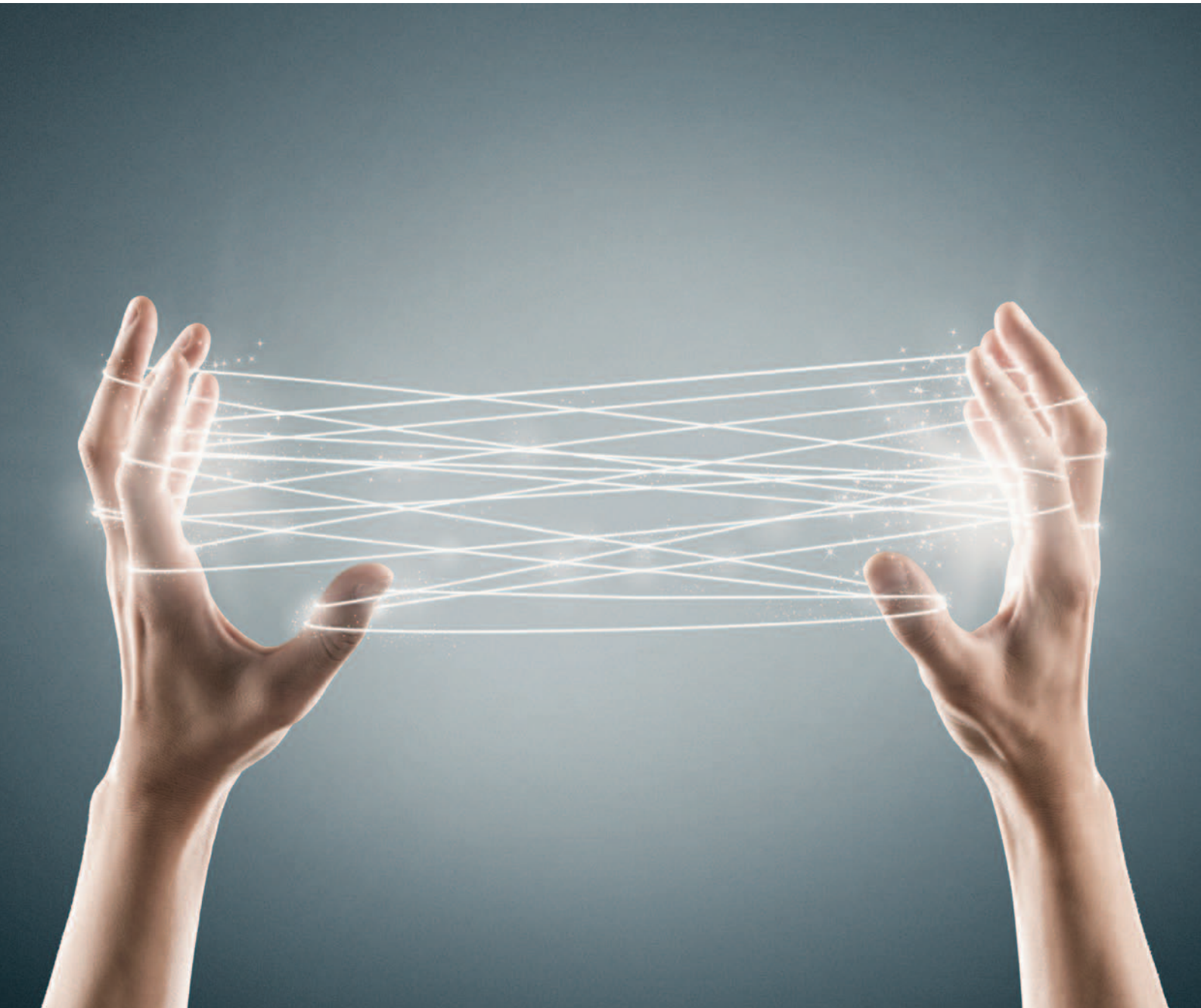
CODIF
R&N

# NEUROGUARD

## The New Science of Aging

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# NEUROGUARD

## THE NEW SCIENCE OF AGING

Neuroguard opens the door to new neurocosmetic strategies. Beyond nerve-cell communications, it decrypts, targets and provides preventive and curative treatment against skin neuroaging. Our laboratories have indeed demonstrated that the aging of neurons directly affects the communication between nerves and fibroblasts, triggering a fall in fibroblasts vitality and a decrease in collagen and elastin synthesis. Neuroaging promotes Fibroaging.

## FIGHTING AGAINST SKIN NEUROAGING

*Why ?*

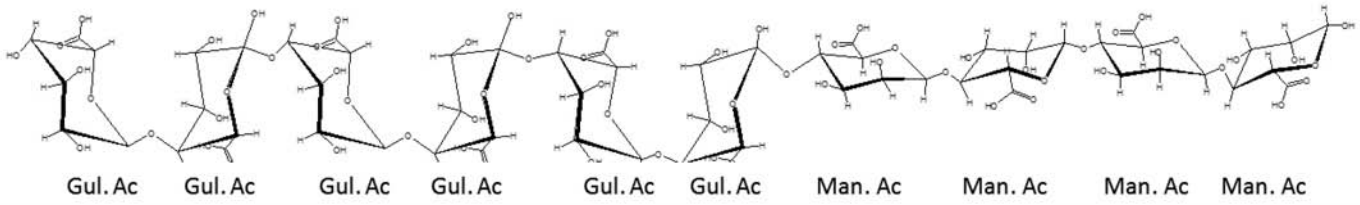
To re-establish a healthy communication between nerves and fibroblasts and to reactivate dermal cells previously weakened by neuroaging toxicity. Fighting against neuroaging enables delaying fibroaging. Thanks to the reactivation of collagen and elastin synthesis, NEUROGUARD treats deep wrinkles within 28 days.

*For whom ?*

For ranges dedicated to mature skins and wishing to offer an anti-wrinkle solution specific to cellular disorders occurring with aging. For all ranges offering solutions to prevent and delay the mechanisms of skin aging.

## NEUROGUARD
## The Enzymatic Biotech Expertise of CODIF R&N

Neuroguard is an oligosaccharide mainly composed of 2 Uronic acids: guluronic acid (Gul. Ac) and mannuronic acid (Man. Ac) obtained by enzymatic depolymerization from saccharides coming from brown algae.
Among the brown algae used to obtain these starting saccharides: *Laminaria hyperborea* and *Lessonia nigrescens*.



# WHAT IS SKIN NEUROAGING?

The network of nerve endings extends throughout the cutaneous layers. In 2014, scientists showed that information delivered by our touch experiences was directly decoded and analyzed by nerves, in the skin, before being sent to the brain [1]. This discovery gives new importance to the key function and place of nerve endings in the skin. It also gives rise to numerous questions on the role of nerve endings in the physiological evolution of the skin.

**How your SKIN is smarter than you think: Researchers find neurons can carry out advanced calculations to tell the brain exactly how we are being touched**

- Swedish team find skin processes touch information before sending it to brain
- Neurons in skin carry out complex calculations

By MARK PRIGG FOR MAILONLINE

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If you've ever looked in awe at smart skin being developed for robots, don't worry - your own is far more intelligent.

Neurons in human skin perform advanced calculations scientists previously believed only the brain could carry out, it has been revealed.

Researchers say that in fact our skin passes far more information to the brain that had previously been thought.

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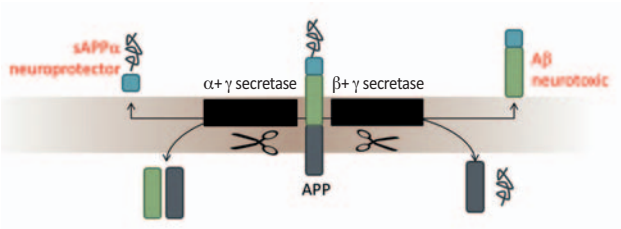
## ARE NEURONS SUBMITTED TO AGING?

Medical searches have demonstrated that nerves are subjected to aging as other cellular types; this process is called neurodegeneration [2]. The study of the neurodegeneration process has highlighted the major role of a neurotoxic peptide called amyloid beta (Aβ), which forms senile plaques on the surface of nerve endings [3].

## WHO IS Aß?

This neurotoxic peptide comes from a membrane protein known as Amyloid Precursor Protein (APP), which is located on the neuron surface. As its name implies, this protein is a precursor that can be cleaved in two different ways. First type of cleavage is triggered by a β secretase enzyme and gives rise to the liberation of neurotoxic Aβ. The second type of cleavage is triggered by a α secretase enzyme and gives rise to the liberation of a sAPPα peptide, which, contrary to Aβ, has neuroprotective properties [4].

These two cleavages exist in a balanced manner within a neuronal population that could be qualified as young. Aging, and in general the accumulation of oxidizing stress, leads to a gradual imbalance, where the cleavage liberating the neurotoxin becomes dominant [5], triggering the neuronal degeneration process that we call neuroaging.




## WHAT IS THE IMPACT OF NEUROGAING ON SKIN PHYSIOLOGY?
## IN THE SKIN, NERVES CONTINUOUSLY COMMUNICATE WITH OTHER CUTANEOUS CELLS THROUGH NEUROTRANSMITTERS. COULD NEUROAGING IMPACT THIS COMMUNICATION? COULD NEUROAGING IMPACT FIBROBLASTS ACTIVITY AND YOUTH? COULD NEUROAGING TRIGGER FIBROAGING?

# STUDY OF NEUROAGING PROCESS AND ITS CONSEQUENCES ON FIBROBLASTS ACTIVITY

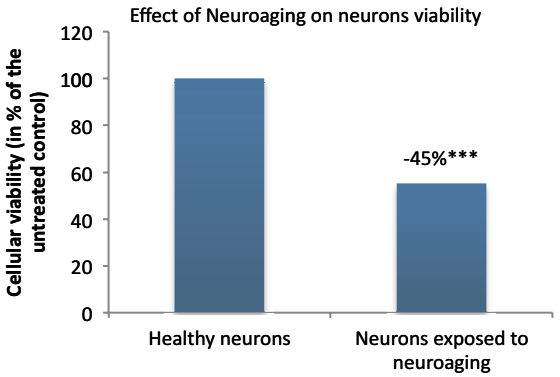
*Preliminary Study - CODIF R&N Laboratories*

The hypothesis we are testing here is whether neurons placed under Neuroaging conditions secrete messengers that are potentially toxic for fibroblasts. In the following studies, Neuroaging is simulated by the addition of the Aβ factor (1µM) to cultures of embryo cortical neurons.

## VIABILITY OF NEURONS EXPOSED TO NEUROAGING DECREASES BY 45%.***

****p<0.001 Student test*

*Protocol: simulation of neuroaging process by addition of neurotoxic Aβ (1µM) to cultures of embryo cortical neurons. Measure of cellular viability by MTT analysis after 24H incubation*

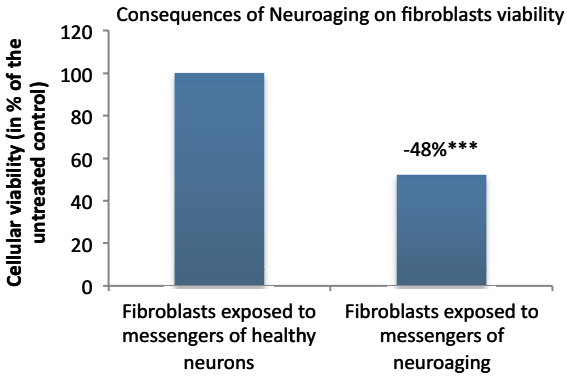



## VIABILITY OF FIBROBLASTS EXPOSED TO NEUROAGING MESSENGERS DECREASES BY 48%***

****p<0.001 Student test*

Culture medium of neurons previously exposed to neuroaging is then inoculated into human dermal fibroblast cultures. The drop of viability confirms the hypothesis that neurons exposed to neuroaging secrete messengers which directly impact fibroblasts viability.

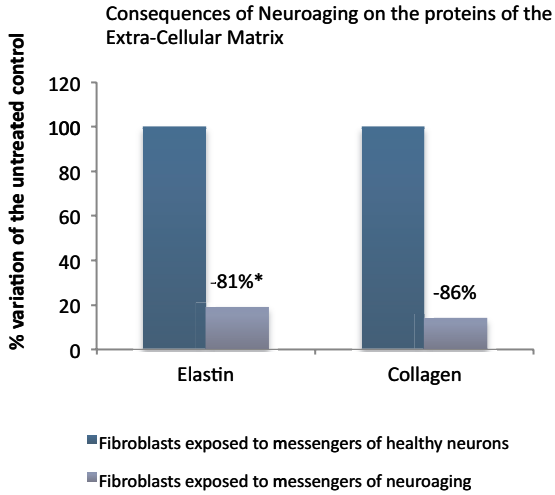
*Protocol: inoculation of culture medium of neurons exposed to neuroaging inside cultures of human dermal fibroblasts. Measure of cellular viability by MTT analysis after 48H incubation.*




## TOXICITY OF NEUROAGING TRIGGERS A DROP OF COLLAGEN AND ELASTIN SYNTHESIS

- 86% collagen synthesis
-81%* elastin synthesis
**p<0.05 Student test*

*Protocol: inoculation of culture medium of neurons exposed to neuroaging inside cultures of human dermal fibroblasts. Quantification of fibers synthesis by 2D and 3D image analysis.*




**p<0.05 Student test*
****p<0.001 Student test*

## WHEN THEY ARE EXPOSED TO NEUROAGING, NERVES RELEASE MESSENGERS THAT DIRECTLY IMPACT FIBROBLASTS VIABILITY. THE COMMUNICATION BETWEEN NERVES AND FIBROBLASTS BECOMES TOXIC AND ACCELERATES SKIN AGING.

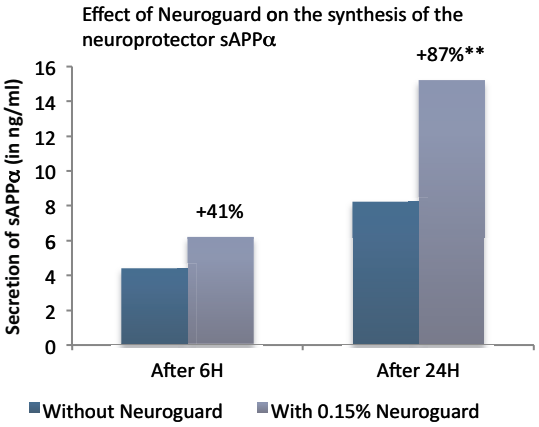
# *IN-VITRO TEST*
# NEUROGUARD PROTECTS NEURONS FROM NEUROAGING

## NEUROGUARD INCREASES THE PRODUCTION OF NEUROPROTECTOR sAPPα BY 87%.**

***p<0.01 Student test*

Meanwhile, Neuroguard provides 42% protection against H2O2 toxicity on neurons viability. Indeed, it has been demonstrated that the accumulation of free radicals leads to the gradual imbalance that appears with age between the neuroprotector sAPPα and the neurotoxic Aβ.

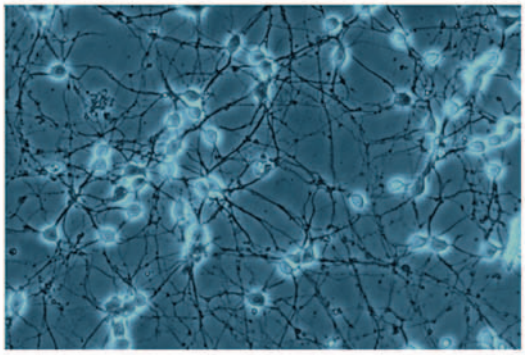
*Protocol: embryo cortical neurons cultivated with 0.15% Neuroguard for 6 and 24 hours. Quantification of sAPPα using ELISA test.*



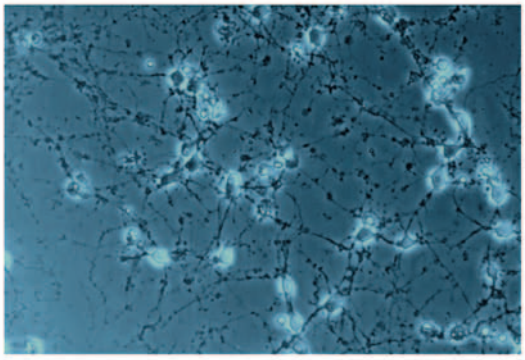

## NEUROGUARD PROTECTS NEURONS FROM NEURODEGENERATION

Whereas neurons which have not been exposed to neuroaging show active synapses, an extensive network and integral cell nuclei, the effect of neuroaging is characterized by fragmented nerve endings, "burnt-out" nuclei and a limited network. By stimulating the sAPPα neuroprotector, Neuroguard enables recovering the characteristics of neurons that have not been exposed to neuroaging.

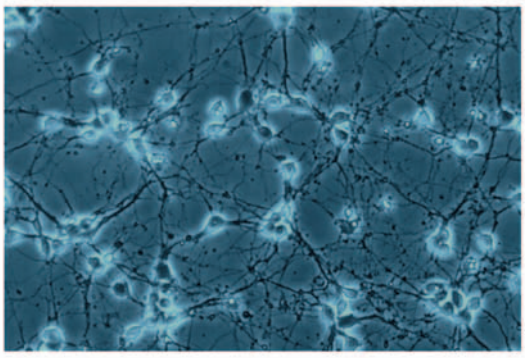
*Protocol: embryo cortical neurons cultivated with or without 1µM Aβ in presence or not of 0.15% Neuroguard for 24H.*



Culture of healthy neurons



Culture of neurons submitted to neuroaging



Culture of neurons submitted to neurogaing with 0.15% Neuroguard

## NEUROGUARD PREVENTS AND PROTECTS FROM NEUROAGING

The protective action of Neuroguard on neurons viability is effective when it is used before neuroaging starts as well as when neuroaging is already underway.

34% PROTECTION
when Neuroguard is used while Neuroaging starts.

45% PROTECTION
when Neuroguard is used before Neuroaging starts.

52% PROTECTION
when Neuroguard is used before and while Neuroaging starts.

*Protocol: embryo cortical neurons cultivated with or without 1µM Aβ in presence or not of 0.15% Neuroguard for 24H.*

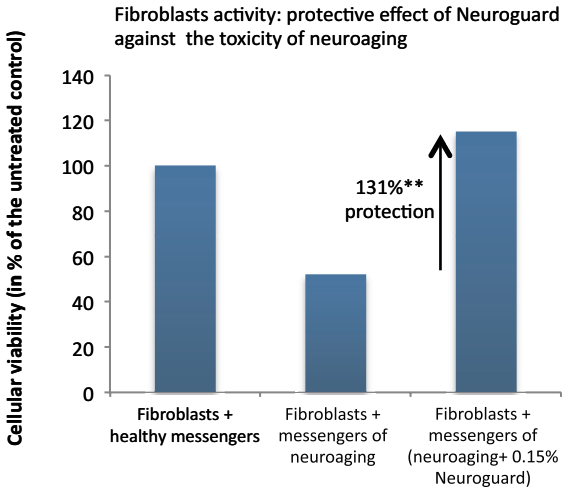
*IN-VITRO TEST*

# NEUROGUARD RE-ESTABLISHES A HEALTHY COMMUNICATION BETWEEN NERVES AND FIBROBLASTS

## NEUROGUARD PROTECTS THE COMMUNICATION BETWEEN NERVES AND FIBROBLASTS FROM THE TOXICITY OF NEUROAGING AND PRESERVES FIBROBLASTS ACTIVITY

131%** protection of fibroblasts activity.
**p<0.01 Student test

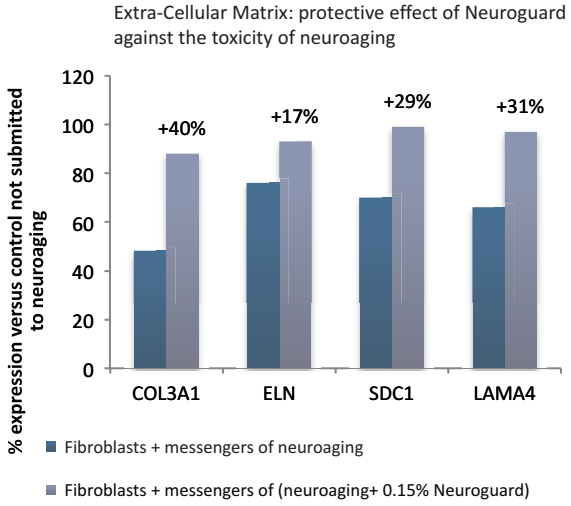
*Protocol: inoculation of culture medium of neurons exposed to neuroaging with or without 0.15% Neuroguard, inside cultures of human dermal fibroblasts. Measure of cellular viability after 48H incubation.*




## NEUROGUARD PROTECTS THE EXTRA-CELLULAR MATRIX FROM THE TOXICITY OF NEUROAGING

By protecting the activity of fibroblasts from neuroaging toxicity, Neuroguard reactivates the expression of genes coding for Extra-Cellular Matrix proteins:
+40% COLLAGEN III (COL3A1)
+17% ELASTIN (ELN)
+29% SYNDECAN (SCD1)
+31% LAMININ IV (LAMA4)

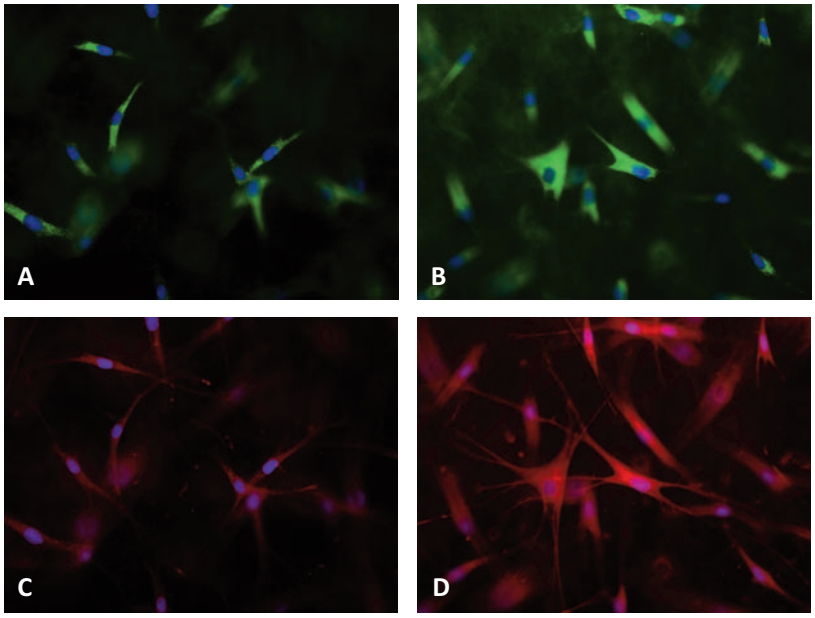
*Protocol: inoculation of culture medium of neurons exposed to neuroaging with or without 0.15% Neuroguard, inside cultures of human dermal fibroblasts. Measure of genes expression by PCR array.*




## NEUROGUARD PROTECTS THE SYNTHESIS OF COLLAGEN AND ELASTIN FROM NEUROAGING TOXICITY

+405%** COLLAGEN synthesis compared to the synthesis level reached with neuroaging conditions.
+832%* ELASTIN synthesis compared to the synthesis level reached with neuroaging conditions.
*p<0.05 ;**p<0.01 Student test

*Visualization of collagen fibers (green fluorescence) in cultures of fibroblasts exposed to neuroaging messengers without (A) and with (B) 0.15% Neuroguard. Same conditions to visualize elastin synthesis (red fluorescence), without (C) and with (D) 0.15% Neuroguard.*



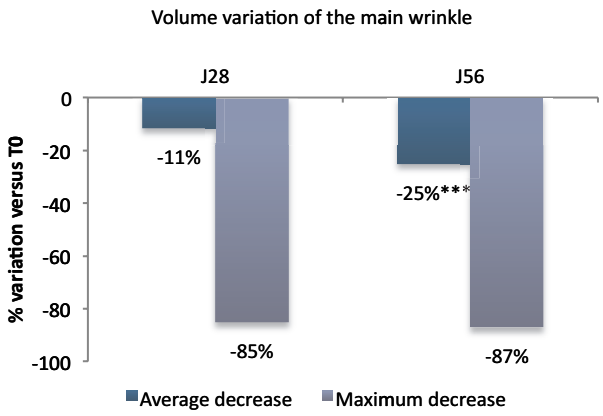
# *IN-VIVO TEST*
# EFFICACY OF NEUROGUARD P ON DEEP WRINKLES

*PROTOCOL*
*20 volunteers aged 62 to 74.*
*Twice daily applications of a cream formulated with 1.5% Neuroguard P for 56 days.*
*Application on the whole face.*
*Analytical method: fringe projection.*

## EFFECT OF NEUROGUARD P ON THE VOLUME OF CROW'S FEET WRINKLES

After 28 days treatment:
-11% on average
And up to -85%

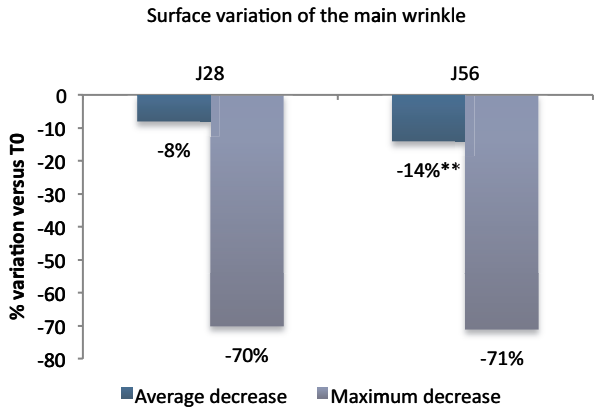
After 56 days treatment:
-25%*** on average
And up to -87%
***p<0.001 Student test




## EFFECT OF NEUROGUARD P ON THE AREA OF CROW'S FEET WRINKLES

After 28 days treatment:
-8% on average
And up to -70%

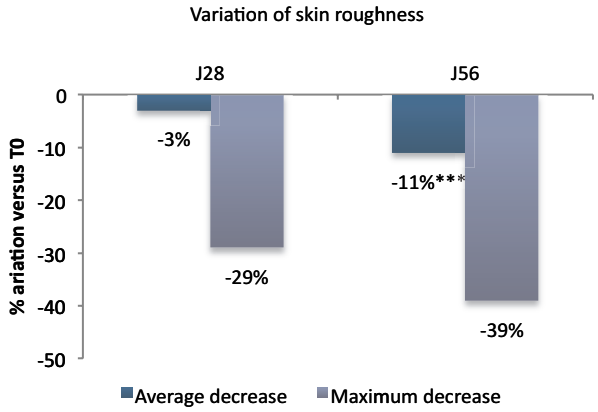
After 56 days treatment:
-14%** on average
And up to -71%
**p<0.01 Student test




## EFFECT OF NEUROGUARD P ON SKIN ROUGHNESS

After 28 days treatment:
-3% on average
And up to -29%

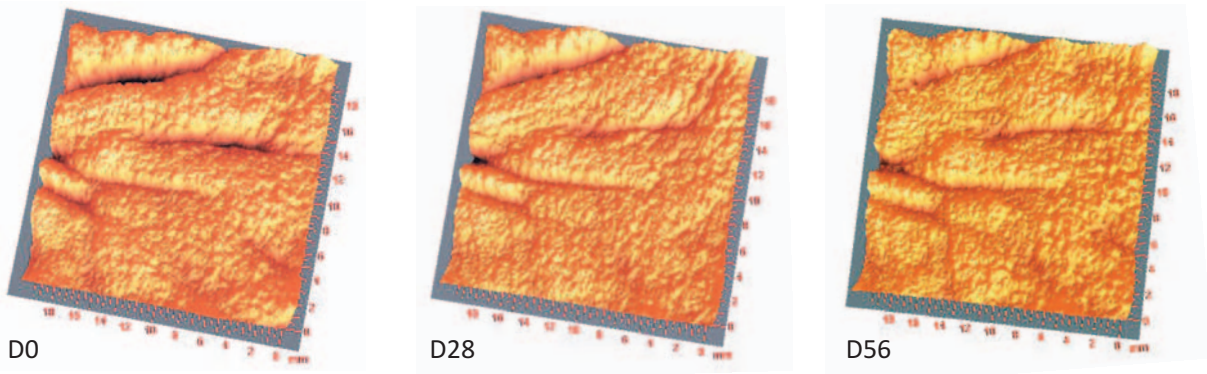
After 56 days treatment:
-11%*** on average
And up to -39%
***p<0.001 Student test



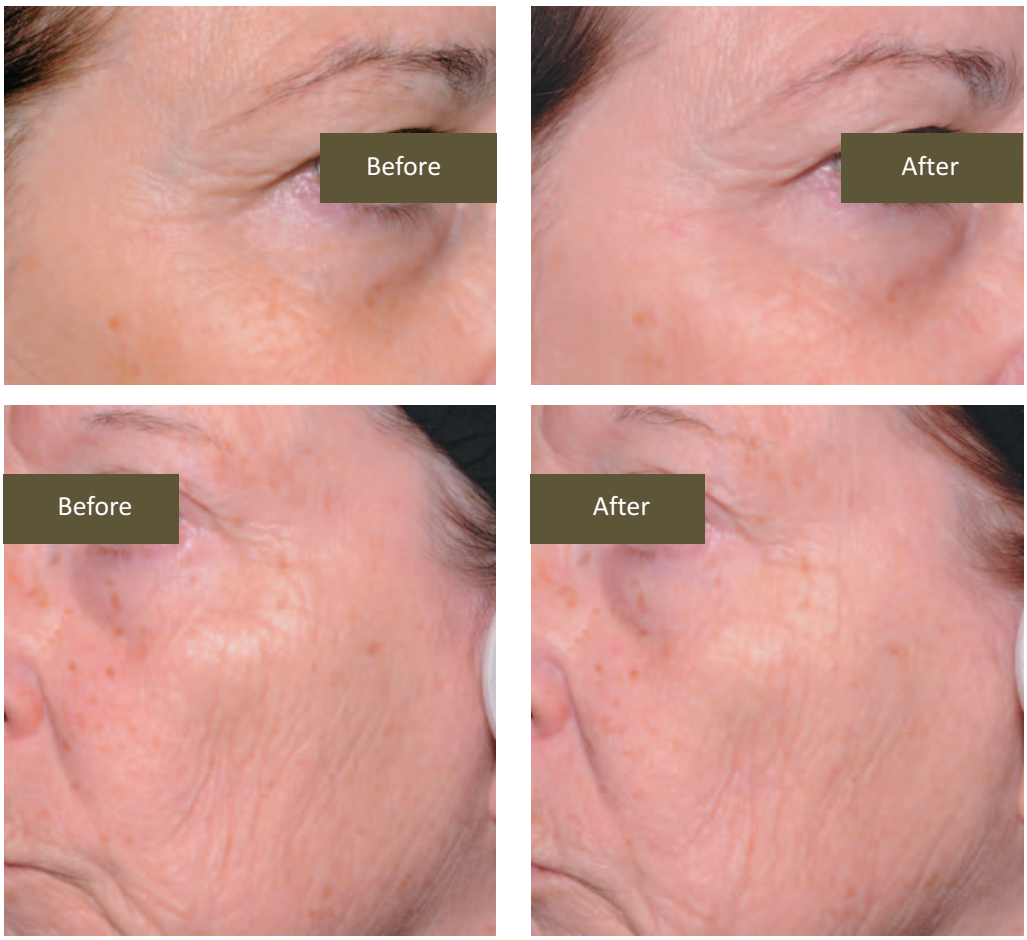
*IN-VIVO TEST*

# VISUALIZATION OF THE EFFICACY OF NEUROGUARD P ON DEEP WRINKLES

## DECREASE IN THE VOLUME AND AREA OF CROW'S FEET WRINKLES




## OVERALL EFFICACY




NEUROGUARD REVOLUTIONIZES NEUROCOSMETIC BY DIRECTLY TARGETING THE AGING OF NERVE ENDINGS. ITS NEUROPROTECTIVE ACTION BLOCKS NEUROAGING AND RE-ESTABLISHES A HEALTHY COMMUNICATION BETWEEN NERVES AND FIBROBLASTS, THUS PROTECTING DERMAL CELLS FROM PREMATURE AGING. UNDER THESE CONDITIONS, COLLAGEN AND ELASTIN SYNTHESIS ARE REACTIVATED, DEEP WRINKLES ARE DIMINISHED AND SKIN TEXTURE IS SMOOTHER.

# FORMULATION GUIDELINE
## INTENSIVE SKIN COM REPAIR

This anti-aging cream has been formulated with two Neurocosmetic active ingredients: NEUROGUARD P and an ingredient targeting dark spots: NEUROLIGHT 61G.

| Phase | Raw Material / Trade name | INCI name | % |
|---|---|---|---|
| A | distilled water | aqua | 64,73 |
| | elestab cpn | chlorphenesin | 0,27 |
| B | synthalen m | Carbomer | 0,4 |
| C | glycerin bidistillee codex | glycerin | 3 |
| | butylene glycol 1-3 | butylene glycol | 2 |
| | keltrol cgsft | xanthan gum | 0,2 |
| D | eumulgin ba 25 | beheneth-25 | 2,5 |
| | plurol diisostearique cg | polyglyceryl-3 diisostearate | 0,5 |
| | nacol 22-98 | behenyl alcohol | 2 |
| | lanette o | cetearyl alcohol | 1 |
| | silicone dc 580 wax | stearoxytrimethylsilane stearyl alcohol | 1 |
| | cetiol cc | dicaprylyl carbonate | 2 |
| | crodamol gtcc / miglyol 812 / waglinol | caprylic/capric triglyceride | 3 |
| | silicone (dimethicone (100cs)) | dimethicone | 2 |
| | cegermil | zea mays oil (and) glycine soja oil (and) helianthus annuus seed oil | 3 |
| | huile jojoba lite | simmondsia chinensis oil | 2 |
| | cremeol sbe / cegesoft sbe | butyrospermum parkii butter extract | 2 |
| | lipocire a pastilles | c10-18 triglycerides | 2 |
| | phenoxyethanol | phenoxyethanol | 0,8 |
| E | soude (solution 5 n) | Aqua (and) sodium hydroxide | 0,5 |
| F | jurimer - sj touch 1 | polymethyl methacrylate (and) aqua | 2 |
| G | NEUROGUARD P | Aqua (and) algae oligosaccharides (and) phenoxyethanol | 1,5 |
| | NEUROLIGHT 61G | Glycerin (and) aqua (and) pancratium maritimum extract | 1,5 |
| | parfum fleur rose r13386 | Parfum (and) geraniol (and) coumarin (and) isoeugenol (and) citronellol (and) benzyl alcohol (and) limonene citral | 0,1 |

**Protocol :**
Heat up phase A to 80°C with gentle stirring. Disperse B into phase A with strong stirring (1500rpm) during 30min. Add premix C with strong stirring (1500rpm) during 15min. Heat up phase D to 80°C. Emulsify phase D into phase ABC with very strong stirring (2500rpm) during 10min. Add E with very strong stirring (2500rpm) during 20min. Cool down to 50°C with gentle stirring. Add F with gentle stirring. Cool down to 35°C with gentle stirring. Add perfume and actives (G) with gentle stirring during 15min.

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# NEUROGUARD : THE NEW SCIENCE OF AGING

## COSMETIC ACTIVITY

***Prevents from Neuroaging***

Protects neurons from the toxicity of free radicals

Stimulates the synthesis of the neuroprotector sAPP$\alpha$

Protects neurons from neuroaging

***Protects the communication between nerves and fibroblasts***

Re-establishes a healthy communication between nerves and fibroblasts

***Prevents from Fibroaging***

Protects collagen synthesis

Protects elastin synthesis

***Decreases deep wrinkles***

Significantly decreases wrinkles depth

Significantly decreases wrinkles area

Significantly decreases skin roughness

## INCI NAME

NEUROGUARD P: Water (and) Algae oligosaccharides (and) Phenoxyethanol
NEUROGUARD G: Glycerin (and) Water (and) Algae oligosaccharides

## RECOMMENDED % OF USE

NEUROGUARD P: 1,5%
NEUROGUARD G: 3%

CHARACTERISTICS: hydrosoluble active ingredient from marine origin – 100% natural

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