RETINOL+ EMULSION 0.3 DERMAL EPIDERMAL JUNCTION (DEJ) EFFECTS

STUDY OBJECTIVE

Retinol+ Emulsion 0.3 was evaluated for its ability to influence gene expression in the skin, with particular attention to effects on genes pertaining to differentiation, maintenance, and repair of the DEJ.

STUDY DESIGN

Differential gene expression was evaluated 48 hours after an application protocol designed to mimic typical at-home product use. Gene sequencing was performed. Differential gene expression analysis and biological pathway analyses were performed using several bioinformatics software programs.

This laboratory method[1 2 3 4 5] incorporates ribonucleic acid (RNA) sequencing using next-generation sequencing (NGS) to quantify messenger RNA (mRNA). This allows the ability to look at changes in global gene expression patterns over time. This technology offers advantages of higher specificity, sensitivity, and a broader range over previous laboratory technologies such as gene microarrays. This allows for millions of small stretches of genes to be mapped and tracked back to the human genome. Statistical analysis is used to evaluate differentially sequenced genes (DEGs) and their significance with respect to the genome.

Multiple combinations of genes are involved in different biologic processes throughout the skin. These were also statistically evaluated. To be classed as showing significant changes, values were required to reach a statistical significance of $p \geq 0.05$ and a log2 fold change value of 0.06.

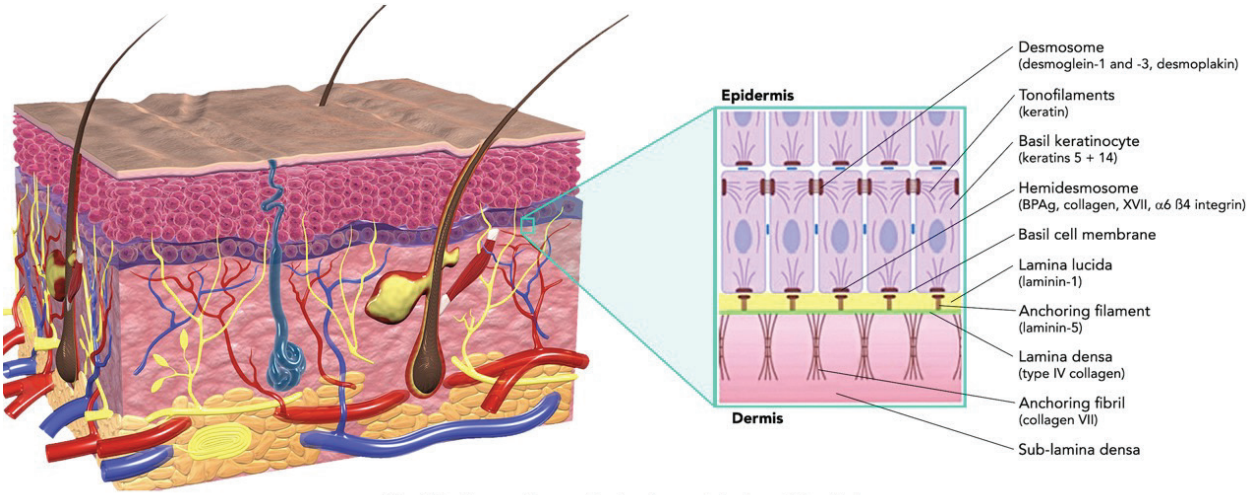
Quadruplicate testing was performed and Control tissues to which normal saline (0.9% saline) was applied were also included in the analysis.

SIGNIFICANCE OF STUDY

A skincare product that improves the maintenance and integrity of the DEJ would have important anti-aging applications and also be important in healing injuries of all types, from surgical wounds to sunburns to skin tears.

The DEJ is an especially important area of human skin. The DEJ attaches to the epidermis above and the dermis below to provide skin integrity and resistance to external mechanical forces.[6]

It is a basement membrane that separates the epidermis from the dermis but is also responsible for adhesion and communication between these 2 parts of the skin. The DEJ anchors basal keratinocytes and the vital stem cells contained within this layer, transmits information from and to cell neighbors, and is a growth factor reservoir.[7] Laminin-5 is an anchoring fibril that fulfills the key anchoring role of the DEJ. Laminin-5 must be intact for the DEJ to function properly in all its roles. The genes coding for this structure were examined in this study.



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The DEJ gradually fragments with age as laminin-5 fails and loses its ability to anchor the epidermis and dermis together. With advancing age, this presents as a commonly seen problem of skin tears.[8] Skin tears occur when a mechanical force is applied to the skin, such as with a fall or scrape, and the epidermis and dermis literally come apart. However, as with almost all problems related to aging, damage occurs gradually and incrementally[9] but also relentlessly over time. Thus, the DEJ of a 40-year-old does not exhibit the strength of integrity of a 20-year-old.

Epigenetic data was used in this study to evaluate the effects of Retinol+ Emulsion 0.3 on the DEJ. Operational epigenetic 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 and can be a very sensitive measurement of product actions. 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.

The practical use of the specific laboratory methods used herein combined with bioinformatics platforms allowed for a broader and more specific evaluation of a topical product's actions on skin than has been previously possible. This yields important and more comprehensive information about the effectiveness and usefulness of a product.

RESULTS AND CONCLUSIONS

The genes LAMB3 and LAMC2 had very high linear fold changes of 4.0 and 5.0 respectively. Retinol+ Emulsion 0.3 epigenetically activated these genes very strongly, consistent with strong messages for maintenance and repair of the DEJ basement membrane.

Laminins are a family of basement membrane proteins and are important in the skin's DEJ for adhesion and attachment. These code for laminin which is a key anchoring structure, fulfilling this role as the DEJ holds the epidermis and dermis together. (see DEJ illustration above) Laminins are the primary noncollagenous components of basement membranes, such as those of the DEJ.[10] They are important in a wide variety of functions, including cell adhesion, differentiation, cell migration, and signaling.[11] The laminin 322 glycoprotein, also known as laminin-5, is composed of 3 chains, each of which is coded for by a specific gene and all of which are necessary for functionality of the protein. LAMB3 encodes for one of these chains in laminin 332[12] and laminin 332 is especially important in the DEJ. LAMBC2 codes for a portion of Laminin 332 that is especially important as an epithelium attachment molecule.[13]

Retinol+ Emulsion 0.3 strongly activates genes involved in maintenance and repair of the DEJ basement membrane. This has important anti-aging and wound healing applications.

REFERENCES

[1]Chu, Y. and Corey, D.R., 2012. RNA sequencing; platform selection, experimental design, and data interpretation. *Nucleic acid therapeutics*, 22(4), pp.271-274.

[2]Finotello, F. and Di Camillo, B., 2015. Measuring differential gene expression with RNA-seq: challenges and strategies for data analysis. *Briefings in functional genomics, 14*(2), pp.130-142.

[3] Wang, T., Zhou, Z., Luo, E., Zhong, J., Zhao, D., Dong, H. and Yao, B., 2021. Comprehensive RNA sequencing in primary murine keratinocytes and fibroblasts identifies novel biomarkers and provides potential therapeutic targets for skin-related diseases. *Cellular & molecular biology letters, 26*(1), pp.1-15.

[4]Whitley, S.K., Horne, W.T. and Kolls, J.K., 2016 Research techniques made simple: methodology and clinical application of RNA sequencing. *Journal of Investigative Dermatology, 136*(8), pp.e77-e82.

[5]Luo, W., Friedman, M.S., Shedden, K., Hankenson, K.D. and Woolf, P.J., 2009. GAGE: generally applicable gene set enrichment for pathway analysis. *BMC bioinformatics, 10*(1), pp.1-17.

[6]Kiritsi D, Has C, Bruckner-Tuderman L. Laminin 322 in junctional epidermolysis bullosa. 2013 Jan 1. Cell Adh Migr. 7(1): 135-141.

[7]Yancey KB. Adhesion molecules. II. Interactions of keratinocytes with epidermal basement membrane. 1995. J Invest Dermatol. 104: 1008-1014.

[8]Fleck CA. Preventing and treating skin tears. Adv Skin Wound Care. www.woundcarejournal.com. 2007 Jun. 20(6): 315-321.

[9]Le Varlet B, Chaudagne C, Saunois A, Barre P, Sauvage C, Berthouloux B, Meybeck A, Dumas M, Bonte F. Age-related functional and structural changes in human dermo-epidermal junction components. J Invest Dermatol. Symposium proceedings. 1998 Aug. 3(2): 172-179.

[10]www.genecards.org. LAMC2 gene. 2023 May 22.

[11]Ibid.

[12]Nat Lib Med, Center Biotechnol Info. LAMB3 laminin subunit beta 3 (Homo sapiens). Gene ID 3914. 2023 June 21.

[13]www.genecards.org. Op cit.

DISCLOSURES

Study performed at Genemarkers, LLC.

Study type: Gene Activation

Study summarized by Charlene DeHaven MD, Clinical Director, Innovative Skincare®