

Optimera[®] firming body contour cream contains SIG-1273[™], a molecule with cell lipolysis and anti-aging properties that clinically promotes skin firmness

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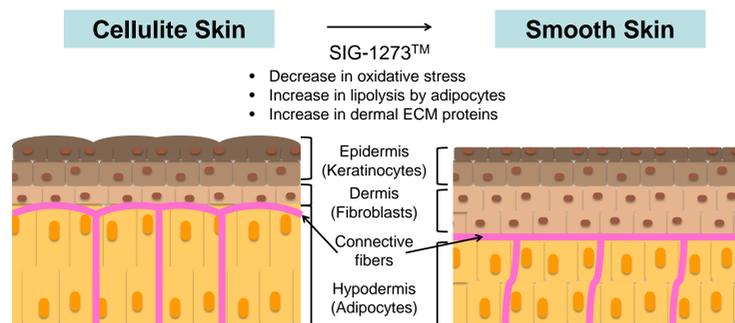
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Abstract

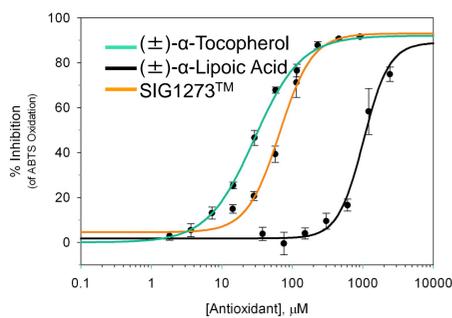
Cellulite is a cosmetic condition characterized by an increase in the subcutaneous adipose tissue along with decrease in the dermal matrix. Although genetic factors have been suggested to predispose individuals to cellulite development, common therapeutic approaches target lipid metabolism. It is known that isoprenylcysteine (IPC) compounds can modulate the intracellular signal transduction activity of G-proteins and modulate gene expression in many skin cell types. Therefore, we tested IPC analog and cosmetic ingredient, SIG-1273[™] for antioxidant and lipolytic properties required for the reduction of cellulite formation. SIG-1273[™] exhibits antioxidant activity in dermal fibroblasts and also demonstrates lipolytic properties *in vitro* by increasing glycerol release in differentiated human adipocytes in a concentration-dependent fashion with maximal stimulation at 30-100 nM. SIG-1273[™] was applied to 3D human skin cultures to assess the effect on key aging and cellulite gene markers topically. Results show SIG-1273[™] potentially targets basement membrane structural integrity by increasing collagen IV and dermal matrix remodeling by enhancing hyaluronic acid synthase-2 gene and protein expression. In addition, SIG-1273[™] stimulates HIF1a gene expression, which has been suggested to help mediate the hypoxic environment created by cellulite. Lastly, SIG-1273[™] as part of the Optimera[®] Firming Body Contour Cream formulation reduced the appearance of cellulite and improved the appearance of upper thigh and skin tone contours at 2, 4, 8 and 12 weeks of use among clinical study participants. Moreover, improvements were observed in firmness and elasticity as determined by an expert clinical grader beginning after only 2 weeks of use.

Fig 1. SIG-1273[™] targets cellulite skin



Skin cellulite is triggered by enlargement of hypodermal adipocytes stressing surrounding connective fibers exerting force within the skin. This promotes the formation of uneven skin surface or dimpling effect. Cellulite is more prevalent in the skin of thighs, hips, buttocks and abdomen of adolescent and adult women. SIG1273[™], has been shown previously to be a novel Cosmetic Functional Ingredient (CFI) with multiple actions including anti-inflammatory in dermal and epidermal cells with anti-microbial activity. In this study, we tested SIG1273[™] for antioxidant and lipolytic properties in human fibroblasts and differentiated adipocytes, respectively.

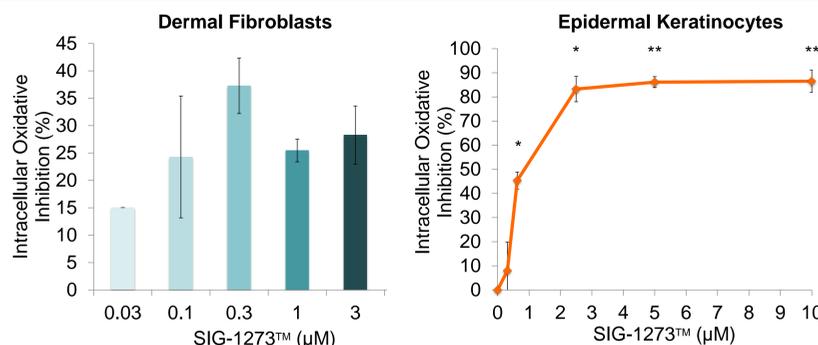
Fig 2. SIG1273[™] is a cell-free ROS scavenging antioxidant



Compound	IC ₅₀ ^a (µM)
SIG-1273 [™]	68 ± 8
α-Tocopherol	25 ± 0
α-Lipoic Acid	682 ± 169

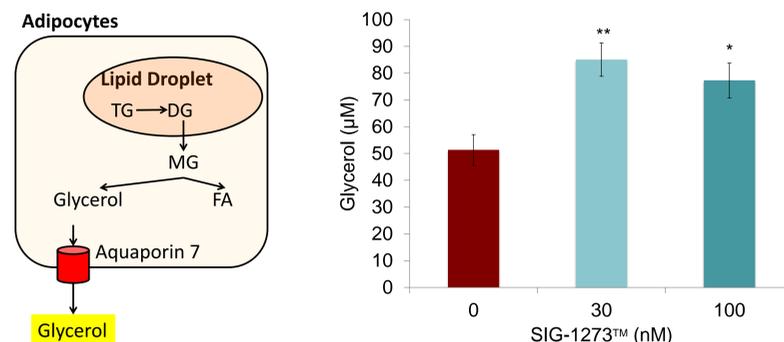
Total antioxidant capacity of compounds was measured by the oxidation inhibition of ABTS^{•+} (2,2'-Azino-di-[3-ethylbenzothiazoline sulphonate]) to ABTS^{•+} by metmyoglobin. The amount of ABTS^{•+} produced was monitored by reading the absorbance at 750 nm. All compounds were tested at the concentrations shown. The IC₅₀ is the concentration of compound producing half maximal inhibition.

Fig 3. SIG-1273[™] demonstrates antioxidant properties in skin cells



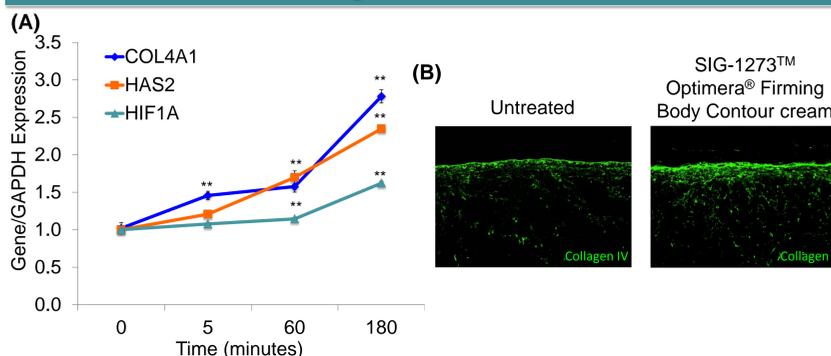
Primary Human Dermal Fibroblasts (HDFs) or epidermal keratinocytes (NHEKs) were incubated with SIG-1273[™] (≤ 10µM) for 3 hours. Control cells received vehicle-only. Intracellular oxidative activity was determined using DCFH-DA marker and hydrogen peroxide as inducer of oxidative stress. * p value ≤ 0.05; ** p value ≤ 0.01 by Student *t* test compared to H₂O₂-only treated cells.

Fig 4. SIG-1273[™] lipolysis in human adipocytes



Effects of SIG-1273[™] on glycerol release by human adipocytes. Differentiated human adipocytes were incubated with SIG-1273[™] (30-100 nM) for 24 hours. Control cells received vehicle-only. Glycerol concentrations in the culture medium were determined using a colorimetric assay. * p value ≤ 0.05; ** p value ≤ 0.01 by Student *t* test compared to untreated tissues. Triglyceride (TG); Diglyceride (DG); Monoglyceride (MG); Fatty Acid (FA).

Fig 5. Optimera[®] cream containing SIG-1273[™] activity in 3D human skin model



Human Reconstituted Skin (EpiDerm-FT[™]) cultures at the air-liquid interface were topically treated with SIG-1273[™] formulated in Optimera[®] cream for 3 hours or 7 days for gene expression and immunohistology analysis, respectively. (A) Total RNA was harvested from tissues and converted to cDNA. Subsequently, gene expression was measured by qPCR using GAPDH as control. (B) After 7 days, tissues were fixed in formalin, paraffin embedded and stained with primary anti-Collagen IV and Alexa Fluor[®]-488 tagged secondary antibodies. ** p value ≤ 0.01 by Student *t* test compared to untreated tissues.

Fig 6. Clinical Study

Study Design	
Enrollment	
<ul style="list-style-type: none"> ≥40 women, ages 35-65 with cellulite on the upper thigh area Subjects who were current users of any slimming or cellulite improvement product, were excluded 	
Procedure	
<ul style="list-style-type: none"> Apply product once daily into each targeted area (upper thigh, front and back, below buttocks) in a circular motion for a minimum of 20 seconds 	
Endpoints (BL, W2, W4, W8, W12)	
<ul style="list-style-type: none"> Expert Clinical Grade Evaluation Instrumental Evaluation: Caliper and Cutometer[™] Subjective Questionnaire 	

Table 1. Clinical study Demographics

Characteristic	Completed (n=42)
Age Range	51.2 ± 6.9 36 – 64
Fitzpatrick skin type (I-VI)	I – 1 III – 11 IV – 13 V – 14 VI – 2
Body skin type	Combination – 1 Dry – 17 Normal – 23 Very Dry – 1

42-female subjects completed a 12-week monadic study using the Optimera[®] Firming Body Contour cream formulated with SIG-1273[™] to determine the effects on the improvement of the appearance of cellulite. Cream was applied once daily on the upper thigh area. Assessments occurred at Baseline (BL), and at Weeks 2, 4, 8, 12 and included expert clinical grading, non-invasive instrumental evaluation and photographic documentation.

Fig 7. Optimera[®] cream containing SIG-1273[™] improves appearance of skin cellulite

Table 2. Clinical Grade Evaluation from Week 2-12

Assessment Measured	Mean % Improvement from Baseline
Thigh Contour	21 - 49%
Skin Tone	16 - 42%
Firmness	21 - 48%
Elasticity	19 - 42%
Cellulite	6 - 44%



Results revealed significant improvement from Baseline (BL) in mean scores for the appearance of thigh contour, skin tone (tactile), firmness, elasticity (tactile) and cellulite after 2-12, weeks of test product use. No significant changes from Baseline were observed in subjects' mean weight at any visit. Standardized photographs of the upper thigh area were taken at baseline and after 2 weeks application of Optimera[®] Firming Body Contour cream containing SIG-1273[™].

Summary/Conclusions

- SIG-1273[™] displays antioxidant properties by inhibiting ROS scavenging activity in a cell-free system and intracellularly in both human keratinocytes and fibroblasts.
- SIG-1273[™] demonstrates lipolytic properties by increasing glycerol release in differentiated human adipocytes in a concentration-dependent fashion.
- SIG-1273[™] formulated in Optimera[®] Firming Body Contour cream produced positive effect in the expression of both skin aging (COL4A1, HAS2) and anti-cellulite (HIF1A) related genes.
- SIG-1273[™] in an existing topical formulation (Optimera[®] Firming Body Contour cream) tested in a clinical study shows significant improvement in the appearance of upper thigh cellulite, elasticity and firmness as compared to baseline.
- Overall, the SIG-1273[™]-containing Optimera[®] Firming Body Contour cream formula provides beneficial effects to skin as it generates significant improvements in anti-cellulite related activity.