Insulin/Insulin-like Growth Factor Signaling Pathway's Effect on the 4 Biomarkers of Aging

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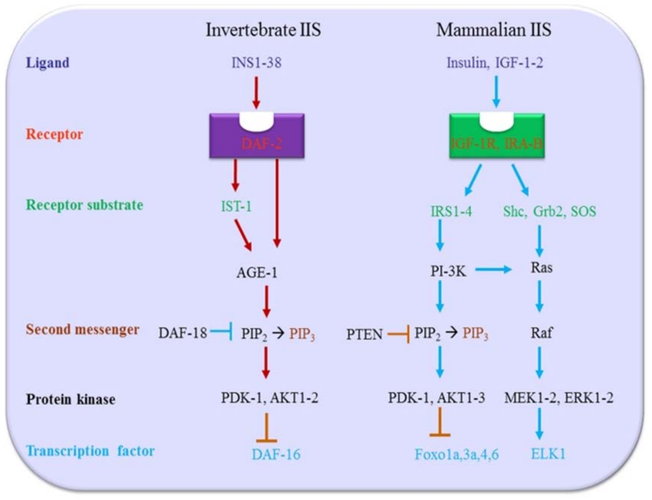
Abstract

Throughout history, humans have demonstrated the innate drive to control both their external and internal environment. However, despite tremendous technological advances in several fields, people cannot control one of the basic portions of their life cycle, i.e. aging. This paradox is a major reason I chose to research the human aging process. Commonly, human aging is studied using animal models, such as roundworms, fruit-flies, and mice, due to the aging process being well conserved from invertebrates to mammalians through the insulin/insulin-like-signaling pathway(IIS). This pathway utilizes insulin growth factor-1(IGF-1) to activate other pathways which then leads to measurable effects on four major biomarkers of aging; skin, the reproductive system, muscles, and the immune system. I reviewed dozens of case studies and scientific journals to better understand the role of the IIS pathway and IGF-1 on the four biomarkers of aging. Although researching aging most likely won't allow us to completely avert the aging process itself, it might give us more insight into the role of health and disease on aging of the body and the mind.

The IIS pathway's main role is activating the signaling of transcription factors that then stimulate growth and development. Researchers use worm and flies to study this pathway instead of humans because of the invertebrates' short lifespan and simpler pathways. The IIS pathway is highly conserved between both invertebrates and mammals. IGF-1 is the initiating step in mammalian IIS. IGF-1 is secreted from inside the liver and through the activation of the AKT pathway via the IIS pathway, it is then established throughout the entire body. The mechanism of action of IGF-1 is that it is the primary mediator of growth hormone 1 (GH). GH is made in the anterior pituitary gland and then is released into the blood stream which then stimulates the liver's production of IGF-1. IGF-1 then promotes systemic body growth.

Breaking down the specificities of the IIS pathway includes several of the following: ligands (molecules that bind to other usually larger molecules), receptors (protein molecules that receive chemical signals from outside the cell), receptor substrates (the substance on which the receptor acts), secondary messengers (signaling molecules released by the cell to trigger physiological changes), protein kinases (a kinase enzyme that modifies other proteins by chemically adding phosphate groups to them), and transcription factors (proteins involved in the process of converting, or transcribing, DNA into RNA).

In mammals there are three different insulin ligands (insulin, IGF-1, and IGF-2) rather than just one (INS1-38 in invertebrates). There are also two different receptors (IGF-1R and IRA-B). After the ligands bind to the receptors, the activated IGF-1/2 (ligand) phosphorylates several intracellular receptor substrates (IRS1-4, Shc, Grb2, and SOS). IRS1-4 Shc, Grb2, and SOS provide docking sites for effectors (cells that act in response to a stimulus) like PI-3K (a regulatory subunit) and Ras (a growth factor). Once docked, the pathway spits into two separate components. Starting at PI-3K, the activation of the kinases protein PTEN occurs. PTEN phosphorylates PIP2 to the secondary messenger PIP3. Elevated level of PIP3 leads to the activation of two more protein kinases known as PDK-1 and AKT1-2 (very similar to what happens in invertebrates). These two proteins lead to the phosphorylation of FoxO1a, 3a, 4, and 6 (transcription factors). Going back to Ras (the growth factor/effector), the activation of Raf occurs. Raf is a protein kinase whose elevated levels leads to the activation of other kinases: MEK1-2 and ERK1-2. These cause the phosphorylation of ELK1 (another transcription factor). The phosphorylation of FoxO1a, 3a, 4, and 6 and ELK1 results in the activation of the AKT pathway; this pathway controls the metabolic and mitogenic effects of IGF-1 signaling.



IIS Related to Skin Aging from UVR

1. IGF-1 (secreted from fibroblasts) regulates the IIS pathway's control of skin cell proliferation
2. Local paracrine signaling affects keratinocyte growth and response to external stressors
3. Fibroblasts create less IGF-1 with age
4. Decreased activation of IGF-1R leads to decreased keratinocytes function
5. Sub-functioning keratinocytes exposed to UVR leads to suppression of DNA repair
6. Without reparative functions, DNA damage occurs
7. The skin becomes damaged and wrinkled

Skin aging from external forces is typically a result of extended ultraviolet exposure. UV radiation (UVR) has long been known to generate photoproducts in genomic DNA that promote genetic mutations, these drive skin aging. IGF-1 levels are shown to be related to skin aging from UVR exposure. Keratinocytes (skin cells that produce keratin) are the most abundant cell type in the epidermis thus are major targets of UVR. Though keratinocytes can respond to UVR autonomously; other proteins can influence keratinocytes' response (i.e. IGF-1 influences keratinocytes). Growth factors like IGF-1 regulate the IIS pathway's control of cell proliferation within general biological cells including the skin. Its local paracrine signaling (meaning it only effects the vicinity around which it is secreted) affects keratinocyte growth and response to external stressors (such as UVR). IGF-1 is related to keratinocyte because keratinocytes express the IGF-1 receptor (the place where IGF-1 binds: for more details look back at the flow chart). Although keratinocytes cannot produce IGF-1, IGF-1 receptors in the skin are stimulated by IGF-1 (created by fibroblasts in the skin) leading to the activation of a variety of intracellular pathways (such as ARK, PI3K, and MAPK).

As a person gets older, fibroblasts, or cells within connective tissue that produces collagen among other fibers, create less and less of IGF-1; this reduction is observed in the skin of 65+ year old patients. Consistent with the idea that dermal production of IGF-1 impacts the activation status of the IGF-1 receptors in keratinocytes, an examination of IGF-1 receptor phosphorylation as a measure of its activation revealed it to be decreased in epidermal keratinocytes of geriatric skin in comparison to that in young adult skin, thus showing a correlation between IGF-1 and aging skin. The reduced production of IGF-1 leads to inactive IGF-1 receptors in epidermal keratinocytes. Exposure of these cells to UVR leads to an inappropriate response: suppression of DNA repair leading to the wrinkling and aging of skin.

Insulin/IGF-1 signaling (IIS) regulates *C. elegans* reproductive aging by modulating multiple aspects of the reproductive process, including embryo integrity (quality of embryo), oocyte fertilizability, chromosome segregation fidelity (how sister chromatids pair in DNA replication), DNA damage resistance, and oocyte and germline morphology (the quality of the structures). Systematic studies on *C. elegans* examined the reproductive aging process from embryonic viability to distal germination in order to determine which steps are susceptible to aging and which are altered in mutants with extended lifespans (both via IIS). The data found establishes that oocyte and distal germline quality correlate well with reproductive success, and that IIS signaling regulates reproductive aging primarily through its control of these two aspects of reproduction. The results also concluded that *C. elegans* with increased levels of IIS signaling were prone to accelerated aging when compared to the longer lived *C. elegans* with decreased levels of IIS signaling.

One may think that comparing worm reproduction and human reproduction is not a viable because worms and humans have vastly different life spans and reproductive strategies, but, the cellular and molecular bases of reproductive span regulation are strikingly similar. Oocyte quality declines with aging (usually after age 40 in humans) and is the major reason for human reproductive capacity decline, resulting in sterility and developmental birth defects. Chromosomal abnormalities, in particular aneuploidies (abnormal chromosome count), are the major defect in human embryos from aging mothers; this increase in chromosomal nondisjunction (mutations) is also seen in *C. elegans*. Another comparison between worms and humans in reproductive aging is oocyte fertilizability, stress resistance, and morphology decline with aging humans as well as aging *C. elegans*, but these declines are delayed by IIS mutants (reduction-of-function IIS).

In worms, genes that are over-regulated by IIS have striking similarities to human oocyte genes that have declined with age, suggesting that many of the molecular mechanisms underlying reproductive cessation are shared between *C. elegans* and humans. The result that IIS (a highly conserved pathway from worms to humans) has a key role in regulating the rate of reproductive aging through utilizing similar mechanisms suggests that IIS is also evolutionarily relevant in humans. Several recent genome-wide association studies of human reproductive development and menopause identified genes that regulate development and longevity in *C. elegans* due to *C. elegans* possessing similar genes as humans. These genes include FoxO3a (transcription factor; shown in flow chart), the human homolog of the DAF-16/FoxO transcription factor downstream of the IIS pathway; this correlation shows another reason why worm reproduction and human reproduction can be compared.

IIS Related to Reproductive Aging

1. Oocyte quality declines with age
2. Through paracrine signaling, IIS controls oocyte quality
3. Increased levels of IIS (which occurs through aging) leads to less production of IGF-1 and thus lowering oocyte quality, influencing fertilization and chromosome maintenance
4. IIS is highly conserved from C. *elegan* to humans (as shown in the flow chart) suggesting that IIS is evolutionarily relevant to human reproductive aging

IIS Regulation of Muscles through Myofibers

1. IGF-1 stimulates the AKT pathway in myofibers
2. The AKT pathway paths a key role in cell proliferation in myofibers
3. Myofibers are crucial in the size and structural architecture of muscles
4. IGF-1 levels decrease with age
5. Less IGF-1 leads to reduced activation of the AKT pathway
6. Reduced activation causes less development of myofibers because of the reduction-function of cell proliferation
7. Muscle atrophy

As humans age, muscles begin to change. Muscle size and architecture are both altered with advanced adult age. An example of this is overall muscle atrophy which occurs in older adults. Further changes in myofibers (multinucleated single muscle cells) include impairments in several physiological domains including muscle fiber activation, excitation-contraction (EC) coupling, actin-myosin cross-bridge interaction, energy production, repair, and regeneration; all of these affect the structure and function of muscles. As aging occurs, body composition changes; this means there is an increase in body fat percentage and a decrease in muscle mass. With a decrease in muscle mass comes a decrease in strength and mobility. All of the changes stated above are factors in why muscle atrophy occurs in older adults.

Local IGF-1's autocrine and paracrine properties result in it being a major controller of muscle tissue growth. Several studies done on mice have shown that mice without IGF-1 have extreme muscle/growth retardation and died shortly after birth. Interestingly, if IGF-1 is introduced to the mice before death, muscle growth is restored. As the control group of mice aged, levels of IGF-1 in muscle tissues decreased proportionately to the increase in muscle atrophy suggesting that there is a correlation between IGF-1 and muscle aging. The levels of IGF-1 is also being studied; the results of increased introduction of IGF-1 to mice has shown to block muscle atrophy in old mice. The effects of introducing more IGF-1 to older mice has not yet been directly studied, however, it is predicted to have similar affects. The over and under-expression of IGF-1 has already been shown to stimulate and reduce the AKT pathway in humans; the AKT pathway is key in cell proliferation of the localized area it resides, the AKT pathway is stimulated by IGF-1's activation of transcription factors, thus why IGF-1 relates to skeletal muscle growth

Dendritic cells (an antigen-presenting cell found in the skin, nose, mouth, and throat) present processed antigen materials to T cells of the immune system. Dendritic cells are considered the most potent professional antigen-presenting cells. Immune system aging is related to IGF-1 and the IIS pathway through these dendritic cells. Localized IGF-1 promotes the activation of AKT signaling in immune cells including dendritic cells.

There are several studies researching the relationship between IGF-1 and dendritic cells through the expression of klotho proteins. Klotho proteins and IGF-1 work closely together. Klotho proteins participate in the regulation of the AKT pathway, the pathway that is activated by IGF-1 and IIS signaling. In one fairly current study, bone marrow from mice was tested with increasing IGF-1 and decreasing IGF-1. The higher levels of IGF-1 resulted in decreased klotho-deficient dendritic cells. Klotho proteins protect dendritic cells from wear and tear thus keeping the youth of the immune system. The mice with reduced IGF-1 resulted in an increase in klotho-deficient dendritic cells; this is because IGF-1 stimulates the AKT pathway, klotho proteins regulate the AKT pathway; if the AKT pathway is never stimulated then klotho proteins remain inactive and unable to bind with dendritic cells of the immune system. With klotho lacking, dendritic cells are vulnerable to degradation thus leading to a suppressed immune system and overall immune system aging.

IIS Regulation of Immunity through Dendritic Cells

1. Localized IGF-1 activates the AKT pathway in dendritic cells
2. Klotho proteins are activated once the AKT pathway is activated
3. Klotho proteins protect dendritic cells from wear and tear
4. IGF-1 reduces with age
5. With less IGF-1 comes less activation of AKT, without AKT activated klotho proteins are unable to bind to dendritic cells
6. Dendritic cells become susceptible to degradation
7. Suppressed Immune system

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