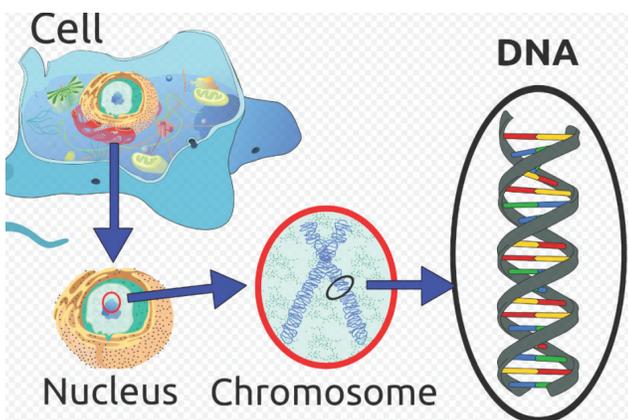


## GENE EFFECTS STUDY – **genexc™** SERUM

### STUDY OBJECTIVE

Genes form the language of every cell's command center. Within the center of every skin cell is the nucleus and its contained language of DNA patterns. Every change that occurs in human tissue begins with a message from the gene directing a specific function. For example, in order to heal a surgical wound in the skin, many genes are activated including those for inflammation, blood vessel formation, immunity, cell division, collagen synthesis, growth factors, skin barrier function, as well as production of many types of required proteins for each of these processes.

For decades it has been known that genes give direct instructions for protein synthesis and the functions of cells. Furthermore, there are many, many genes "talking" to the human body at all times about all kinds of processes. The newer science of epigenetics has explained that this conversation goes both ways – from genes to tissues and from tissues back to genes. Genes can be "turned on" or "turned off" by any number of factors -- from emotional state to sunscreen use. These events, in turn, influence events then controlled by genes.<sup>1</sup>



ANTIOXIDANT GENE MAY BE "TURNED ON" BY WELL-DESIGNED COSMECEUTICAL PRODUCT CONTAINING THE BEST INGREDIENTS

Genes and their ultimate protein products govern critical skin processes related to aging and skin health.<sup>2</sup> Gene expression affected by application of a cosmeceutical product may be measured. Information of this nature would measure when genes were "turned on" or "upregulated" by applying the product to the skin. The ability of a cosmeceutical to "speak" to the skin and improve skin health may be evaluated by advanced laboratory techniques of Qualitative Polymerase Chain Reaction (qPCR). The qPCR monitors the amplification of a DNA molecule in real time. This study determined the ability of GeneXC Serum to speak to skin cells and improve skin health in a variety of ways.

### STUDY DESIGN

Skin was exposed to GeneXC Serum and gene expression was measured. Messenger RNA (ribonucleic acid) was isolated from epidermal cell layers and qPCR methods used to measure gene expression for multiple genes within skin cells. Gene expression panels including 107 target genes and 5 endogenous control genes were analyzed. Control genes are those that influence multiple other genes and are important in overall control processes. Families of genes were grouped according to their specific jobs within skin. Statistical analysis software was used to verify that the resulting data was statistically significant and results were tabulated.

### SIGNIFICANCE OF STUDY

Testing large numbers of genes grouped by gene function provides valuable information regarding biologic functions affected by a cosmeceutical product. Operational data about gene functions "turned on" or "turned off" explains the many ways a skin product may work to affect cells and tissues downstream from the genes. In any biologic process, genes become activated as the first step. All other physiologic events then occur in response to direction given by the genes. Genes do not operate in isolation but are part of the entire holistic milieu of skin. Genes may be influenced by many factors including application of cosmeceutical products.

A skin care product affecting many genes favorably will have widespread positive effects on skin health.

## RESULTS AND CONCLUSIONS

Beneficial skin effects should be expected in the following categories. These categories were discovered according to groups of genes affected by application of GeneXC Serum.

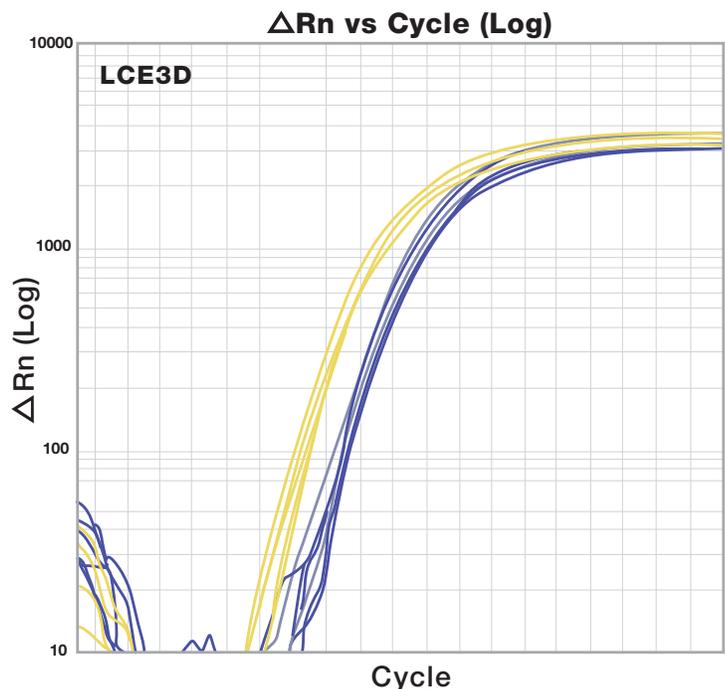
- **ANTI-AGING** – Several known genes are related to how fast aging occurs and the aging process itself. GeneXC Serum “turned on” anti-aging genes, including the very important Forkhead Box O3 (FOXO3). About one percent of epidermal keratinocytes are stem cells but these are responsible for renewal of skin and division into new “daughter” cells. Cell renewal is required after injuries including procedures, as well as cell death from all other causes. With aging, this one percent population of stem cells becomes more fragile and less able to respond to the demands of producing new skin cells. Stem cell renewability should be improved with GeneXC Serum. The FOXO3 gene is important in the process of apoptosis or “programmed cell death”. During apoptosis, an aged cell dies rather than continuing to “take up space” within the skin while doing nothing. It is better for damaged and very aged cells to die and be replaced by young cells with a vibrant metabolism.

- **INCREASED ANTIOXIDANT PROTECTION** – All cells are subject to free radical damage from ongoing cellular metabolism. In addition, skin cells are subject to huge amounts of additional free radical damage due to sun exposure and photoaging. Antioxidants neutralize free radicals and protect cells and tissues from oxidative damage. GeneXC Serum “turned on” antioxidant genes, including Superoxide Dismutase 2 (SOD2). The SOD2 is important in antioxidant protection for mitochondria, the tiny organelles within cells that create energy. One of the critical events of aging is that mitochondrial function and energy creation decline. SOD is a critical part of the body’s own powerful antioxidant system. Only minute amounts of SOD are required to neutralize a very large number of free radicals.

- **IMPROVED HYDRATION** – Skin hydration is essential for healthy skin function. Genes for skin hydration were upregulated by GeneXC Serum.

- **ENHANCED SKIN BARRIER FUNCTION** – An improved skin barrier is indicated. Skin barrier function is important in many ways, including immunity, hydration, solar protection, and others. Ceramide synthesis genes that are essential for a healthy skin barrier and appearance were “turned on”.

Although many positive gene markers were found in this study, an example of gene function increased by GeneXC Serum is given below -- the Late Cornified Envelope 3D (LCE3D) gene () that is important in stratum corneum cornification and skin barrier function. The graph shows that the gene LCE3D is “turned on” by GeneXC Serum. The yellow line indicates the positive response in skin barrier function from product application. The clear separation between the yellow and blue lines indicates the marked improvement in gene function related to product application.



The yellow "Treated with GeneXC Serum" line shows increased gene expression and an earlier appearance of gene function compared to the blue "Control" line without product application. This gene is an important determinant of skin barrier function and is dramatically "turned on". A 772 percent increase in expression of this gene occurred with product application. (See additional results in bar graph shown subsequently.)

- **EXTRACELLULAR MATRIX INTEGRITY** – These gene functions increased. The extracellular matrix or "ECM" is the structural environment in which skin cells are held and proper skin shape is maintained. The integrity of the ECM is controlled by collagen, elastin, hyaluronic acid, and glycosaminoglycans (GAGs).

- **ABILITY TO RESPOND TO STRESS** – Skin cells, in particular, are exposed to stress, including free radical damage from photoaging and cellular metabolism, pollution, toxins and particulates in air, weather extremes, dehydration, and ambient cigarette smoke. The skin's ability to respond to stress should improve related to function of antioxidant and other genes.

- **ENHANCED CELL RENEWAL AND REPAIR** – Abilities of skin cells to renew and repair themselves benefit from GeneXC Serum. Homeostatic mechanisms are processes whereby skin "self-corrects" from damage, injuries, procedures, and changes of aging. These homeostatic abilities are improved as skin renews and repairs itself more easily.

- **ANTI-INFLAMMATORY, ANTI-ITCH, AND ANTI-PAIN** – Results demonstrated strong anti-inflammatory changes. Excess inflammation occurs with aging and all skin diseases and disorders. This product quiets excess inflammation and is soothing. It has strongly anti-itch and anti-pain qualities.

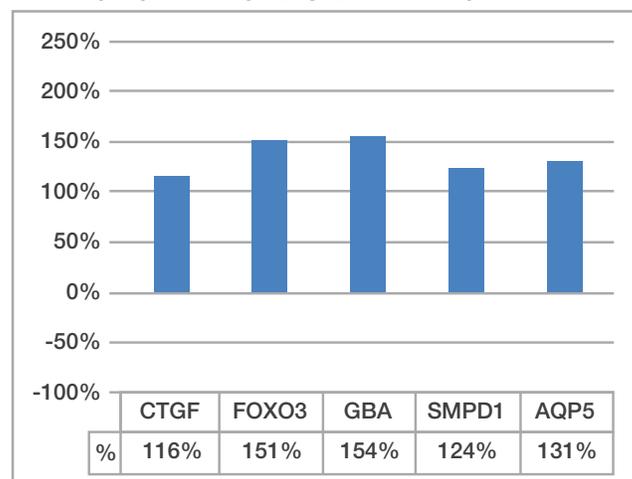
- **ENHANCED IMMUNE RESPONSE** – The enhanced immune response found with GeneXC Serum is helpful, not only in fortifying the skin barrier and protecting

against infections, but also for cancer surveillance and combating the development and progression of skin cancer cells.

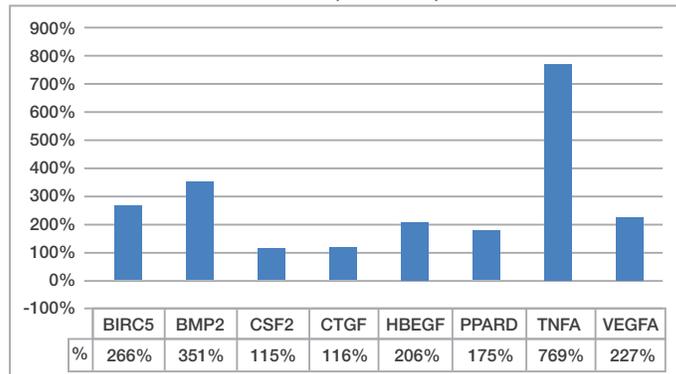
- **BRIGHTENING EFFECT** – Product brightening effects counteract changes of photoaging, hyperpigmentation, and dull skin tone associated with aging.

The bar graphs below are examples indicating percentage change increases in gene functions with application of GeneXC Serum. These are categorized by gene groups important in specific functions of healthy skin.

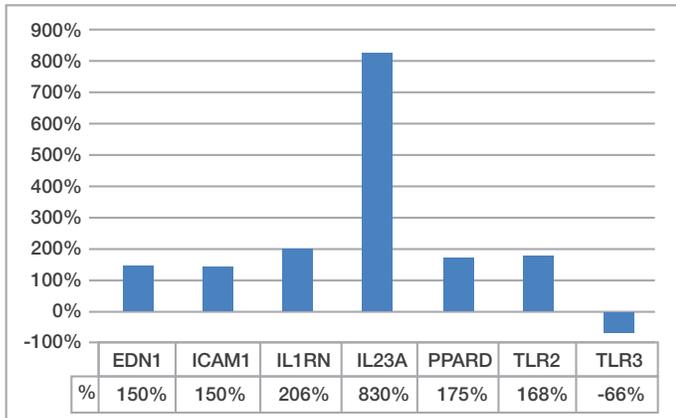
#### ANTI-AGING AND INCREASED HYDRATION



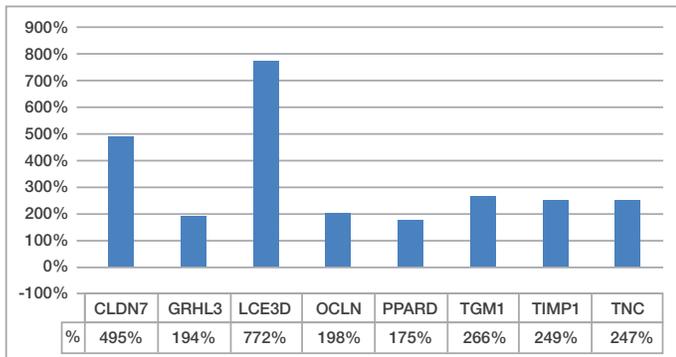
#### INCREASED CELL RENEWAL, REPAIR, AND REGENERATION



### ENHANCED IMMUNE RESPONSE OR ANTI-INFLAMMATORY /-ITCH/-PAIN



### IMPROVED BARRIER INTEGRITY / EXTRACELLULAR MATRIC MAINTENANCE



#### GENE ABBREVIATION KEY

Key to gene abbreviations and functions in bar graphs above.

- AQP5 (Aquaporin 5) – AQP5 is important in skin moisturization and fluid mechanics. It encodes for a water channel protein important in cell membranes for moisturization and water transport.
- BIRC5 (Survivin) – In keratinocytes, this gene is involved in cell cycle growth and proliferative potential. When increased, this is a marker for epidermal homeostasis, metabolic balance, and epidermal survival.
- BMP2 (Bone Morphogenetic Protein 2) – BMP2 belongs to the superfamily controlling transcription

growth factors. In epidermis, this is important in tissue remodeling, regeneration, and wound healing.

- CLDN7 (Claudin 7) – This gene is crucial in skin barrier integrity, permeability and passage of substances between cells in keratinocytes throughout all epidermal layers. Claudin proteins interact with tight junction proteins.
- CSF2 (Granulocyte-Macrophage Colony Stimulating Factor 2) – This gene regulates cytokines involved in wound healing, expression of important extracellular matrix proteins in fibroblasts and a number of other cell types during healing, and immune responses to fight infection, including inhibition of fungal survival. It induces keratinocyte proliferation and differentiation to facilitate wound healing. It is involved in the synthesis of extracellular matrix proteins in fibroblasts for wound healing, including tenascin, fibronectin, and Collagen Type1.
- CTGF (Connective Tissue Growth Factor) – This gene governs the fibroblast regulatory protein that increases with wound healing. CTGF synthesis is impaired with aging and increased CTGF gives anti-aging and healing effects.
- EDN1 (Endothelin 1) – EDN1 is involved in melanin synthesis, melanosome formation, and pain sensation during UV activation. EDN1 encodes for a protein secreted by endothelial cells and is involved in vasoconstriction as well as Epidermal Growth Factor Receptor activation.
- FOXO3 (Forkhead Box 03) – The Forkhead transcription family is important in resistance to stress, longevity, and insulin signaling. This gene locus is very important in anti-aging and anti-senescence. Forkhead Box genes are also important in programmed cell death (apoptosis), inflammation, and cellular proliferation. FOXO3 increases anti-oxidant responses, protects against oxidative stress, increases expression of the superoxide dismutase antioxidant system through SOD2, decreases melanin synthesis via antioxidant mechanisms, and improves keratinocyte differentiation.

- GBA (Glucosylceramidase Beta) -- This locus is important in stratum corneum ceramide synthesis and conversion of sphingolipids to ceramides. This area is important in maintaining a healthy skin barrier and moisturization.

- GRHL3 (Grainyhead-Like Protein 3 Homolog) – This gene codes for a protein important in the development of stratified epithelial cells and epithelial wound repair. It is essential in forming the epidermal barrier. It is required for epidermal reformation following injury and epidermal repair after immune-related damage.

- HBEGF (Heparin-Binding EGF-Like Growth Factor/ Heparin-Binding Epidermal Growth Factor-Like Growth Factor) – This gene encodes for a mitogenic and chemotactic growth factor that is important in fibroblast-to-fibroblast cell signaling during wound healing, repair, and regeneration. One of its identifying chemical properties is that it will bind to heparin.

- ICAM1 (This gene encodes for a cell-surface glycoprotein that is important in immune function. It binds to integrins and is expressed on cells of the immune system and endothelial cells.

- IL1RN (Interleukin 1 Receptor Antagonist) – This gene is strongly anti-inflammatory and anti-aging. It has therapeutic potential early in the healing cascade via messages to macrophages and granulation tissue components.

- IL23A (Interleukin 23 Alpha) – This gene is involved in adaptive immunity and sends messages to Helper T-Cells. It is important in inflammatory and immune responses against infection.

- LCE3D (Late Cornified Envelope 3D) – This gene governs synthesis of the LCE3D protein that is important in formation of the epidermal barrier and its integrity. It governs peptide cross-linking involved in keratinocyte differentiation and cornification.

- OCLN (Occludin) – This gene regulates synthesis of the protein Occludin that is expressed in the epidermal stratum

granulosum layer and regulates communication between these cells. It enhances epidermal barrier repair, especially in response to wounding, UV damage, and chemical damage.

- PPARD (Peroxisome Proliferator-Activated Receptor Delta) – This gene along with several other related genes in its group govern nuclear hormone receptors that decrease inflammation, improve epidermal differentiation, regulate programmed cell death (apoptosis), improve skin barrier function after injury, and are anti-aging. They decrease activation of MMPs (Matrix Metalloproteinase enzymes).

- SMPD1 (Sphingomyelin phosphodiesterase 1) – SMPD1 is important in ceramide synthesis from sphingomyelin. This is important in skin barrier maintenance and moisturization.

- TGM1 (Transglutaminase 1) – This gene codes for the enzyme transglutaminase 1 that is important in cornification and epidermal barrier function. It forms strong cross-links between the structural proteins composing the cornified cellular envelope, providing strength and stability to the epidermis.

- TIMP1 (Tissue Inhibitor of Metalloproteinase 1) – This gene encodes for an inhibitor of the matrix metalloproteinases. It is involved in preservation of the extracellular matrix and is anti-apoptotic. It promotes epidermal proliferation and differentiation.

- TLR2 (Toll-Like Receptor 2) – This gene is a member of the TLR family that is important in inflammatory and innate immune responses. In response to bacterial lipoproteins, it can encourage apoptotic signaling.

- TLR3 (Toll-Like Receptor 3) – This gene is a member of the TLR family that is important in innate immune responses and can sense foreign double-stranded RNAs important in viral infections – including the viral replication process and processes particular to retroviruses.
- TNC (Tenascin C) – This gene encodes for the glycoprotein tenascin that is expressed in the extracellular matrix. It is important in repair during tissue injury.

- TNFA (Tumor Necrosis Factor Alpha) – TNFA has strong anti-inflammatory effects and is anti-aging. It encodes for a protein important in the acute phase inflammatory reaction and inhibition of tumor development.
- VEGFA (Vascular Endothelial Growth Factor Alpha) – The VEGF genes regulate vascular endothelial cells. VEGFA promotes healing, wound repair, and balances collagen deposition during wound repair. It mediates and regulates epithelial and angiogenic growth and migration.

## CONCLUSIONS

It is very unusual for a skin care product to affect this number of genes. These results indicate an active product with high ability to improve skin health and normal skin processes. These changes occurred at the critically important level of gene expression. This is a strongly regenerative product that accelerates normal processes of skin repair.

## REFERENCES

- <sup>1</sup> The genie in your genes. Church D. 2009. Energy Psychology Press: Santa Rosa CA.
- <sup>2</sup> Molecular targets of aging processes. DeHaven C. 2013 May. *Cosmetics and Toiletries*. 128(5):340-348.

---

Study performed by GeneMarkers, LLC