Exploratory study to examine the gene expression effects of retinol on aging-related molecular pathways in individuals of East Asian descent

Linna Guan[1,2], Ludan Zhao[1], Jin Xu[1], Rui Li[1], Dale Kern[3], Helen Knaggs[3], Anne L.S. Chang[1]

[1]Department of Dermatology, Stanford School of Medicine, Redwood City, CA. [2]Case Western Reserve University School of Medicine, Cleveland, OH. [3]NuSkin Enterprises, Provo, UT.

Introduction

Topical vitamin A (retinol) and its analogs have long been used to improve the appearance of photo-aged skin and to decrease fine wrinkling in skin of older individuals. *Kafi et al.* demonstrated that topical 0.4% retinol applied for up to 3 times per week was able to induce clinically significant differences in fine wrinkling of sun-protected skin in as little as 4 weeks. However, to date, it remains unclear whether retinol merely reduces the appearance of skin aging features through structural changes or whether classic aging-related molecular pathways may also be affected.

The objective of our study is to use unbiased whole transcriptomic analysis of sun-protected skin to identify molecular pathways that may be altered by topical retinol.

Study Methods

After IRB approval and written consent, 100 women of East Asian descent between ages 55-75 were recruited. Patient screening, selection, and treatment assignment is shown in the "Study Outline" flowchart. RNA sequencing (RNA-seq) was performed on arm skin biopsies from patients at the end of the study. Pairwise comparisions were made between RNA-seq results from retinol and vehicle treated skin of each patient and differentially expressed genes (DEGs) were identified. Pubmed litereature search was conducted to identify protein encoding genes that have previously been reported to be involved in aging. Results of our literature search are shown in the venn diagrams*. Gene ontology (GO) terms were also applied to DEGs to further examine biologic themes.

References

1. Kafi R, et al. Improvement of naturally aged skin with vitamin A (retinol). Arch Dermatol. 2007 May;143(5):606-12.
2. Shao Y, et al. Molecular basis of retinol anti-aging properties in naturally aged human skin in vivo. Int J Cosmet Sci. 2017 Feb; 39(1): 56–65.

Financial Disclosures

Funding for this study was provided by NuSkin Enterprises. DK and HK are employees of NuSkin Enterprises. LG, LZ, JX, RL, and ALSC have no other relevant financial relationships to disclose.

RESULTS

Study Outline

Recruitment of 100 women ages 55-75 of East Asian descent in compliance with IRB guidelines.

↓

Collection of high quality digital photography and demographic data of all 100 women.

↓

Selection of 11 participants with the most fine wrinkles and 10 participants with the least fine wrinkles of the face based on analysis of digital photography by two expert dermatologists.

↓

Selected participants were randomly assigned to apply 0.4% retinol cream to one sun-protected arm and vehicle cream to the contralateral arm daily for 12 weeks.

↓

Collection of punch biopsy skin samples from both arms for RNA-Seq.

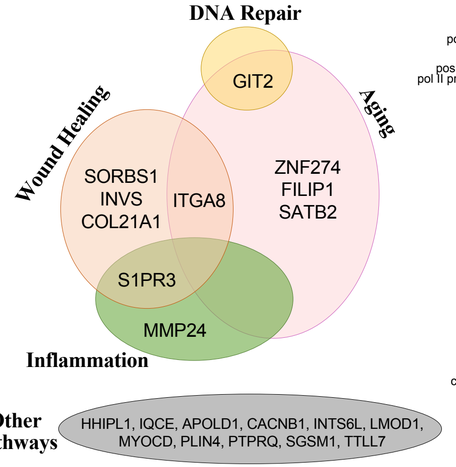
↓

Pairwise comparisons of RNA-seq results were made to generate a list of DEGs in retinol versus vehicle treated skin. GO term analysis of the top 100 up and downregulated DEGs.

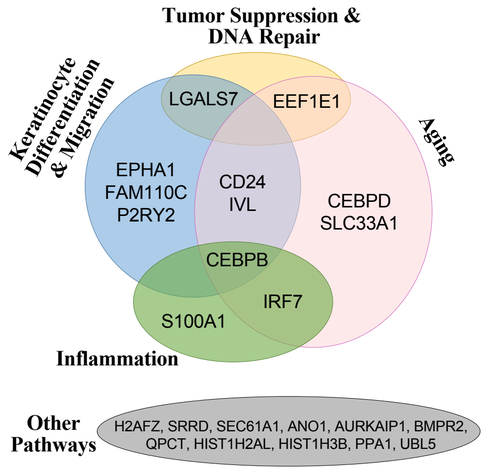
Patient Demographics

	ALL OLDER ADULTS (N= 100)	LESS FINE WRINKLES (N=10)	MORE FINE WRINKLES (N=11)
AGE AT VISIT, MEDIAN (RANGE)	64 (55 - 74)	67 (61 -75)	66 (57-63)
AGE AT VISIT, MEAN (RANGE)	63.7 (55 - 74)	67.4 (61 -75)	64.8 (57-63)
BODY MASS INDEX (KG/M²), MEDIAN (RANGE)	22.3 (17.3 - 34.6)	21.8 (19.4 -35.9)	23.2 (19.9 -32)
CUES SCORE, MEDIAN (RANGE)	$3.6x10^5$ ($1.2x10^5$ – $1.3x10^6$)	$5.0x10^5$ ($1.3x10^5$ – $1.1x10^5$)	$5.4x10^5$ ($1.8x10^5$-$7.3x10^5$)
HISTORY OF SKIN CANCER, N(%)			
NO	97 (98)	10 (100)	11 (100)
YES	2 (2)	0 (0)	0 (0)
FITZPATRICK SKIN TYPE, N(%)			
2	4 (4)	1 (10)	0 (0)
3	46 (46)	3 (30)	8 (73)
4	45 (45)	6 (60)	2 (18)
5	3 (3)	0 (0)	0 (0)
6	1 (1)	0 (0)	1 (9)

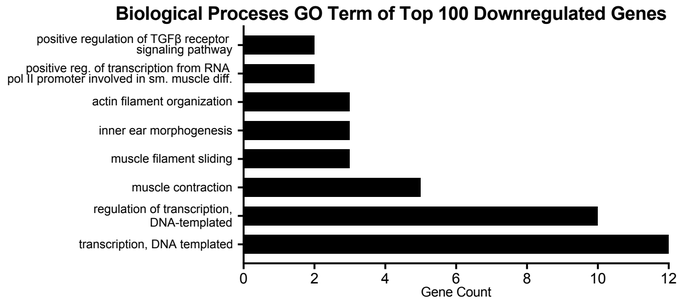
Downregulated DEGs

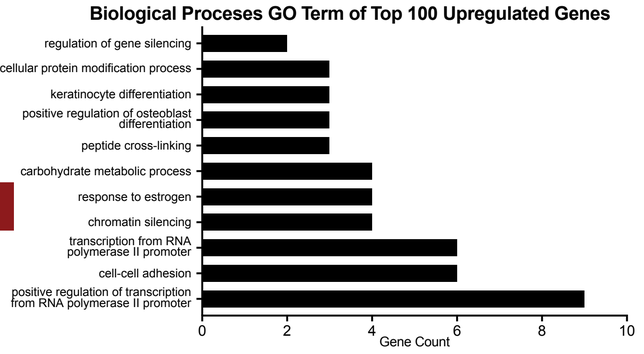


Upregulated DEGs



Gene Ontology Term Analysis





Results Summary & Discussion

- In total, 55 DEGs were identified ($p<0.01$) in retinol versus vehicle treated skin.
- Of these, 12 genes have been reported to be associated with aging.
- GO term analysis revealed that top biologic processes include transcription regulation and cell-to-cell adhesion.

These data suggest that topical retinol may have effects beyond reducing the appearance of wrinkles. Our study provides clues as to potential anti-aging pathways affected by retinol and merit further study.

*Genes that were differentially expressed but whose function has not been well-demonstrated in literature and which are not represented in the diagrams include 8 downregulated genes (AL359075.1, RBFADN, ANKRD20A7P, LINC00637, AL132989.1, RC3H1-IT1, PRRT3-AS1, LINC00393) and 5 upregulated genes (AC134312.3, AL162274.2, AC005288.1,)