

## SkinTone

**INCI Nomenclature:** Yeast Extract **Suggested Use Levels:** 2 – 5 % **Suggested Applications:** Anti-wrinkle, Toning, Elasticity

Communication is key! How often have we heard this phrase uttered in reference to our personal or work relationships?

Yet one would rarely thinks to apply such a phrase to other situations. However effective communication amongst the various cells of our body is vital for our survival, and also dramatically affects our appearance.

The epidermis is the outer layer of our skin; underneath the 5 layers of the epidermis is the dermis, which provides oxygen and nutrients that are needed for maintaining the epidermis. Anatomically the site at which both the dermis and the epidermis meet is referred to as the dermal epidermal junction (DEJ), and it consists of an area approximately 100 nm thick. DEJ integrity is vital for the communication that occurs between the dermis and the epidermis, and it plays a role in numerous processes including cellular differentiation, migration, proliferation and repair. The DEJ is also involved in immune system responses such as the inflammatory



response mechanism, which is triggered via chemical signals relayed across the dermal epidermal junction. SkinTone is intended to improve the integrity of the DEJ by increasing the synthesis of its components; thus improving the overall appearance of the complexion by increasing tone and elasticity while decreasing the appearance of fine lines and wrinkles.

The dermal epidermal junction is predominantly constructed out of collagen types IV and VII as well as glycoproteins such as integrin 2 1, laminins and other proteins. Many hypothesize that the condition of the DEJ directly affects the appearance of the epidermis, and its condition is responsible for wrinkle formation, elasticity and tone. Therefore if one were to create a cosmetic to target the DEJ, it would have to effectively improve the various components that make up the DEJ. SkinTone is intended to improve the DEJ by increasing the production of collagen IV and VII as well as integrin 2 1. We have isolated a specific peptide sequence from yeast that may actually increase the production of collagens IV and VII while improving the concentration of various glycoproteins.

The efficacy of SkinTone was verified by several in vivo and in vitro studies. A 15 subject panel of women between the ages of 40 and 55 was constructed to determine the effects of SkinTone on epidermal characteristics such as elasticity, tone and fatigue. The results indicated that SkinTone may actually improve elasticity and tone while minimizing epidermal fatigue. As a result SkinTone may be added to anti-aging products designated to reduce wrinkle appearance and formation.



SkinTone

An in vitro analysis involving fibroblast migration revealed that SkinTone increased fibroblast migration to wound sites after 24 and 48 hours. The study involved the use of fibroblasts cultured in fibroblast-cultured medium and grown on glass slides. Cultures were wounded and incubated with either 0.5% SkinTone, TGF- 1 and the control. Fibroblast migration was then observed with a compound light microscope.

SkinTone was also effectively used to increase the synthesis of collagen IV, collagen VII and integrin 2 1. Fibroblasts were incubated with 2% SkinTone, or TGF- 1 and the control for 72 hours. Reverse mRNA transcriptase and PCR were used to identify the coding region for each protein. The concentration of the coding sites is expressed with the intensity of the bands produced via PCR. SkinTone was observed to increase the concentration of the coding sites of collagen IV, collagen VII and integrin 2 1. Therefore it may be



used to increase synthesis rate of collagen IV, collagen VII and integrin 2 1.

SkinTone may be incorporated into lotions, creams and gels that are intended to act as anti-aging products as well as other products that are intended to reduce the appearance of physical damage. The various in vitro and in vivo studies performed on SkinTone reveal that it may be used to target the DEJ and improve the overall appearance, tone and elasticity of the epidermis. Since it is a fermented yeast extract that is water-soluble so it may be used in virtually any aqueous system.



## SkinTone

Specification	Parameter
Appearance	Clear to Slightly Hazy Liquid
Odor	Characteristic
Color	Yellow
рH	6.0 – 7.0
Solids (1g – 1 hr- 105ºC)	3.0 – 4.2%
Sugar Content	2.2 – 3.5%
Microbial Content	< 100 opg no pathogens

Compositional Breakdown:

%
92.00 - 96.00
3.50 - 5.50
0.45 – 0.55
0.05 – 0.15

To our knowledge the above material is free of materials classified as CMR in accordance with the Directive 2004/93 of 21 September 2004.



SkinTone

Abstract:

The efficacy of SkinTone was determined by measuring changes in epidermal characteristics such as elasticity, fatigue and tone before and after a 28 day treatment with a Carbopol gel containing 5% SkinTone.

Materials and Methods:

A six subject panel of women between the ages of 32 and 58 was asked to apply a mixture containing 5% SkinTone to their forearms twice daily for 28 days. Subjects abstained from using products on the test site prior to analysis. The SEM 575 Cutometer was used to non-invasively quantify modifications in epidermal elasticity via suction. The process began by placing the suction probe in contact with the skin for a period of 5 seconds, this was then repeated 5 times consecutively. Sensors at the tip of the probe measured the amount of epidermis drawn into the probe to determine the structural integrity of the epidermis e.g. tensile property. All testing was performed in conjunction with a placebo.

Recorded values pertinent in determining the efficacy of SkinTone include: the initial contortion (IC), delayed contortion (DC), tensility (T=IC + DC), final tensility (T'), epidermal recoil (ER), secondary recoil (mean of cutometer measurements taken 5 times consecutively)(ER').

The following formulas were used to analyze results:

Toning:

%Δ T'=(Day 28 T/ Day 0 T) \*100

Fatigue:

% Δ ER'=(Day28 ER'/Day0 ER')\*100

Elasticity:

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% Δ ER=(Day28 ER/Day0 ER)*100
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**Results:** 



# SkinTone

### Fig.1 Fatigue Assessment:

SkinTone				Placebo		
Volunteer	Day 0	Day 28	Change	Day 0	Day 28	Change
1	0.063	0.043	-0.020	0.049	0.095	0.046
2	0.037	0.035	-0.002	0.045	0.093	0.048
3	0.035	0.048	0.013	0.047	0.125	0.078
4	0.047	0.046	-0.001	0.058	0.103	0.045
5	0.059	0.066	0.007	0.067	0.105	0.038
6	0.500	0.043	-0.007	0.072	0.132	0.060
Mean	0.049	0.047	-0.002	0.056	0.109	0.053

### Fig. 2 Elasticity Assessment:

SkinTone				Placebo		
Volunteer	Day 0	Day 28	Change	Day 0	Day 28	Change
1	1.023	0.097	-0.052	0.968	0.970	0.002
2	0.851	0.813	-0.038	0.953	0.953	0.085
3	0.842	0.843	0.001	0.796	0.796	0.003
4	1.120	0.989	-0.131	0.876	0.851	-0.025
5	0.937	0.857	-0.080	0.923	0.928	0.005
6	0.827	0.768	-0.059	1.060	1.032	-0.028
Mean	0.933	0.874	060	0.915	0.922	0.007



## SkinTone

SkinTone				Placebo		
Volunteer	Day 0	Day 28	Change	Day 0	Day 28	Change
1	0.395	0.387	-0.008	0.425	0.428	0.003
2	0.405	0.357	-0.048	0.369	0.374	0.005
3	0.378	0.362	-0.016	0.353	0.352	-0.001
4	0.342	0.327	-0.015	0.408	0.413	0.005
5	0.356	0.358	0.002	0.321	0.319	-0.002
6	0.348	0.337	-0.011	0.373	0.372	-0.001
Mean	0.371	0.355	-0.016	0.375	0.376	0.002

### Fig. 3 Toning Assessment:





#### Fig 4 Discussion:

It is understood that firm skin is elastic and that there is direct relationship between elasticity and recoil. As elasticity improves so does recoil and the values for epidermal recoil decrease as skin becomes toned. The integrity of the epidermis effects how it reacts under stressful conditions such as fatigue. Observing how the epidermis responds under stress may be a good indicator of the epidermal integrity. One may deduce that an improved tensor effect may be related to an improvement in epidermal integrity and a reduction in fatigue. In order to detect fatigue the cutometer was used for 5 successive trials to determine changes in elastic recoil.



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The results indicate that SkinTone reduces epidermal recoil therefore exhibiting a tightening effect on the skin. Epidermal fatigue was also reduced which indicates that perhaps SkinTone may improve the overall integrity of the epidermis. The combined improvement in tightness and the decrease in fatigue indicate that SkinTone improves skin tone.

#### Abstract:

Fibroblasts are responsible for tissue and collagen formation; when tissue becomes damaged fibroblasts migrate to the wound site to facilitate tissue repair via collagen production. To determine the efficacy of SkinTone to minimize the appearance of scarring an in vitro model was used to observe changes in fibroblast colonization.

#### Materials and Methods:

Fibroblasts were grown to confluence on glass slides with fibroblast culture medium (FCM). FCM components include: MEM/199 (3/1, V/V), penicillin (50 IU/ml), streptomycin (50 g/l), sodium bicarbonate (0.2% w/V) and fetal calf serum (10% V/V). The glass slides were kept in an incubator at 37 C with a 5% concentration of carbon dioxide. Cells were then treated with mitomycin C (10 ng/ml) for 2 hours to hinder cellular division. The centers of confluent fibroblast cultures were then scraped to mimic tissue damage. Cultures were then incubated with 0.5% SkinTone or the placebo (TGF- 1 at 10 ng/ml). Changes in fibroblast concentration were noted after periods of 24, 48 and 72 hours.

#### **Results:**

Incubation Time		Control	TGF-β1 (10 ng/ml)	AC Dermapeptide Toning
24 Hours	Trial 1	57	65	78
	Trial 2	60	73	89
	Mean	58.5	69	83.
	%	100	118%	12'
48 Hours	Trial 1	107	157	178
	Trial 2	103	120	136
	Mean	105	138.5	157
	%	100%	132%	150%
72 Hours	Trial 1	140	271	180
	Trial 2	146	243	210
	Mean	143	257	19
	%	100%	180%	136%

Fig. 1 Fibroblast Migration



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### SkinTone

Fig. 2

#### Increase in Fibroblast Migration



Discussion:

An increase in fibroblast proliferation was observed in cultures treated with SkinTone when compared with cultures treated with the control. Therefore one may conclude that SkinTone may be useful in treatments used to reduce the appearance of scarred tissue. It is understood that scarred tissue is most likely to form when there is a delay in tissue repair, by increasing the concentration of fibroblasts at the wound site one may be able to decrease the formation of scarred tissue.



SkinTone

Abstract:

The purpose of this study is to determine whether or not SkinTone is capable of increasing the rate of collagen IV and collagen VII production. Collagen IV serves as a protein in the epidermal scaffolding matrix and as well as a component of the dermal epidermal junction. Collagen VII is predominantly localized in the epithelium however it forms the anchoring fibrils and filaments at the dermal epidermal junction. The dermal epidermal junction is the point at which the dermis and epidermis bind.

Materials and Methods:

Confluent human fibroblasts were incubated with 2% SkinTone in the presence of 5% CO2 for a period of 72 hours. RNA was then extracted from the fibroblasts and transcribed to produce complementary DNA via reverse transcription. PCR analysis was then performed to isolate the segments of DNA present that code for collagen IV and collagen VII production. The DNA expression for fibroblasts treated with 2% SkinTone were compared to those of fibroblasts treated with TGF- 1 and -actin mRNA as the placebo.

The intensity of the bands formed on agarose via PCR were quantified using a Bio-Profil system, BIO-1D software (Vilber Lourmat, France). The intensity of the bands for cells treated with SkinTone were compare to those of cells treated with TGF-1 and -actin. The following ratios were used to discern the percent difference in mRNA production.

Variable Intensity (VI)=PCR band intensity for Variable/PCR band for -actin

Placebo Intensity (PI)=PCR band intensity for placebo/PCR band for -actin

% Difference= VI/PI\*100

Results:

Fig 1. Collagen VII mRNA Expression:

SkinTo	ne	TGF-1
VI	0.73	0.97
PI	0.52	0.52
% Difference	140	187

Fig. 2 Collagen IV mRNA Expression:

SkinTo	ne	TGF-1
VI	1.55	1.45
PI	1.02	1.02
% Difference	152	142



SkinTone

SkinTone TonIng & Collagen IV mRNA Expression

Fig. 4

Fig 3.





#### **Discussion:**

The results implicate that SkinTone may be efficacious in increasing the expression of mRNA that codes for collagen IV and VII thereby increasing the production of both collagen IV and VII. The increase in collagen production may also lead to improvement in the dermal epidermal junction integrity as well as an improved scaffolding matrix.