. Furthermore, when circulating GDF11 was elevated by daily injections of a recombinant form of GDF11 for 4 weeks, no significant effect could be detected in the aged mouse hearts in terms of weight, myocyte size, contractile function, and pathological gene induction. These 2 new studies add to another recent report arguing against the validity of the initial claim that GDF11 is a

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circulating factor in young blood responsible for its effect to reverse age-related pathological changes in heart and muscle.\textsuperscript{15-21}

These seemingly contradictory results highlight the need to address some of the key questions raised on the GDF11 hypothesis. Is GDF11 a specific antiaging circulating factor, and if so, how? From earlier genetic analysis, GDF11 and its close homologue myostatin function through a common receptor, activin receptor IIB, and have redundant but not entirely overlapping functions in vivo.\textsuperscript{25} Indeed, a report by Egerman et al\textsuperscript{16} showed that GDF11 and myostatin had a nearly identical downstream signaling pattern and gene expression profile in cultured human skeletal muscle–derived myoblasts. However, there is still a gap in our knowledge about the downstream molecular events specific for GDF11-mediated effects in either cells or intact animals. This concern is compounded by the lack of high-quality antibodies with sufficient specificity and sensitivity to accurately measure GDF11 levels across different animal models and laboratories. One potential approach is to rely on more definitive genetic evidence to prove or disprove the functional role of GDF11 in the aging process. For example, the effect of genetic inactivation of GDF11 on aging should be carefully examined in heterozygous GDF11 null mice or in tissue-specific knockout models.

Is GDF11 sufficient to reverse the aging process in heart, muscle, and brain? This was first demonstrated by intravenous injection for 4 weeks of a recombinant form of GDF11 protein in aged animals.\textsuperscript{2,4} However, there is no uniform quality control or quantitative comparison for the potency of the GDF11 recombinant proteins used in different studies from different laboratories. This issue is of particular importance as GDF11, like myostatin and other transforming growth factor-β family members, is processed via proteolytic maturation into a biologically active dimer form. As indicated in the report by Loffredo et al\textsuperscript{2}, the antiaging effect of GDF11 seems to be dose dependent. For example, only the highest tested concentration of GDF11 was shown to be able to prevent cardiomyocyte hypertrophy in vitro, and elevated plasma GDF11 was dose dependent. For example, the effect of genetic inactivation of GDF11 on aging should be carefully examined in heterozygous GDF11 null mice or in tissue-specific knockout models.

It is important to note that none of these controversies questions the remarkable effect of young blood on aged progenitor cell function as observed in the original heterochronic parabiosis experiment a decade ago.\textsuperscript{1} Clearly, the quest is still on to find the elusive Philosopher’s Stone hidden in the young blood to retard tissue aging via improved regeneration and repair.\textsuperscript{15} Until more studies are performed with more definitive evidence, the controversy and debate about GDF11 in aging is likely to continue.

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Disclosures
None.

References


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