

# An innovative approach for wrinkle treatment: the role of in vivo aging markers in the efficacy of a novel cosmetic formulation

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## Introduction:

- Wrinkles become prominent during aging, resulting in horizontal marks on the forehead, nasolabial folds, and periorbital area<sup>1,2</sup>;

- These changes are driven by modifications in skin molecular composition, such as:



- These alterations generate important functional and structural consequences to the skin<sup>6</sup>:

- increased fragility,
- lack of cell proliferation and migration,
- homeostasis impairment, and
- weakening of skin support

- The aim of this study was to evaluate the efficacy of a novel cosmetic formulation (CHR-1), containing an innovative combination of active ingredients, targeting wrinkle reduction, and its efficacy in stimulating different anti-aging markers in clinical trials.

## Materials & Methods:

### Clinical trials

- Resolution 466/12 of the National Council of Health on Regulatory Guidelines and Standards for Research Involving Humans (n° 35243820.0.0000.5514)

- Subjects: women aged 18-60 yrs, phototype Fitzpatrick I-VI, with different grades of wrinkles (light, moderate, or intense)

- Treatment: CHR-1 was randomly applied in the face and neck, twice a day, for 60d. The control group was set as the skin without application of any product.

### Wrinkle evaluation

- Assessed using image analysis in specific regions of the face: periorbital, nasolabial, and frontal area.

### Skin hydration and cutaneous micro-relief

- Measured using Corneometer and prophylometric analysis

### Dermal density

- Appraised with High-Frequency Ultrasound (HF-US)

### Anti-aging markers assessment

- Evaluated by protein levels in human skin biopsies by immunofluorescence analysis:



### Statistical analysis

- For anti-aging markers: comparison between groups were performed using one-way ANOVA followed by Bonferroni post-test

- Other tests: performed using Student's t-test for independent samples

- For all comparisons p-value was set at 0.05.

## References:

1. Lemperle G (2001) A classification of facial wrinkles. *Plast Reconstr Surg* 106:1733-1734.
2. Zhao Y, Feng B., Lee J et al (2020) A multi-layered model of human skin elucidates mechanisms of wrinkling in the forehead. *J Mech Behav Biomed Mater* 105:103694.
3. Kruglikov I L, Scherer P E (2018) Skin aging as a mechanical phenomenon: the main weak links. *Nutr Healthy Aging* 4:291-307.
4. Giangreco A, Goldie S J, Failla V et al (2010) Human skin aging is associated with reduced expression of the stem cell markers b1 integrin and MSCP. *J Invest Dermatol* 130:604-608.
5. Makrantonaki E, Zouboulis C C (2007) Characteristics and pathomechanisms of endogenously aged skin. *Dermatology* 214:352-360.
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## Results & Discussion:

### Wrinkle evaluation

- 100% of subjects showed improvement in wrinkles and fine lines after 14d, and the reduction of wrinkles after 60d of treatment.

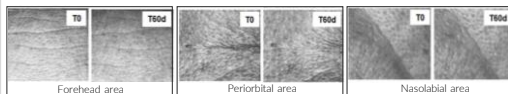


Figure 1. Image analysis of wrinkles in the frontal, periorbital and nasolabial areas and their reduction after 60 days of use of the cosmetic formulation.

### Skin hydration and cutaneous micro-relief

- After 15 min of product application there was a significant reduction in forehead skin roughness by 10.8%. After 2h, a significant reduction of 7.4% in skin roughness was observed.

- Also, the product was able to provide a significant increase in skin hydration up to 37% after 15 min of the application and kept the skin moisturized up to 48h

### Dermal density

- The use of the product resulted in a significant increase in dermal density and a better functional structure in all evaluated facial regions due to the horizontal and more compact organization of the collagen bundles:

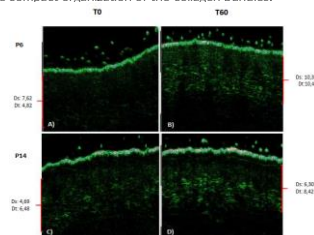


Figure 2. High-Frequency Ultrasound image obtained from subjects P6 and P14 before and after 60d of treatment.

### Anti-aging markers assessment

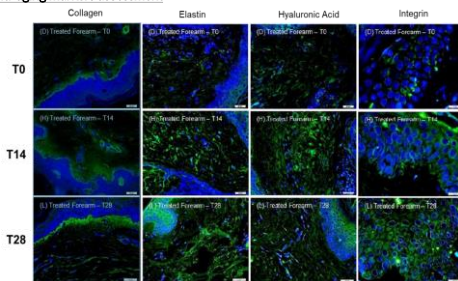


Figure 3. Immunofluorescence evaluation of biological markers in skin biopsies after 14d and 28d after treatment. Molecules of interest are stained in green and blue color corresponds to the cell nucleus. The reference bar corresponds to 10 µm.

## Conclusions:

- Skin molecules, e.g., collagen, integrins, and others, are key factors related to aging and their regulation can improve wrinkle treatment

- Our results showed the potential of biomarkers assessment aligned with other clinical tests to provide a deep understanding of the action mechanism of a cosmetic formulation.