Oxygenation for Skin Rejuvenation: A Comprehensive Study on the Age-Related Decline in Skin Oxygen Levels and the Efficacy of a Novel Oxygen-Delivery Technology

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Abstract

Hypoxia is a significant tissue stressor, has been linked to inflammatory cell infiltration and diminished collagen synthesis in the skin, adversely affecting its regenerative capabilities. Oxygen is essential for various cellular functions, including proliferation, migration, and tissue neo-synthesis, underscoring its importance in skin regeneration and energy overall. This study explores a novel topical oxygen-based therapeutic complex designed to enhance tissue re-oxygenation and provide anti-aging benefits. We evaluated the efficacy of this innovative oxygen delivery system through *in vitro* experiments on cells and on reconstituted epidermis, and assessed its impact on skin oxygenation levels in human subjects. The results demonstrate significant improvements in cellular oxygenation and several downstream effects, suggesting potential for enhancing skin health and anti-aging outcomes.

Keywords: oxygen; hypoxia; anti-aging; cell proliferation; collagen synthesis

Introduction

Oxygen plays a fundamental role in several cellular processes, mainly, in the production of ATP as the terminal electron acceptor in the inner mitochondrial membrane. Previous studies have suggested a correlation between aging and a decline in availability of cellular oxygen and thus, may impact the skin [1]. Barley extracts derived from the *Hordeum vulgare* plant are chlorophyll-rich have garnered considerable attention in skincare owing to their abundant composition of antioxidants, polyphenols, vitamin E, and minerals, and may serve as a sustainable, abundant source of oxygen if solubilized [2]. The objective of this study was to assess the capabilities of a unique extract procured via an exclusive cultivation process that stimulates the concentration of oxygen and endogenous plant metabolites to re-oxygenate and optimize skin health and benefits.

Materials and Methods

Normal human keratinocytes were treated for 72 hours and cell ATP content was assessed by using a bioluminescence-based microplate assay. ATP levels were normalized against total cellular protein content. Cell proliferation was assessed on normal human fibroblasts upon treatment for 24 hours, and quantified by total DNA content by using the CyQuant Cell Proliferation Assay kit. After 72 hours of treatment, Collagen I synthesis was quantified by ELISA. Oxygen levels were measured on reconstructed human epidermis (Mattek Epiderm) incubated with dimethyloxalylglycine diluted in culture medium to repoduce hypoxia conditions. Oxygen content was carried out by a variant of the Winkler method. *In vivo*, skin oxygen levels were measured in 136 healthy women on the inner forearm using a polarographic probe (Clark method). Tolerance and Efficacy on Atopic prone skin patients was evaluated by a Dermatologist, via clinical examination, clinical scoring and instrumentation using TEWAMETER® method: Tewameter 300® (Courage+Khazaka,

electronic GmbH). 2 panels of 21 and 23 atopic prone skin patients in average 46,8 y.o. tested alternatively a formula with oxygenating agent or the placebo formula.

Results

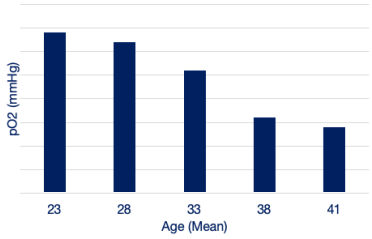

Figure 1: Oxygen partial pressure for female subjects. Mean age of the group is given [1].

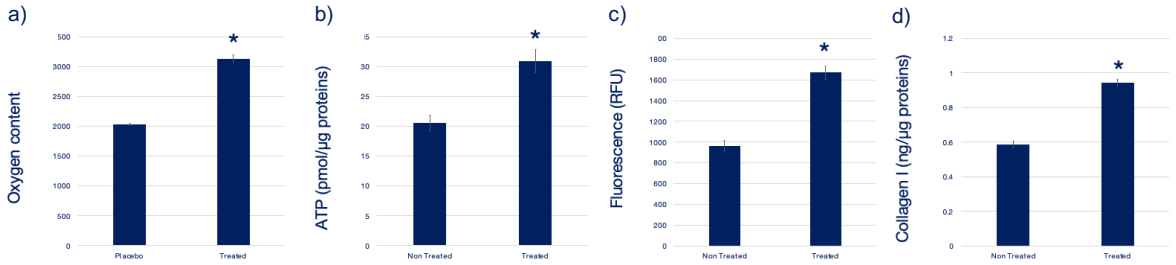

Figure 2: a) Oxygen content on Reconstructed Human Epidermis, mean±sd, n=3, * p<0.05 compared to non-treated. b) ATP synthesis on normal human keratinocytes, mean±sd, n=5, * p<0.01 compared to non-treated. c) Normal Human Fibroblasts proliferation, mean±sem, n=4, * p<0.05 compared to non-treated. d) Collagen I synthesis by normal human fibroblasts, mean±sd, n=3, * p<0.01 compared to non-treated

Results from the measurement of oxygen levels on healthy subjects' skin revealed a clear decrease with age. Notably, it indicated that oxygen content in the skin can decrease by up to 50% between the 20s and the 40s, making it a potential marker of skin aging (Fig. 1). *In vitro* assessments demonstrated significant improvements attributed to the oxygenating agent. Keratinocytes exhibited a significant increase in ATP levels after the application of the active ingredient (Fig. 2b). The oxygenating agent demonstrated a substantial reoxygenating effect on reconstituted epidermis under hypoxic conditions, emphasizing its potential role in enhancing tissue oxygenation (Fig. 2a). The anti-aging effect of the oxygen delivery method technology was further substantiated by improved collagen synthesis and cellular proliferation in fibroblasts (Figs. 2c,d). In the clinical study, Dermatologist found a

good tolerance of the tested product and atopic patients appreciated its action on comfort, redness, reduction of irritations (Figs. 3-4).

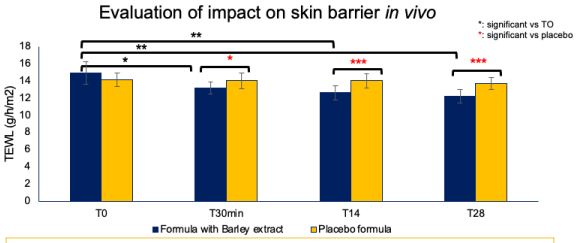

Figure 3: *in vivo* study on atopic prone patients, Instrumental measurement of TEWL
* Or * : * p<0.05 ; **p<0.01 ; ***p<0.001

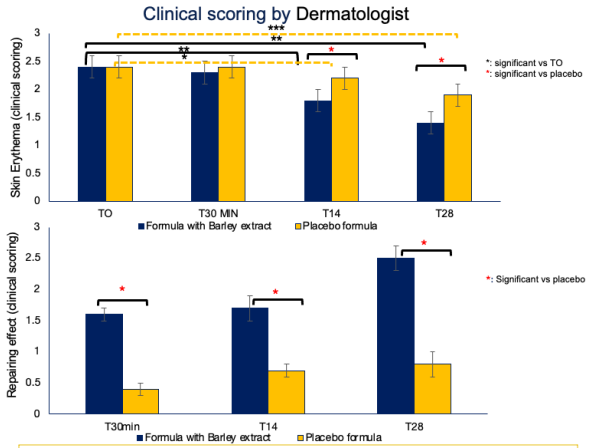

Figure 4: *in vivo* study on atopic prone patients, clinical scoring by Dermatologist
* Or * : * p<0.05 ; **p<0.01 ; ***p<0.001

Discussion

Our study presents a promising approach to support skin regeneration and address skin aging by harnessing the benefits of oxygenation. The significant decline in skin oxygen levels observed with age, coupled with the positive outcomes of our oxygenating ingredient, emphasizes its potential as a valuable addition to specific skincare treatments aimed at maintaining skin youth and function. The active ingredient not only displayed an energizing potential but also exhibited the ability to reoxygenate cells and reconstituted epidermis in hypoxic conditions *in vitro*. Moreover, it demonstrated the potential to boost collagen synthesis and cell proliferation, affirming its anti-aging effects. This innovative technology offers a comprehensive strategy to promote skin rejuvenation, presenting new opportunities for advancing anti-aging skincare solutions.

Conclusion

In this study, we demonstrated that topical application of the oxygenating agent can deliver oxygen to skin and promote several downstream effects such as increase in ATP, upregulation of cellular proliferation and key biomarkers of skin health. Taken together, these suggest that the oxygenating agent may ameliorate compromised skin barriers, potentially elevating skin hydration and comfort among patients afflicted by conditions such as Eczema and Atopic Dermatitis. The promising outcomes have laid the groundwork for ongoing clinical trials, intending to validate and expand upon the observed benefits.

Conflict of Interest Statement

This research was funded by Coty Inc.

References

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