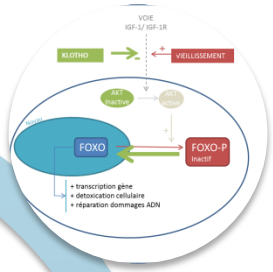
CODIF
R&N

LAKESIS

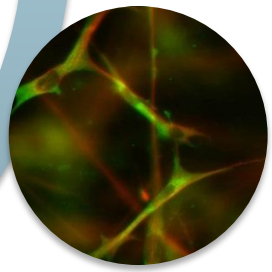


THE CONCEPT

Increase the synthesis of youth proteins to renew density in the dermis and redesign facial contours.

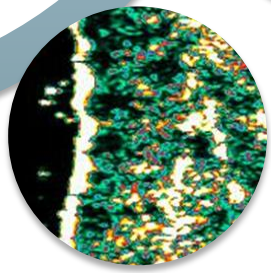
THE LEGEND

An oil extracted from crystal drop gathered from a tree endemic to the Island of Chios (Greece). A Greek goddess who gives her name to a youth protein…



IN VITRO EFFECTIVENESS

Increase in youth proteins and 3D visualization of redensifying process in the dermis.



IN VIVO EFFECTIVENESS

Increase in density and homogeneity of dermal supporting fibers; younger-looking skin seems firmer and plumped.

The precious oil that increases Klotho: LAKESIS

LAKESIS is an oil obtained by supercritical CO2 extraction from crystal drops collected on Pistacia lentiscus, a tree which is native to the Island of Chios (Greece).

The resinous sap produced by this tree forms drops as it falls to the ground. Once solidified, these drops take on a crystalline shape and are known as “crystal tears”.

The name of the oil obtained from these crystals is LAKESIS and refers to the Greek legend of the Three Fates.

The birth of a legend
LAKESIS

The Fates, Clotho, Lachesis and Atropos, are the Greek goddesses of night.

According to legend, Clotho spins the thread of life onto a spindle. Lachesis ("the allotter") measures its length and Atropos ("the inexorable") cuts this thread with her scissors, signifying the end of life.

Each thread represents a life, and the Fates represent destiny. It is said that no one can escape destiny.

The Three Fates by Michelangelo (1475-1564). Clotho, the spinner, is on the left holding a distaff from which the thread of life unwinds. On the right, Lachesis determines the destiny and length of the thread. In the center, Atropos waits to cut the thread at the moment of death.

The birth of a legend
LAKESIS

The goddess Clotho gave her name to the KLOTHO protein identified in 2005 and able to slow-down aging.

The Precious Oil of Crystal Tears increases the synthesis of Klotho protein in the fibroblasts. In reference to the legend, we have called this oil Lachesis, from the name of the second the goddess who measures lifespan.

"Winding the Skein" by Frederic Lord Leighton: Clotho is at right and Lachesis is on the left; painted in 1878

YOUTH PROTEINS

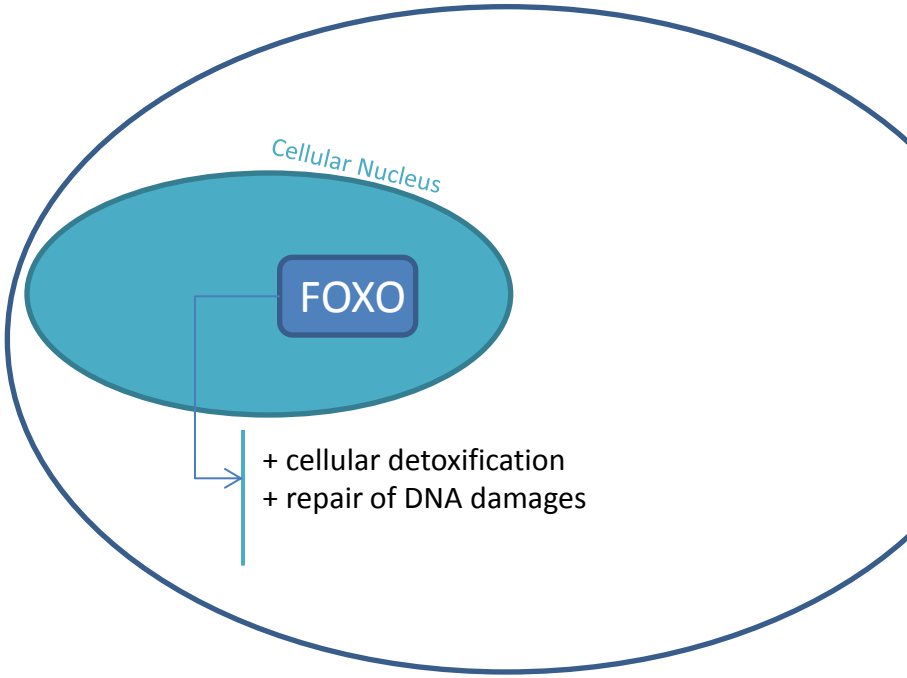
What are they? What role do they play in cellular longevity?

The current youth protein: FOXO

Studied very thoroughly during the past few years, FOXO has being considered to be the youth protein.

Its role in cellular protection and longevity has been well described and targeted to combat aging.

Located directly inside the nucleus, the "nuclear" form of FOXO activates the transcription of genes involved in cellular detoxification and the repair of DNA damages [1].



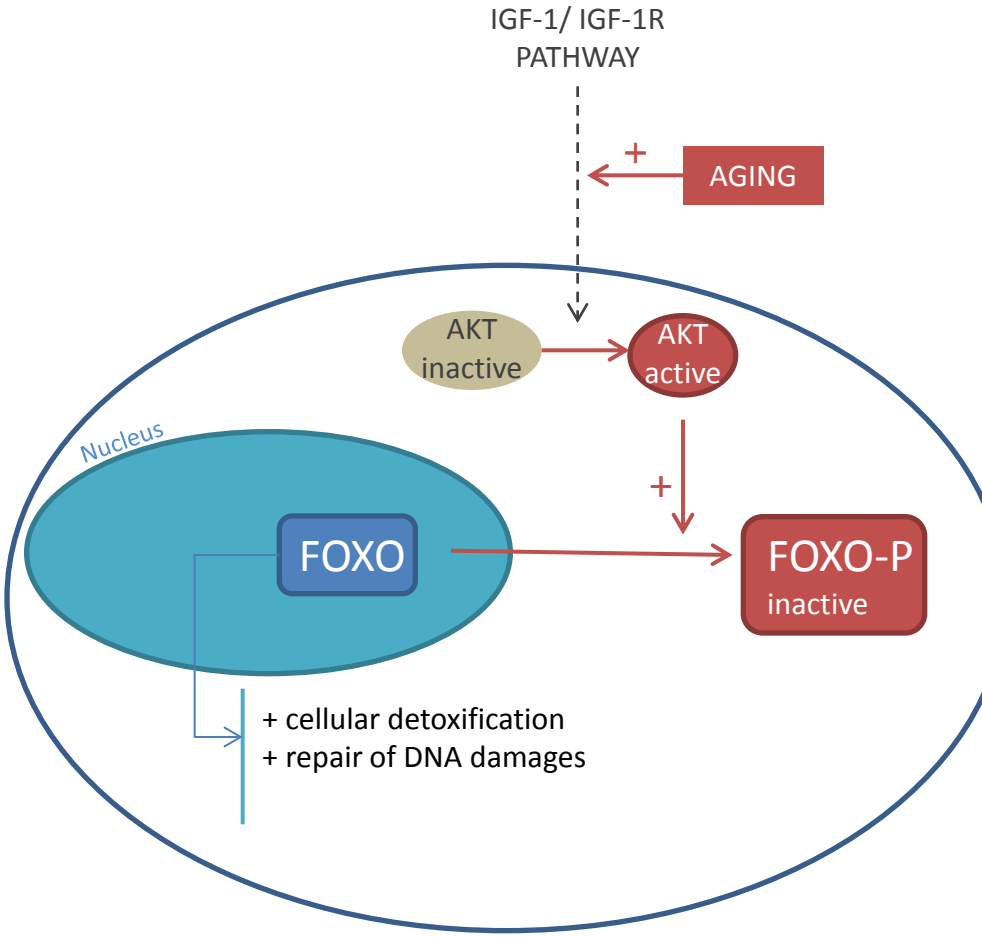
[1] The multiple roles of FOXO transcription factors. A. Brunet. Médecine Sciences, vol.20, no. 10, 2004, p. 856-859.

Effect of aging on FOXO

The FOXO protein exists in two forms inside the cell.

When it is phosphorylated by the AKT factor, FOXO becomes FOXO-P and leaves the nucleus for the cytoplasm, where it loses its properties [2].

Activation of the AKT factor increases with age, thereby provoking a predominance of the inactive form of FOXO.

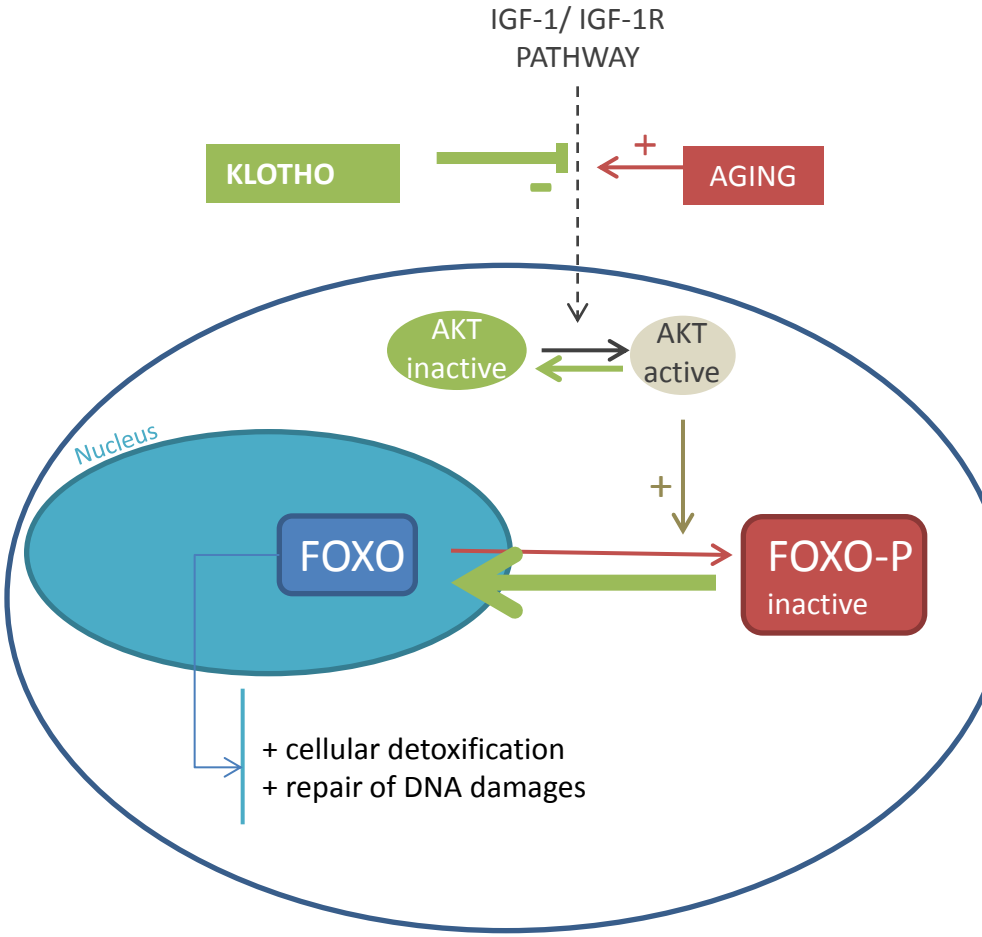


[2] Klotho and aging. M. Kuro-o. Biochem Biophys Acta. 2009 October; 1790(10): 1049-1058.

THE NEW youth protein: KLOTHO

A protein which is able to inhibit AKT activation and therefore maintain FOXO inside the cell nucleus was identified in 2005.

Named KLOTHO in reference to the Greek goddess Clotho, who spins the thread of life, it is now considered by scientists to be the NEW YOUTH PROTEIN.

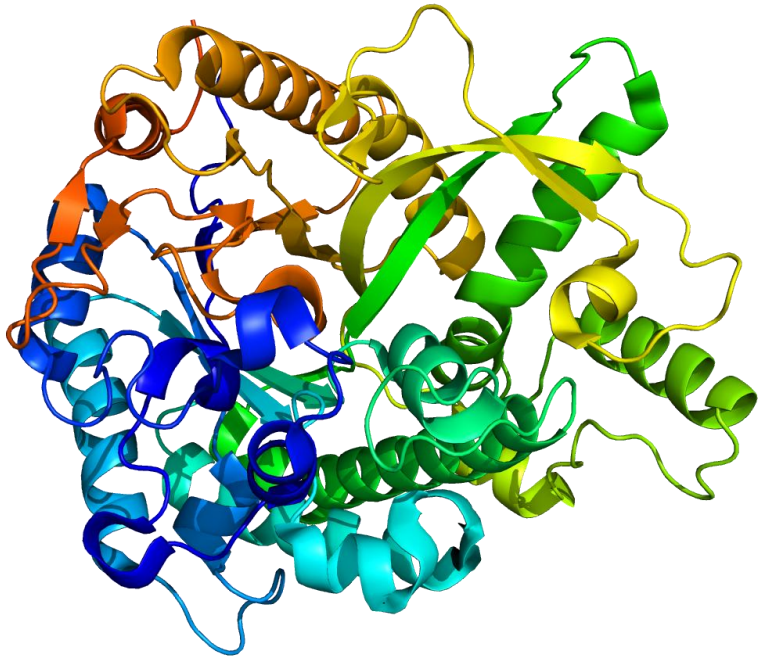


The power of KLOTHO

The KLOTHO protein gene was identified for the first time in mice [3]. Its mutation causes a premature apparition of all the traditional signs of aging and leads to premature death.

On the contrary, when it is overexpressed, this gene leads to a 20% to 30% increase in mice lifespan.

The Klotho protein is involved in a large number of signaling pathways leading to the re-establishment of optimal cellular function.



[3] http://www.washingtonpost.com/wp-dyn/content/article/2005/08/25/AR2005082501224.html

KLOTHO and cosmetic applications

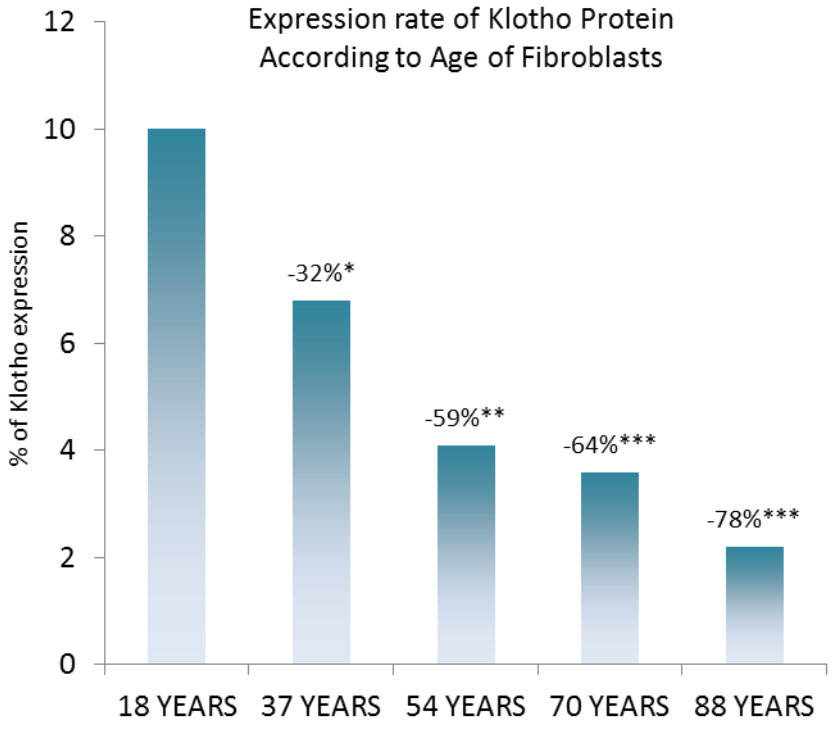
Klotho protein has not yet been studied for cosmetic applications. Our laboratories have shown that its rate of expression is directly linked to the age of human fibroblasts.

Protocol: human dermal fibroblasts coming from donors of different ages. Quantification of KLOTHO protein by immunofluorescence.

RESULTS

The decrease in KLOTHO expression starts very early during the ageing process: from roughly 30 years. Then its expression gradually decreases with fibroblasts ageing.

Increasing Klotho synthesis in the fibroblasts thus represents a new strategy to combat skin aging.



** p<0,05 ; **p<0,01 ; ***p<0,001 Student's Test*

LAKESIS

Action mechanism In-Vitro Effectiveness

LAKESIS: Action Mechanism

REACTIVATION OF YOUTH PROTEINS

1- Increases KLOTHO synthesis

2- Increases inactive AKT

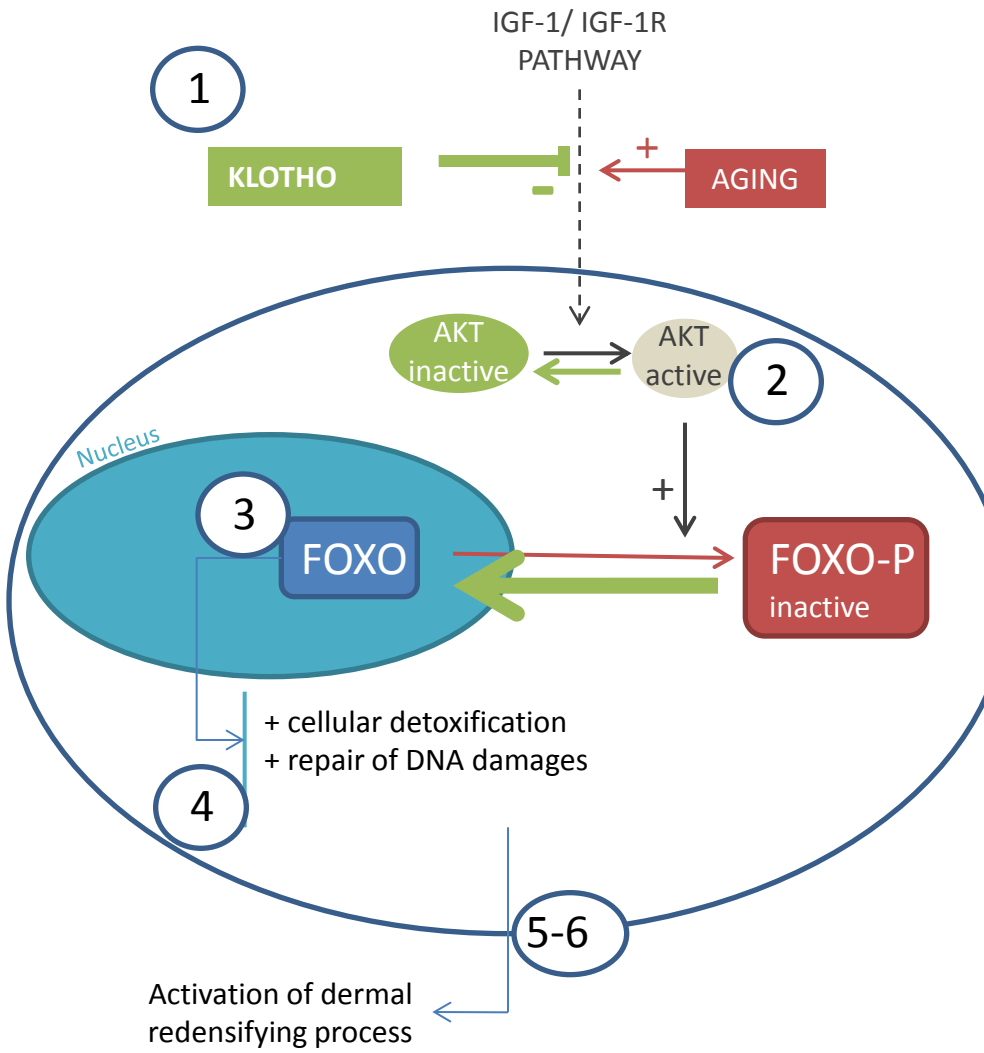
3- Increases the FOXO / FOXO-P ratio

REACTIVATION OF ANTI-AGING ACTION

4- Reactivates the cellular detoxification processes

5- Reactivates cellular activity

6- Promotes dermal redensifying process



0.005%

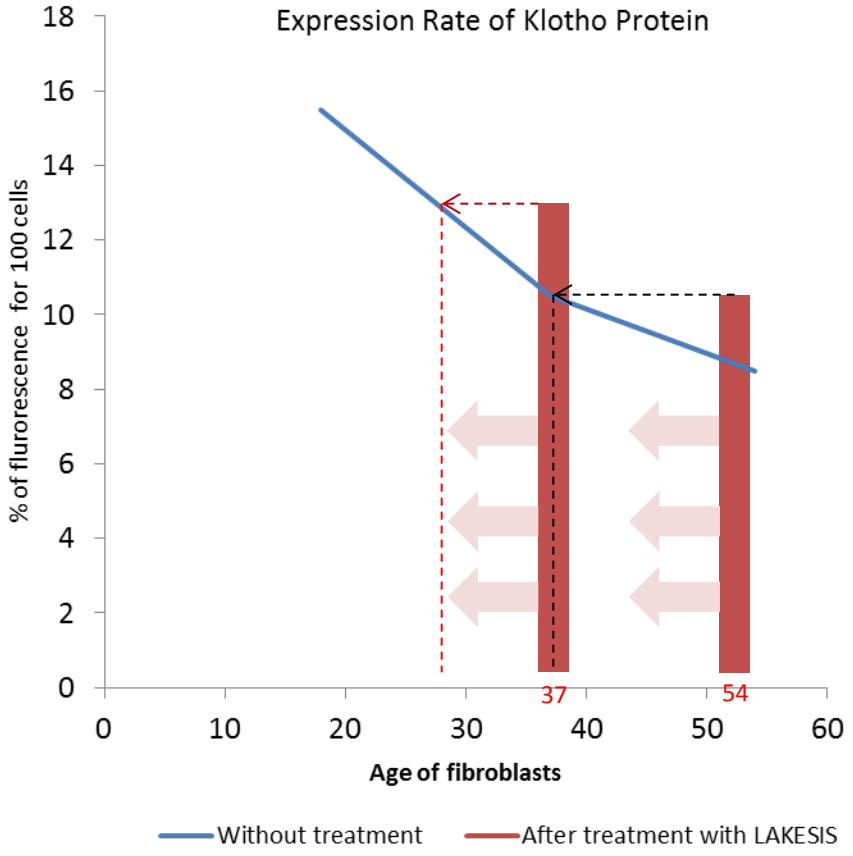
LAKESIS increases KLOTHO

Protocol: human dermal fibroblasts from donors of different ages and incubated with LAKESIS at 0.005% for 24 hours. Observation and analysis of Klotho expression using immunofluorescence.

RESULTS

LAKESIS increases the synthesis of the KLOTHO protein in fibroblasts, from the earlier to the later stages of ageing process.

AFTER TREATMENT, 54-YEAR-OLD FIBROBLASTS EXPRESS THE SAME AMOUNT OF KLOTHO AS 37-YEAR-OLD FIBROBLASTS.
AND 37-YEAR-OLD FIBROBLASTS EXPRESS THE SAME AMOUNT OF KLOTHO AS 28-YEAR-OLD FIBROBLASTS.



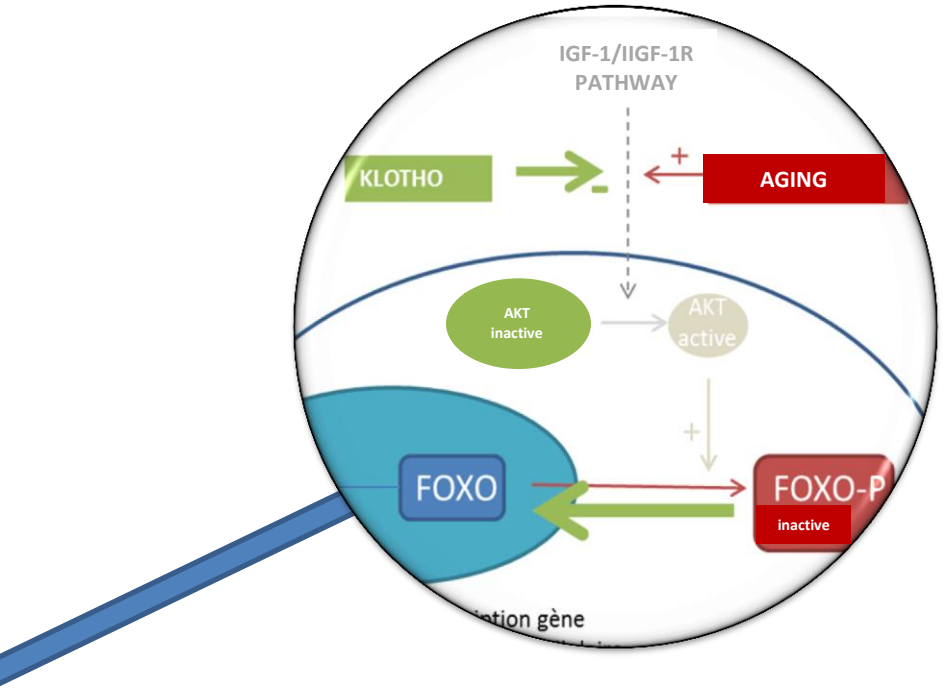
0.005%

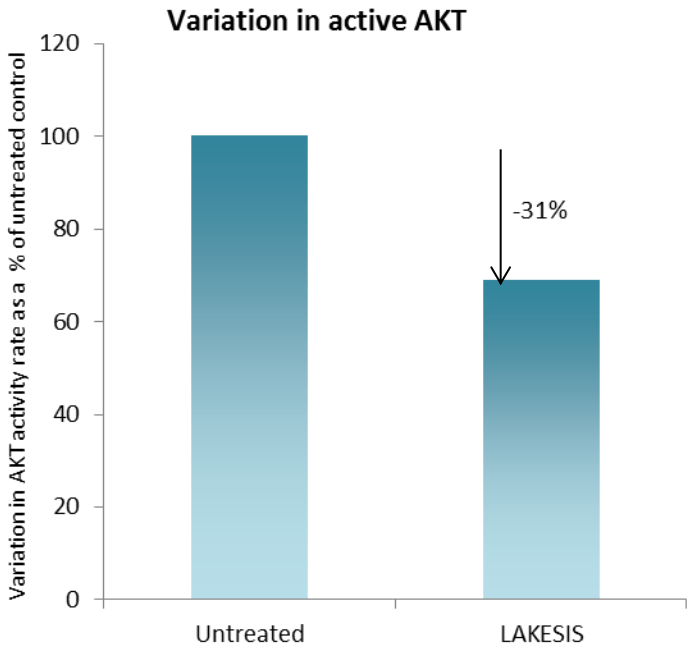
LAKESIS decreases active AKT

Protocol: 54-year-old human dermal fibroblasts were treated or not with LAKESIS at 0.005% for 24 hours. Staining and analysis of phosphorylated and non-phosphorylated AKT.

RESULTS

By increasing Klotho synthesis, LAKESIS decreases the level of active AKT.





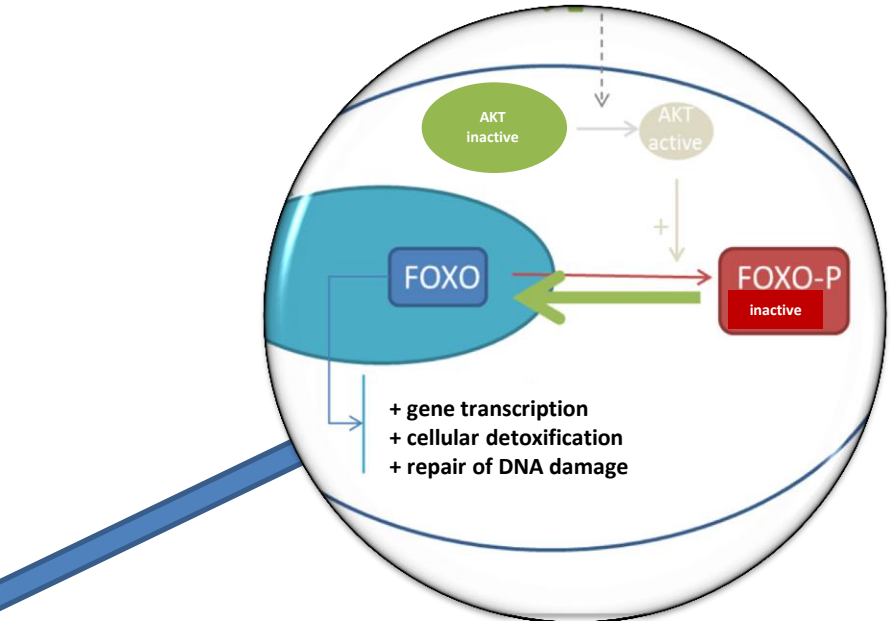
0.005%

LAKESIS increases the FOXO rate

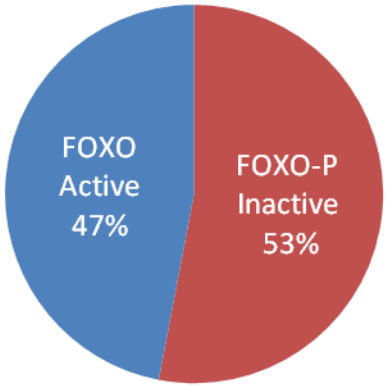
Protocol: 54-year-old human dermal fibroblasts were treated or not with LAKESIS at 0.005% for 24 hours. Analysis of phosphorylated and non-phosphorylated FOXO.

RESULTS

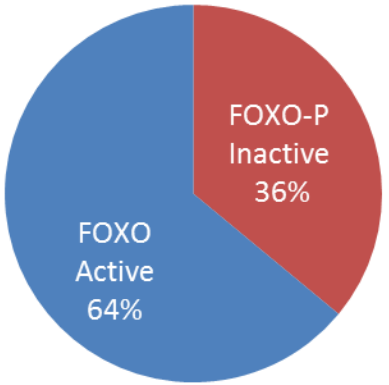
By inhibiting the AKT factor, LAKESIS decreases phosphorylation of the FOXO youth factor. This factor is now mainly located in the nucleus, where it activates the transcription of detoxification and cellular repair factors.



Before Treatment



After Treatment with LAKESIS



0.005%

LAKESIS increases the catalase rate

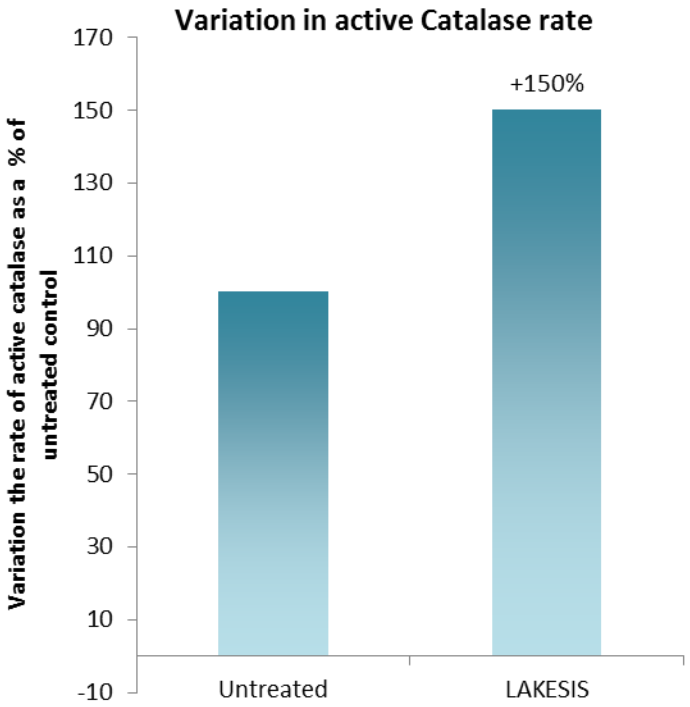
Protocol: 54-year-old human dermal fibroblasts were treated or not with LAKESIS at 0.005% for 24 hours. Staining and analysis of active catalase.

Catalase is part of an enzymatic pool which detoxifies the cell.

RESULTS

By increasing the FOXO factor, LAKESIS relactivates the cellular detoxification processes.

The rate of active catalase increases by 145%.



LAKESIS decreases TXNIP

0.2%

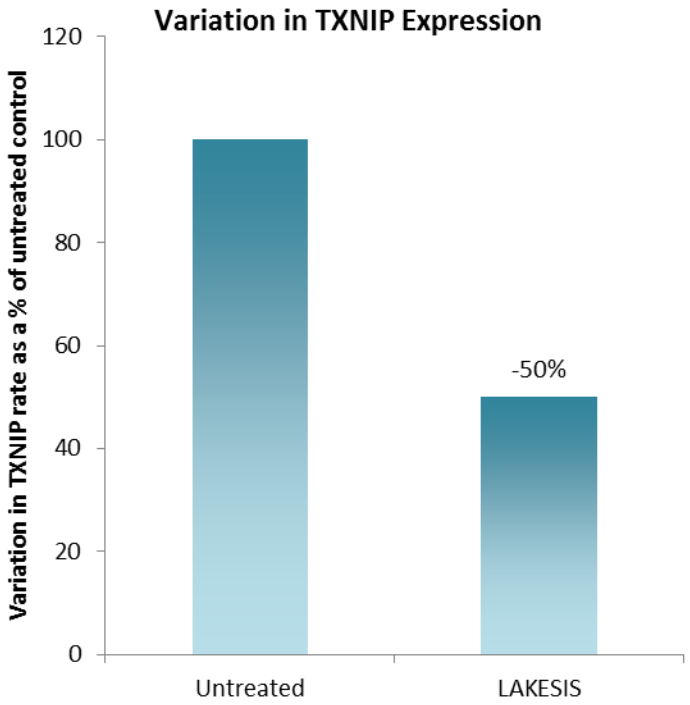
Protocol: reconstituted human skin was treated or not with LAKESIS at 0.2% using a topical application for 24 hours. Gene expression analysis was made with mini-chips.

Thioredoxin is a universal and very powerful antioxidant system. It has been largely studied mainly for its involvement in combating cellular senescence. It plays a major role in the cellular detoxification process.

The TXNIP factor is a thioredoxin inhibitor.

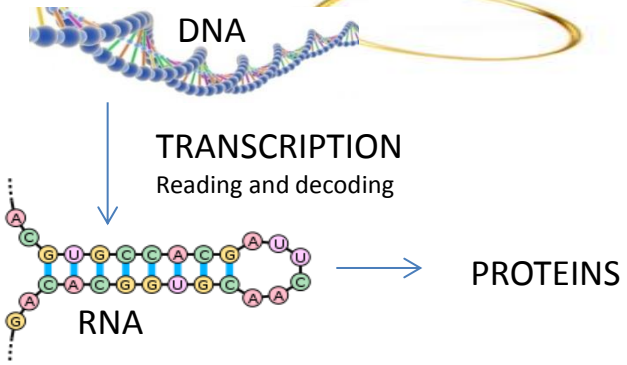
RESULTS

LAKESIS decreases the TXNIP rate by 50%.
This result illustrates its anti-aging action in favor of cellular detoxification proteins.



0.005%

LAKESIS relaunches cellular activity



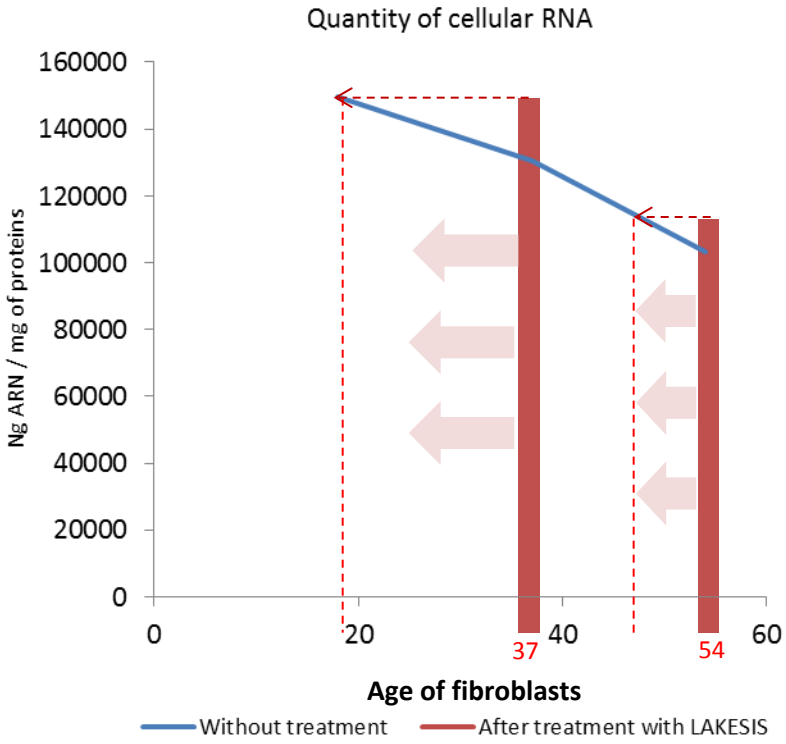
Gene transcription is the process of reading and translating genetic information in RNA.
After transcription, each RNA represents a “recipe” to be used for manufacturing a protein. The amount of RNA present inside a cell provides important information about the rate of cellular activity. With age, it drops.

CELLULAR RNA AMOUNT MEASUREMENT

Protocol: human dermal fibroblasts from donors of different ages. Extraction and quantification of total RNA.

RESULTS

LAKESIS reactivates cellular detoxification. Cells are more active, which is translated here by an increase in the total amount of cellular RNA. 37-year-old fibroblasts achieved an RNA synthesis rate equivalent to that of 18-year-old fibroblasts.



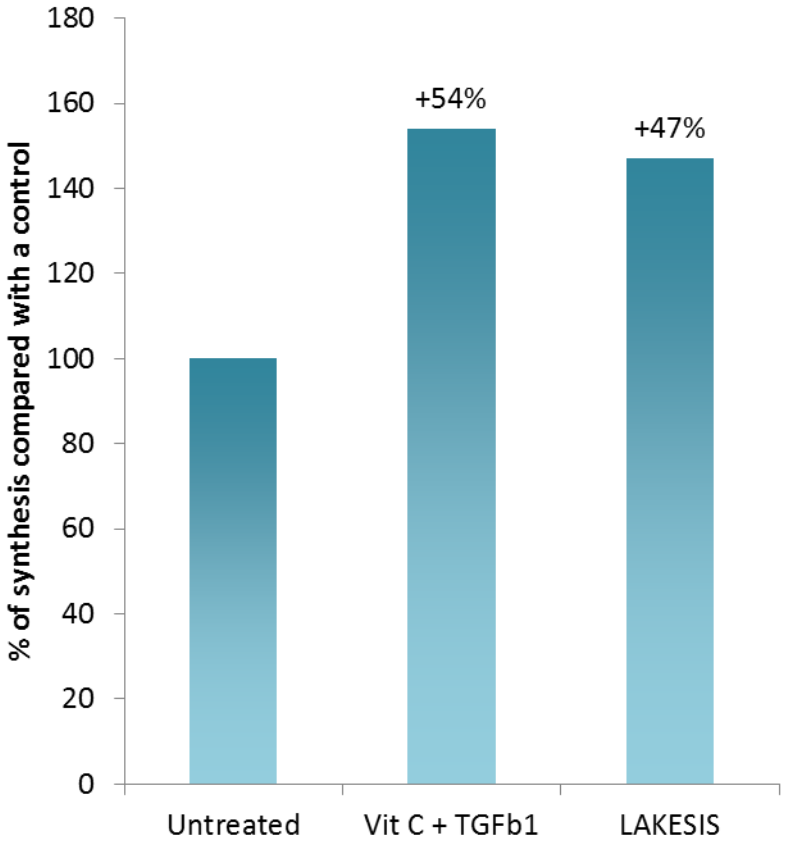
0.0025%

LAKESIS relaunches collagen I synthesis

Protocol: 39-year-old human dermal fibroblasts were treated or not with LAKESIS at 0.0025% for 24 hours.

RESULTS

By relaunching cellular activity, LAKESIS increases collagen I synthesis.



REJUVENATING ACTION OF LAKESIS ON THE EXTRACELLULAR MATRIX

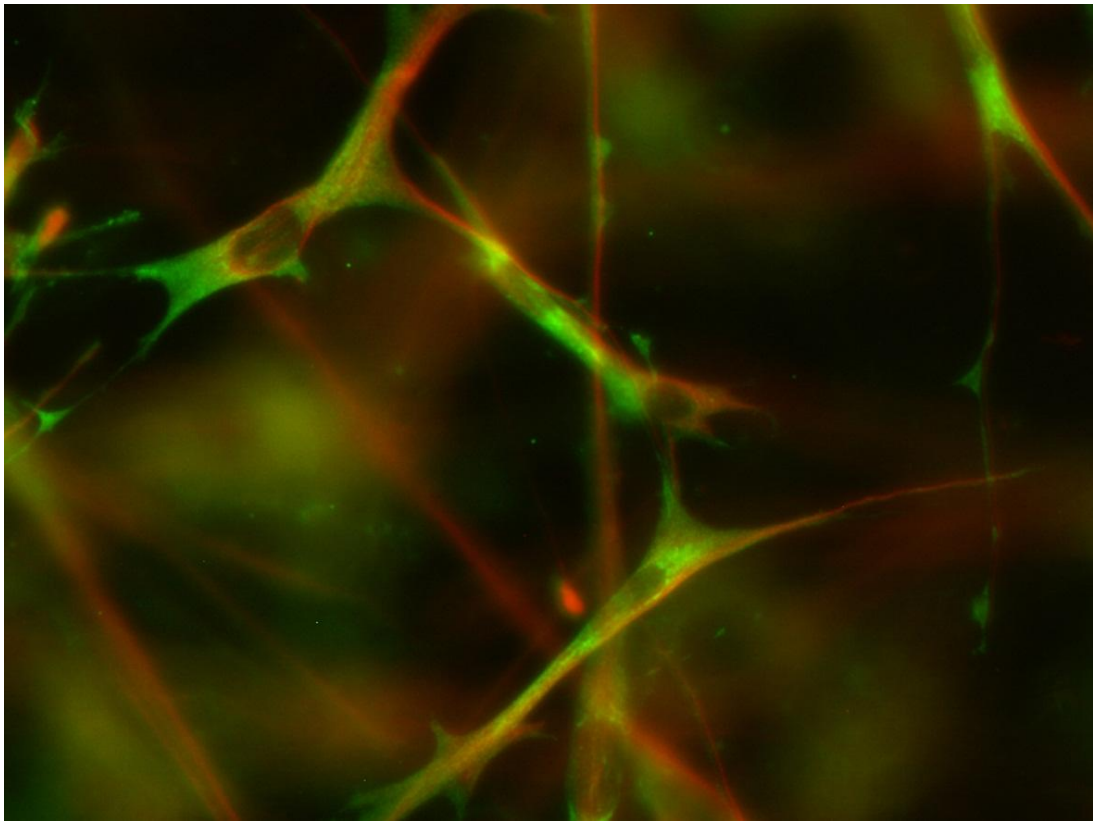
Use of 3D dermis models and visualization of dermal redensification process

IN VITRO TEST

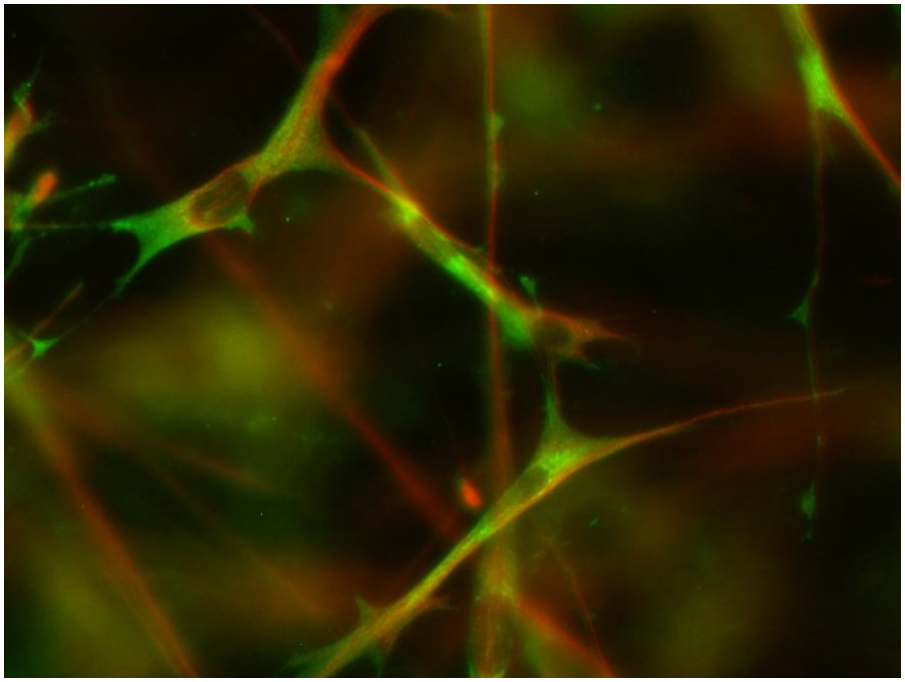
3D dermal model

The use of a 3D dermal model enables visualizing fibroblasts in the extracellular matrix and obtaining data on their interaction and activity within the extracellular matrix.

We used red staining to visualize the fibroblast cytoskeleton (vimentin) and green staining to visualize synthesis of the proteins we are interested in.

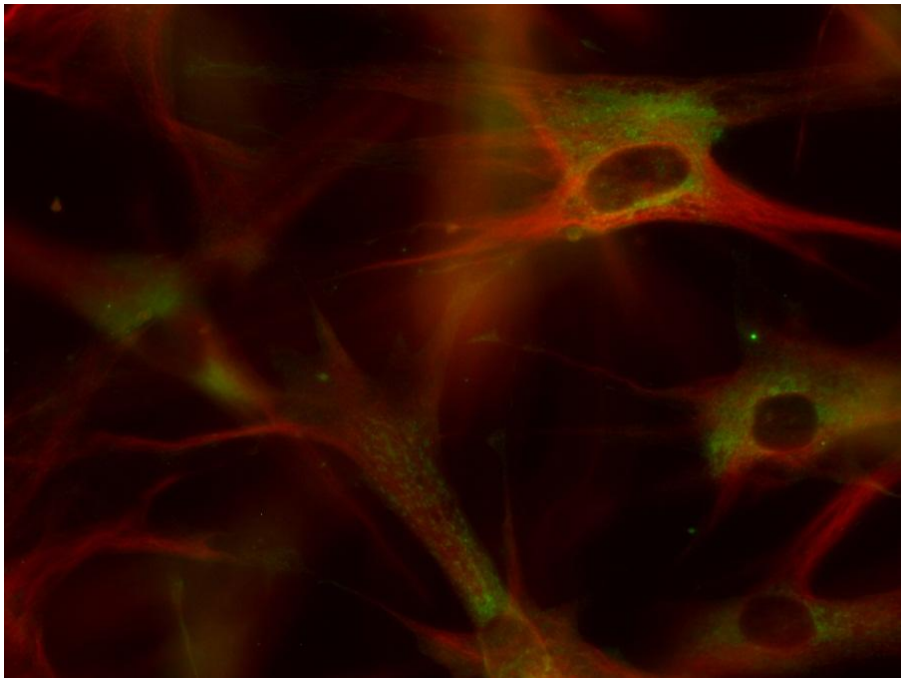


Effect of age on the 3D model



MODEL FROM 18-YEAR-OLD FIBROBLASTS

The very obvious green staining indicates strong protein synthesis activity.



MODEL FROM 54-YEAR-OLD FIBROBLASTS

Green staining is much weaker and the cytoskeleton is much more visible. Protein synthesis activity is weak.

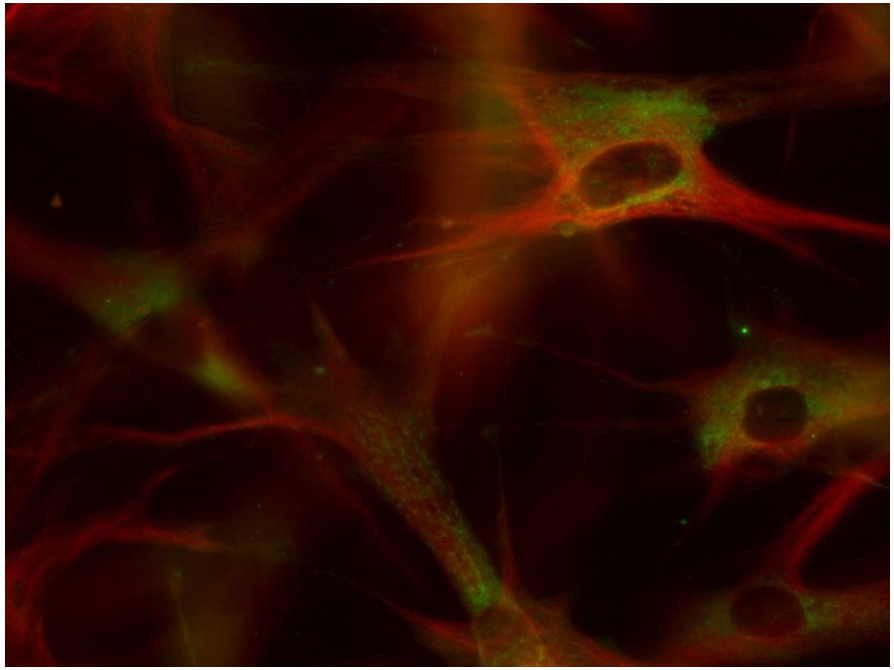
IN VITRO TEST

0.005%

LAKESIS and fibrillin synthesis

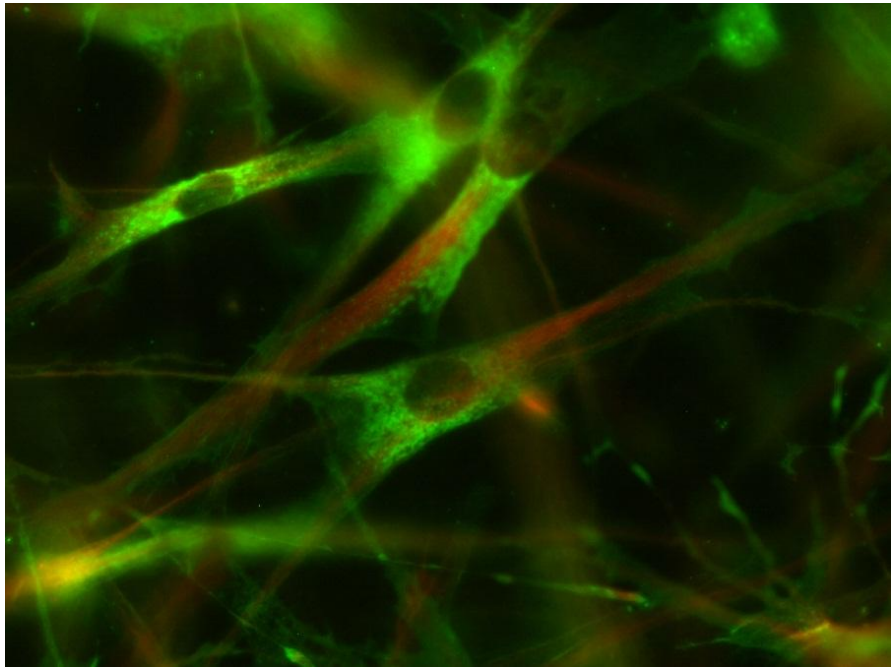
Fibrillin is a major component of elastic fibers

Protocol: 54-year-old human dermal fibroblasts included in a collagen lattice were treated or not with LAKESIS at 0.005% for five days. Visualization of fibrillin using green immunofluorescence.



MODEL FROM 54-YEAR-OLD FIBROBLASTS

Visualization of fibrillin synthesis in green.



MODEL FROM 54-YEAR-OLD FIBROBLASTS TREATED WITH LAKESIS.

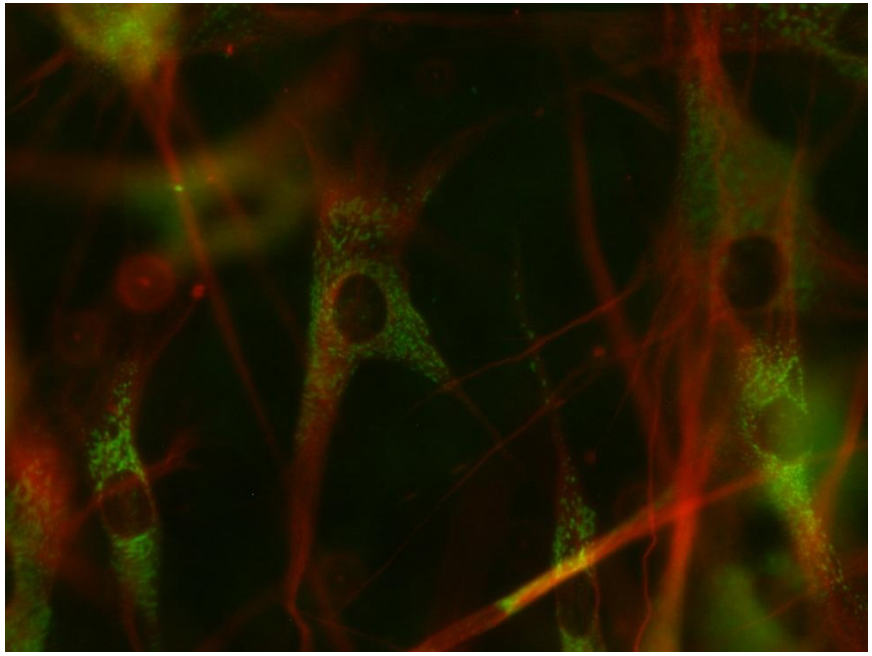
Activation of synthesis and dermis redensification in fibrillin.

IN VITRO TEST

0.005%

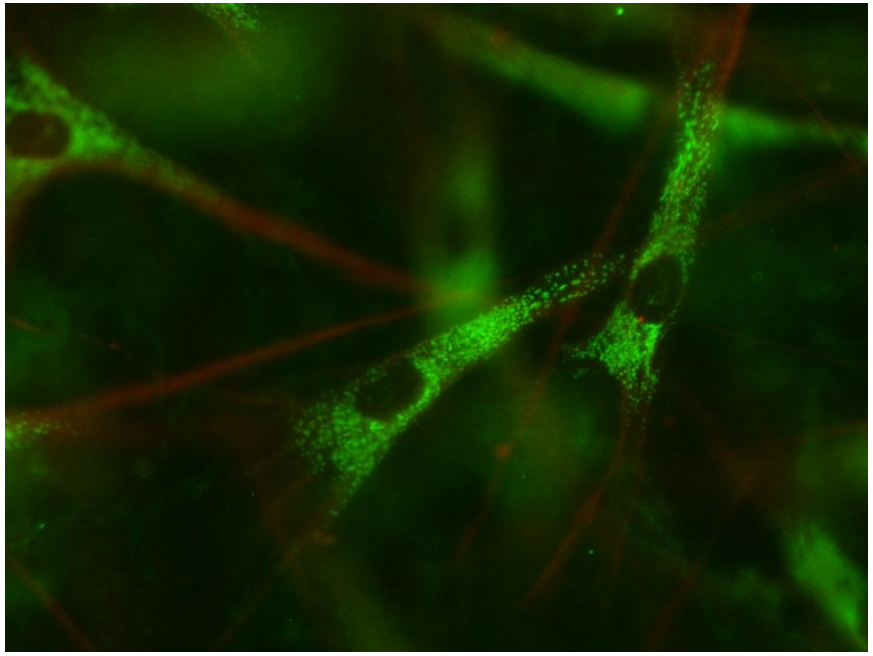
LAKESIS and collagen synthesis

Protocol: 54-year-old human dermal fibroblasts included in a collagen lattice were treated or not with LAKESIS at 0.005% for five days. Visualization of collagen using green immunofluorescence.



MODEL FROM 54-YEAR-OLD FIBROBLASTS

Visualization of collagen synthesis in green.



MODEL FROM 54-YEAR-OLD FIBROBLASTS TREATED WITH LAKESIS

Activation of synthesis and dermis redensification in collagen.

GLOBAL ANTI-AGING ACTION OF LAKESIS

Clinical tests

LAKESIS ANTI-AGING EFFECTIVENESS

0.2%

Clinical measurements

PROTOCOL

- 20 volunteers ages 55 to 65
- Twice-daily application of a cream containing 0.2% LAKESIS for 28 days
- Application area: whole face

PARAMETERS ANALYZED

- Thickness and density of the SENEB (SubEpidermal Non-Echogenic Band)
- Homogeneity and density of the entire dermis
- Overall evaluation of skin youth by a dermatologist
- Overall assessment using an analogical scale of the anti-aging effectiveness of the product by the volunteers: they graded each criterion by positioning a slider on a non-calibrated scale from 0 to 10.

LAKESIS ANTI-AGING EFFECTIVENESS

0.2%

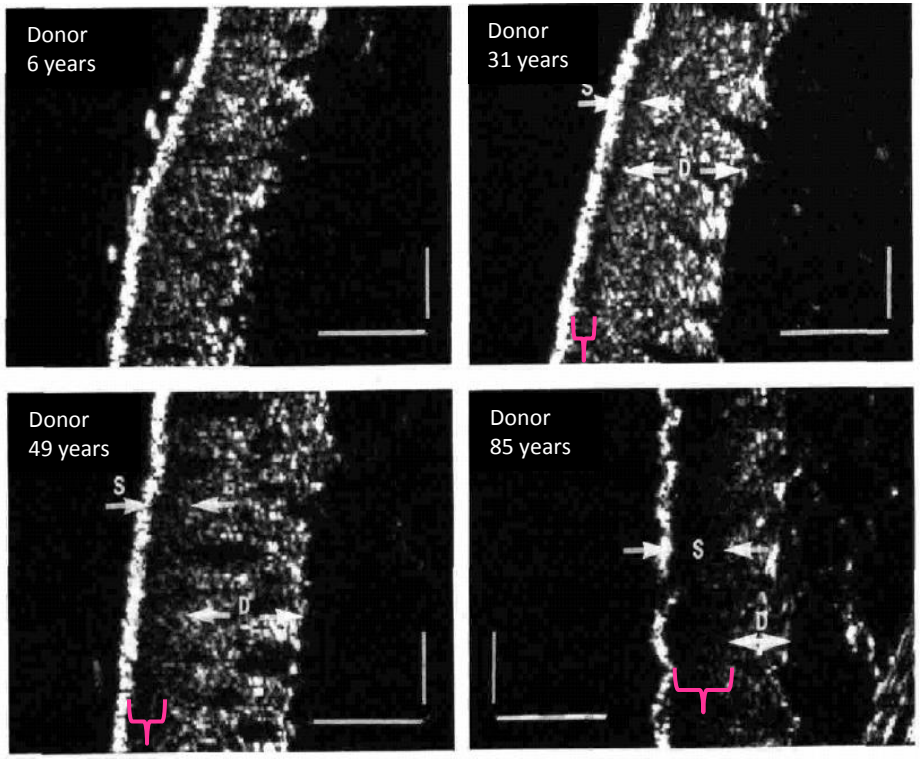
Clinical measurements

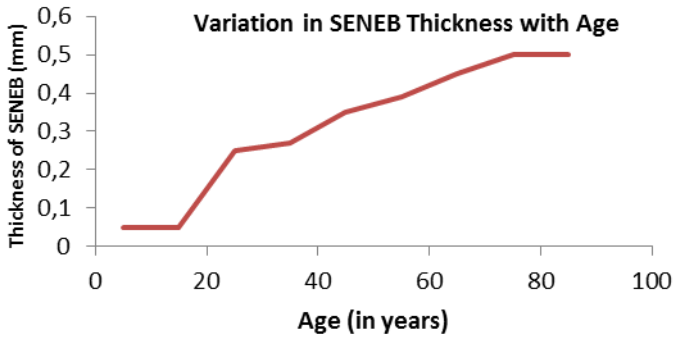
ANALYSIS OF THE SENEB [1]

The SENEB (SubEpidermal Non-Echogenic Band) is found in the most superficial layer of the dermis. It is an area very poor in supporting fibers and corresponds to the black band indicated here by pink brackets. Almost non-existent in children, it increases with age.

The thickening of this fiber-poor area is characterized by a loss of firmness and tissue support.

Researchers have shown that the thickness of the SENEB increases with age.





*[1] Jean de Rigal, Catherine Escoffier, Bernard Querleux, Brigitte Faivre, Pierre Agache and Jean-Luc Lévêque, (1989). Assessment of aging of the human skin by in vivo ultrasonic imaging. Journal of Investigative Dermatology, **93**(5).*

LAKESIS ANTI-AGING EFFECTIVENESS

0.2%

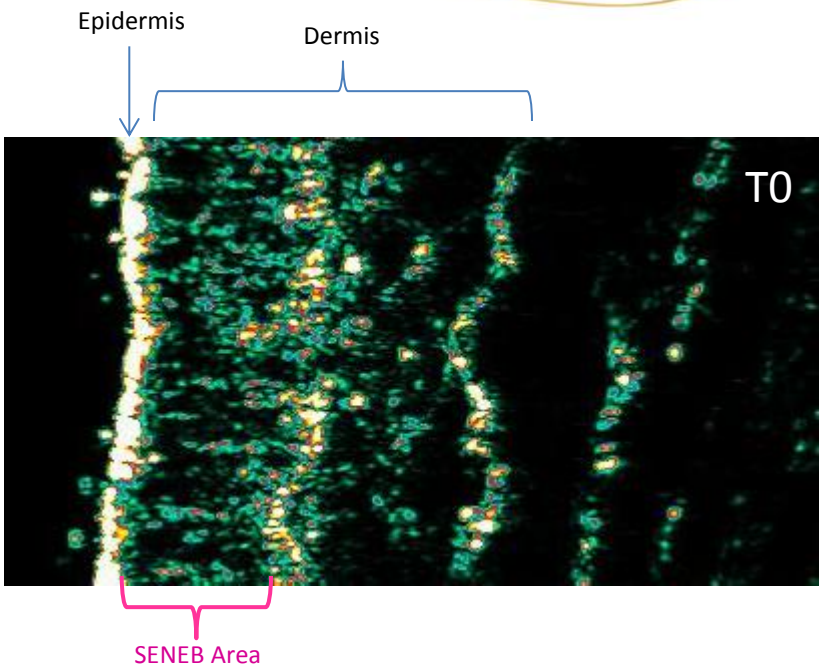
Clinical measurements

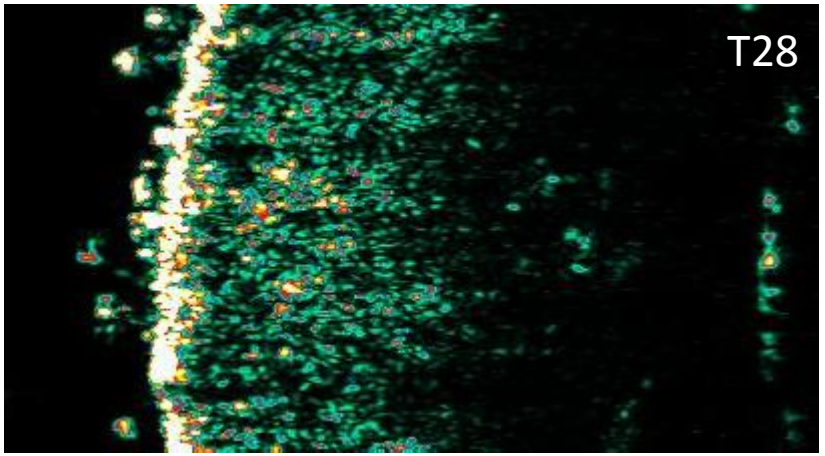
ANALYSIS OF THE SENEB

Density of supporting fibers (in green):
+51% on average ($p<0.001$)
And up to +103%

Variation in SENEB surface
-29% on average ($p<0.001$)
And up to –43%

LAKESIS stimulates the synthesis of supporting fibers in the SENEB and decreases the size of this non-echogenic area. This action reinforces the immediate supporting properties of the dermis.





Volontaire 39

LAKESIS ANTI-AGING EFFECTIVENESS

Clinical measurements

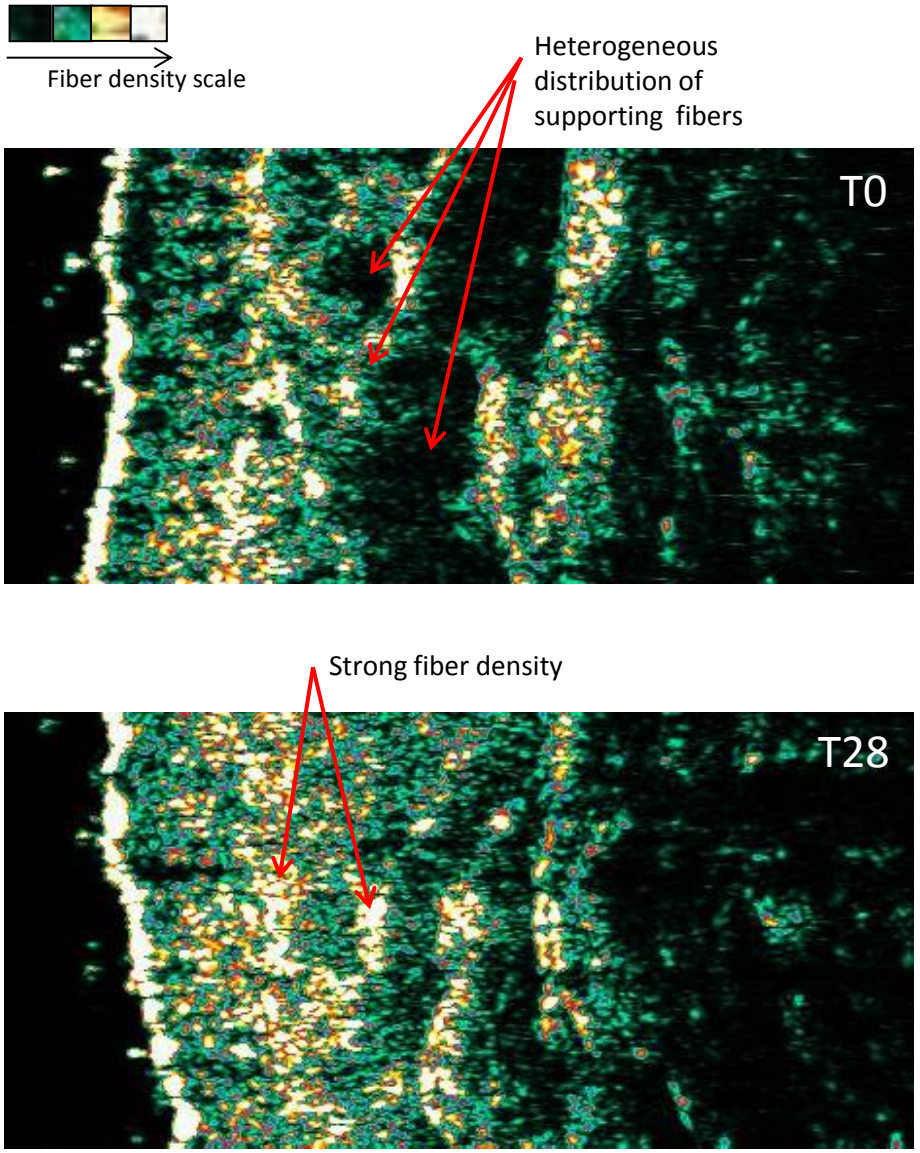
0.2%

ANALYSIS OF THE ENTIRE DERMIS

Homogeneity of supporting fibers distribution:
+16% on average (p<0.005)
And up to +34.6%

Variation of supporting fibers density:
+18% on average (p<0.005)
And up to +37.4%

LAKESIS increases supporting fibers density throughout the dermis and also ensures a homogeneous distribution of these fibers.



Volontaire 3

LAKESIS ANTI-AGING EFFECTIVENESS

0.2%

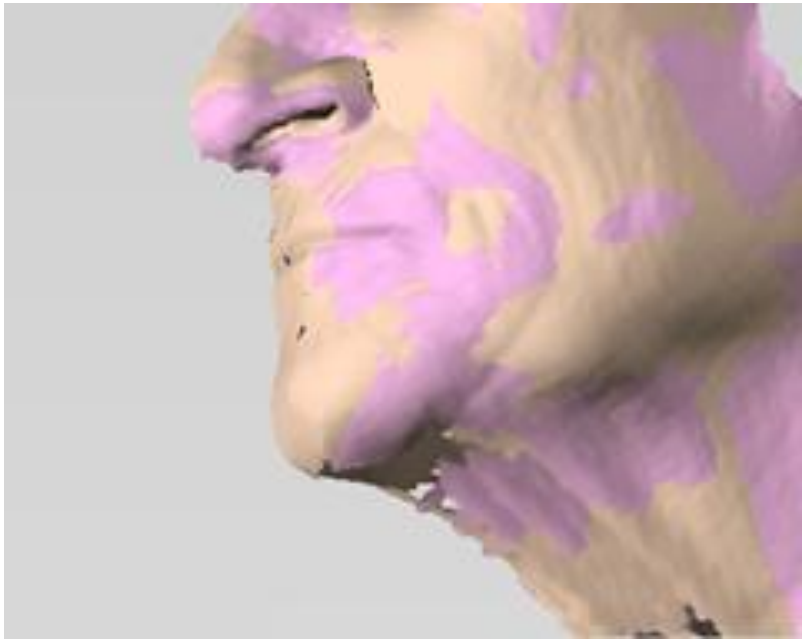
Clinical measurements

ANALYSIS OF FACIAL CONTOURS

Using fringe projection allows reconstituting the volume of the face at T0 and T28. A superposition of both volumes is visible on picture 3. Everywhere the skin is yellow means that the volume of these areas has been lifted after 28 days treatment.

Variation of the volume of the face:
-4.41% on average ($p<0.05$) and up to -16%

More than 2/3 women observed in improvement on their oval face.



Everywhere the skin is yellow means that the volume of these areas has been lifted.

LAKESIS ANTI-AGING EFFECTIVENESS

Clinical measurements

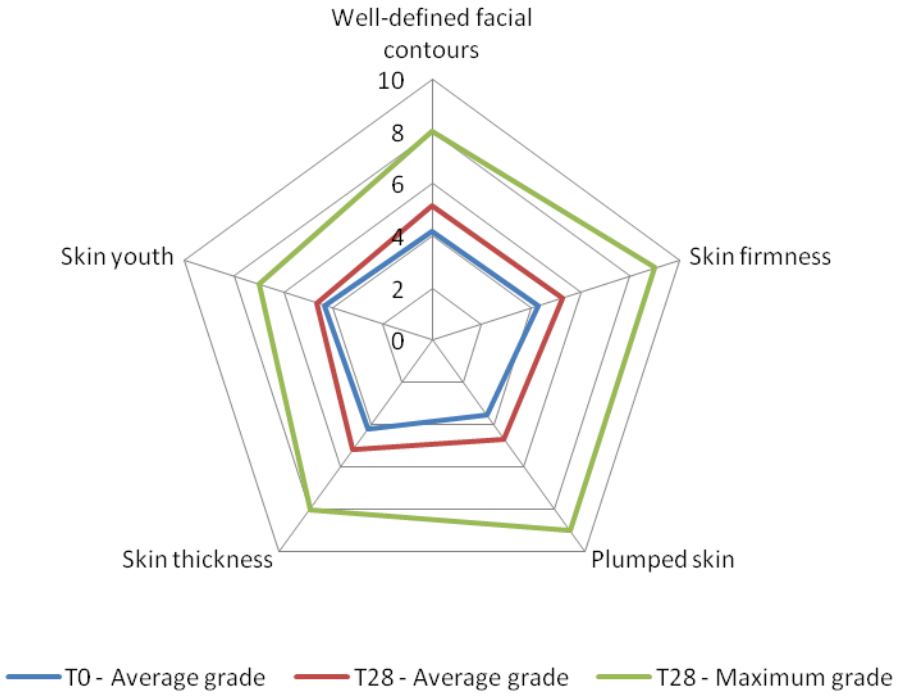
0.2%

ANALYSIS OF GLOBAL ANTI-AGING EFFECTIVENESS

Clinical analysis of skin youth by a dermatologist:
+8.4% on average ($p<0.05$)
And up to 29%

Consumers analysis on an analogical scale:
Facial contour well defined: +24% ($p<0.05$)
Skin firmness: +23% ($p<0.1$)
Skin thickness: +22% ($p<0.05$)

Consumers analysis on an analogical scale:



LAKESIS reactivates youth proteins and improves supporting fibers density in the dermis. Skin seems younger and thicker. Facial contours appear to be redrawn.

LAKESIS

A precious oil extracted from crystal drops, LAKESIS reactivates cellular youth proteins and the synthesis of supporting fibers. The dermis gains density and skin seems plumped and thicker.

IN VITRO:

- Increases synthesis of the KLOTHO youth protein
- Increases the level of the FOXO youth protein
- Reactivates the cellular detoxification process
- Reactivates cellular activity
- Promotes redensification of dermal matrix

IN VIVO:

- Improves density of the SENEB and decreases its surface
- Increases supporting fiber density in the dermis and ensures homogeneous fibers distribution
- Lifts the facial contours
- Skin appears younger
- Skin seems thicker and firmer

LAKESIS

INCI name: Caprylic/capric triglyceride (and) *Pistacia Lentiscus (Mastic) Gum*

OK CHINA

Recommended % of use: *0.2%*

EXAMPLES OF USE

In an anti-aging facial oil - with Stoechiol or Samphira oil

In a global anti-aging product - with Matrigenics.14G and Actiporine 8G

In a youth-reactivating serum - with Phycojuvenine

In a youth-detoxifying night cream – with Phormiskin Bioprotech G

LIFT-FIRMING FACE OIL

Formulation guide

This FACIAL OIL is formulated with LAKESIS and HYDRASALINOL, a hydra-nourishing active ingredient.

MINI-PRODUCT AVAILABLE – 30 ML
A LUXURY GIFT

Phase	Raw Material / Trade Name	INCI Name	%
A	DEODORIZED ORGANIC SESAME SEED OIL (1)	Sesamum Indicum Seed Oil	35.150000
	VIRGIN ORGANIC CARTHAMUS OIL (1)	Carthamus Tinctorius Seed Oil	8.000000
	DEODORIZED SUNFLOWER OIL (1)	Helianthus Annuus Seed Oil	15.000000
	CETIOL LC (2)	Coco-Caprylate/Caprate	41.000000
	NATROX RO (3)	Helianthus Annuus Seed Oil and Rosmarinus Officinalis Leaf Extract	0.200000
	HYDRASALINOL (4)	Caprylic/Capric Triglyceride and Salicornia Herbacea	0.250000
	LAKESIS (4)	Caprylic/Capric Triglyceride and Pistacia Lentiscus Gum Oil	0.200000
	WATER LILY R18717 FRAGRANCE (5)	Fragrance	0200000

How to use:
In an appropriate recipient, add the raw materials and cold mix with a deflocculator at 1000 rpm.
Mix for about 10 minutes until a clear solution is obtained.

CODIF R&N