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“Advancing Hair Care: Unlocking the science of hair reconstruction and the power of adaptive natural solutions”

Rebeca Gasparin ^{1*}, Juliana Amado ^{2*}, Rita Cartaxo ³, Maria Reichenbach ⁴, Marcia Paula ⁵, and Dominik Stuhlmann ⁴

¹ Hair Care Center, Global Innovation Cosmetic Ingredients, Symrise, Cotia, Brazil; ² Advanced Actives - Hair Actives, Cosmetic Ingredients, Symrise, Paris, France; ³ Advanced Actives - Hair Actives, Cosmetic Ingredients, Symrise, Cotia, Brazil; ⁴ Global Innovation Cosmetic Ingredients, Symrise, Holzminden, Germany; ⁵ Advanced Actives - Hair Actives, Cosmetic Ingredients, Symrise, New Jersey, USA

1. Introduction

Hair fibers are keratin-based materials with distinct structures, each serving specific functions and exhibiting unique properties. The outermost layer, the cuticle, contributes to hair's appearance and acts as a protective shield against external stressors. Beneath it, the cortex—the inner part of the fiber—determines hair's strength and mechanical behavior. Meanwhile, the cell membrane complex (CMC) plays a key role in maintaining the integrity of the hair structure and controlling diffusivity [1].

The function of each hair structure is directly linked to its chemical composition and architecture. Cuticle cells consist of multiple layers, displaying a gradient of cystine content that decreases from the outermost to the innermost regions. This arrangement results in a harder exterior while the inner layers become progressively softer toward the protein-lipidic CMC, which connects cuticle cells and transitions toward the cortex. The cuticles are coated externally by a lipid layer rich in 18-methyl eicosanoic acid (18-MEA), which gives hair its natural hydrophobicity. Inside the cortex, α -keratin is organized into intermediate filaments, surrounded by a matrix of amorphous keratin and keratin-associated proteins (KAPs). Together, these components form cortical cells that are interconnected via the CMC [1,2].

Damage to any of these structures directly affects their properties and influences how hair fibers interact with their environment. Oxidative damage leads to the breakage of chemical bonds throughout the hair, making it more charged and less hydrophobic. This, in turn, impacts hair permeability, sensory attributes, and mechanical properties [1,3,4].

Concerns about damaged hair have become so widespread among cosmetic consumers that it is now recognized as a distinct hair type [5]. While consumers remain drawn to hair transformations, they actively seek effective and sustainable ways to restore their hair's health.

This study aims to develop and evaluate naturally derived, biodegradable materials as sustainable and effective solutions for addressing damaged hair. By integrating solutions tailored

to specific hair damage issues, a comprehensive and adaptive 360° approach to hair reconstruction can be developed.

2. Materials and Methods

2.1 Hair samples

Regular and platinum bleached Caucasian hair samples (25 cm in length) were sourced from International Hair Importers & Products Inc.

2.2 Hair preparation

2.2.1 Cleansing

The hair was sectioned into tresses and pre-cleansed using a 10% sodium laureth sulfate (SLES 10%) solution. Each tress was wetted under tap water for 30 seconds (flow rate: 4 L/min; temperature: 33 ± 3 °C), followed by the application of SLES 10% at a dosage of 0.1 mL per gram of hair. The tresses were massaged for 1 minute, then rinsed under identical conditions for 1 minute. Excess water was removed by gently passing each tress between two fingers. The hair tresses were subsequently left to dry overnight under controlled environmental conditions (22 ± 2 °C and $55 \pm 5\%$ relative humidity – RH).

2.3 Hair Care Ingredients

2.3.1 Vegetable-Derived Small Peptides (SPO and SPL)

Aqueous solutions of oat (SPO - INCI Name: Water (Aqua) (and) Avena Sativa (Oat) Peptide (and) 1,2-Hexanediol (and) Caprylyl Glycol) and lupin (SPL - INCI Name: Water (Aqua) (and) Hydrolyzed Lupine Protein (and) 1,2-Hexanediol (and) Caprylyl Glycol (and) Benzoic Acid (and) Sodium Benzoate) small peptides (< 2 kDa) with selected physicochemical characteristics were obtained via enzymatic hydrolysis with a tailor-made enzymatic cocktail.

2.3.2 Vegetable-Derived Protein (SHR)

A native high molecular weight protein (gliadin) was obtained from wheat via hydro/alcoholic extraction (SHR - INCI Name: Glycerin (and) Triticum Vulgare (Wheat) Protein (and) Water).

2.3.3 Vegetable Oily Extract (SHSC)

An oily wheat fraction was obtained from the vegetable material extraction in linoleic acid and blended with Camelia oil and selected emollients (SHSC – INCI Name: Cetearyl Nonanoate (and) Triticum Vulgare (Wheat) Germ Oil (and) Caprylic/Capric Triglyceride (and) Linoleic Acid (and) Triticum Vulgare (Wheat) Bran Extract (and) Triticum Vulgare (Wheat) Germ Extract (and) Camellia Oleifera Seed Oil).

2.4 Cosmetic Formulations

Shampoo and hair conditioner formulations - including both rinse-off and leave-on variants - were prepared with one or more ingredients outlined in Section 2.3 at concentrations ranging from 0.5% to 2% for subsequent application to hair tresses.

2.5 Product Application

The prepared cosmetic formulations were applied to the hair tresses according to the procedure outlined below. Following treatment with the selected formulations, the tresses were left to dry overnight under controlled conditions (22 ± 2 °C and $55 \pm 5\%$ RH).

2.5.1 Shampoo

The hair tresses were wetted under tap water for 30 seconds at a flow rate of 4 L/min and a temperature of 33 ± 3 °C. A shampoo formulation was then applied to each tress at a dosage of 0.1 mL per gram of hair, followed by a standardized massage for 1 minute. The tresses were subsequently rinsed for 1 minute under the same water flow and temperature conditions. Finally, excess water was removed by gently passing each tress between two fingers.

2.5.2 Rinse-off Conditioner

Hair conditioner was applied to wet tresses at a dosage of 0.1 mL per gram of hair. The hair was massaged for 1 minute and allowed to rest for 3 minutes. The tresses were then rinsed for 1 minute under a water flow of 4 L/min at 33 ± 3 °C, and excess water was gently removed by passing each tress between two fingers.

2.5.3 Leave-on Conditioner

The leave-on formulation was applied to wet tresses at a dosage of 0.1 mL per gram of hair. The hair was then massaged for 1 minute to ensure even distribution.

2.6 Efficacy evaluation

2.6.2 Fatigue Test

Fifty bleached hair fibers from each treatment group were subjected to a fatigue test using a Cyclic Tester (CYC801, Dia-Stron, UK). The test was conducted under a constant strain of 5%. Weibull distribution parameters were extracted using UVWin software (Dia-Stron, UK) for statistical assessment of fiber durability.

2.6.3 Breakage Test (Repeated Grooming)

Six bleached hair tresses (2.5 g each) underwent a repeated grooming assessment comprising five cycles of 1,000 combing strokes per cycle. The combing procedure was executed using an Automated Combing Machine (BLPA-300, Bioluz, BRL). Fragments of broken fibers were systematically collected and quantified following each cycle to evaluate mechanical resistance and damage prevention.

2.6.6 Statistical Analysis

Statistical analysis was performed using XLSTAT (Lumivero, USA). Inter-group comparisons were conducted using the Kaplan-Meier method and log-rank test (Fatigue test) or Student's t-test for mean differences (Breakage test), at a 95% confidence interval to determine significant effects.

3. Results

Fatigue Test

The survival probability curves obtained from the fatigue measurements are presented in Figure 1, while the characteristic life (α) values are summarized in Table 1.

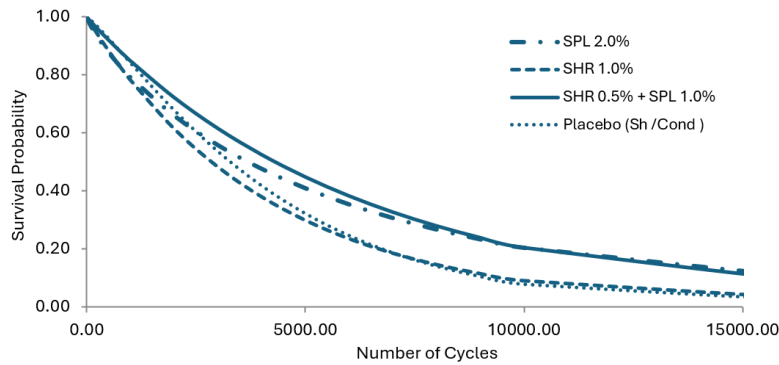


Figure 1. Survival probability curves for regular bleached hair (IHIP) treated with shampoo and conditioner formulations, either containing lupin peptides (SLP) and/or wheat native protein (SHR) or a placebo formulation without these components.

Table 1. Characteristic life (α) values derived from fatigue test results for regular bleached hair (IHIP) treated with shampoo and conditioner formulations, either incorporating lupin peptides (SLP) and/or wheat native protein (SHR) or using a placebo formulation without these components. * $p < 0.05$ log-ranks.

Group	α value
SPL 2.0%	5707
SHR 1.0%	4140
SHR 0.5% + SPL 1.0%	6248*
Placebo	4498

The formulations containing SHR at 1.0% or SPL at 2.0% did not demonstrate a significant enhancement in the survival probability profile of damaged hair compared to the placebo treatment. However, the combined application of SHR at 0.5% and SPL at 1.0% resulted in a 39% increase in characteristic life relative to the placebo, indicating a synergistic effect in improving hair fiber resilience.

Breakage Test (Repeated Grooming)

Figure 2 presents the results of the breakage assessment conducted on platinum bleached hair tresses (IHIP), illustrating the effects of the applied treatments on fiber integrity.

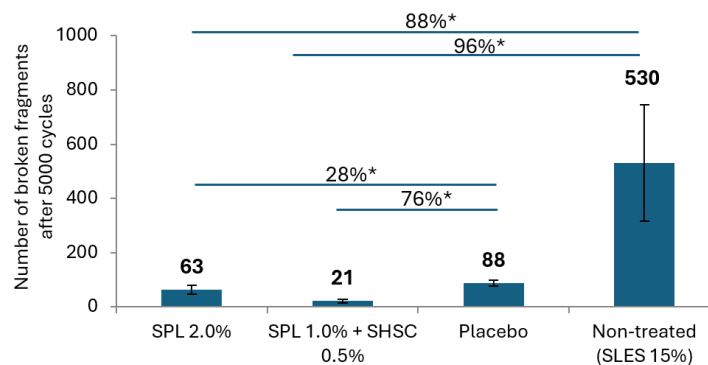


Figure 2. Breakage test results obtained for platinum bleached hair (IHIP) treated with leave-on conditioner formulation containing or not (placebo) lupin peptides (SLP) and/or oily wheat extract (SHSC) versus non-treated control (SLES 15%) after 5000 combing cycles. Percent values represent the improvement against references. * $p < 0.05$ versus placebo or non-treated control.

All tested treatments statistically improved hair breakage results compared to the non-treated control. Both treatments with formulations containing SPL and SPL+SHSC were more efficient in reducing hair breakage due to repeated grooming than placebo-treated hair.

4. Discussion

The oxidation process involved in the bleaching of hair fibers is not melanin-selective and ends up provoking the cleavage of chemical bonds of the keratin structure and other associated proteins. The lipids present in the hair structure, both internally and externally, are also affected by bleaching. Thioester bonds that link the lipids to the protein are broken in the process, making them easy to remove from hair. This oxidative process results in the generation of charges and dramatically alters hair's physicochemical characteristics. Bleached hair is finally a polar, porous, prone to break, dull material [1,3,4].

The polar bleached hair presents a high affinity with polar amino acids or small peptides, which, due to their low molecular weight, can diffuse into the hair cortex and interact with the charged sites [1,6]. Due to their special composition, SPO and SPL are able to deeply penetrate the hair fibers and establish new bonds within the hair cortex structure. The new bonds established between these protein materials and the hair structure collaborate to stabilize hair's internal structure and recover mechanical properties of the hair, improving its resistance to mechanical stress [7].

Big native proteins, on the other hand, can easily deposit on the hair surface. Depending on their physicochemical characteristics, those proteins can present good affinity to damaged sites on the hair surface [6], thus being able to fill in holes and fractures or glue split ends together. SHR was demonstrated to present high affinity to damaged areas on the hair surface, thus delivering the mentioned benefits. In addition, this ingredient contributes to reducing friction during hair combing.

Lipidic materials with specific sizes and compositions, such as SHSC, can both penetrate the hair fibers by diffusing via the CMC, replacing the lost components of this structure, and deposit on the hair surface, restoring the hair's natural hydrophobicity and lubricity [8]. Moreover, SHSC showed efficacy in protecting hair color from fading caused by washing and improving hair volume/frizz control and curl retention. The ingredient was also efficient in promoting hair cuticle sealing, enhancing hair combability, and smoothness [9].

The fatigue test represents an instrumental way to simulate the response of hair fibers to the repeated stresses to which they are subjected every day. These small but repeated stresses generate fractures in the hair fiber, which propagate until hair breakage. Non-damaged and well-conditioned hair is able to better accommodate the stress and to postpone fracture propagation, resulting in bigger survival probabilities and alpha values. The characteristic life or alpha (α) parameter represents the cycle in which 63.2% of the fibers have already broken, indicating the level of resistance of fibers to fracture [10]. At the studied dosage, neither small peptides nor big proteins were effective in increasing hair survival probability compared to the placebo. However, the association of both ingredients at even smaller dosages increased the alpha by 39%. This result suggests that a synergistic effect was achieved when combining ingredients with different mechanisms of action and targets within the hair structure.

The breakage assessment based on repeated grooming evaluates not only hair's resistance to break due to fibers's strength but also the hair lubricity and easiness to comb. Oxidative damage makes the hair easier to break in this test by reducing both its resistance and lubricity. When hair is well conditioned, it is easier to comb and less fibers are broken after repeated

grooming processes [1,11]. Placebo formulation alone significantly reduced the number of broken fragments compared to non-treated control. The addition of SPL at 2% improved the placebo formulation performance in 28%. This result can be attributed to the cortex structure enhancement due to the penetration and bond repair effect of the polar small peptides present in the ingredient. However, the best result, in terms of anti-breakage effect, was obtained with the combination of SPL at 1% and SHSC at 0.5%. While the lupin peptides work on the cortex repair, the rich oily composition of SHSC penetrates the CMC, improving hair cohesivity and maleability, and enhances hair lubricity by forming a hydrophobic film on the hair surface.

5. Conclusion

The results demonstrated that small peptides derived from vegetable sources effectively penetrate the hair cortex, interact with damaged sites, and enhance the mechanical properties of hair. Larger non-hydrolyzed proteins and other natural film-forming agents significantly improved the condition of damaged hair by enhancing its sensory and surface attributes. Furthermore, natural oily extracts contributed to increased hair cohesion, thereby regulating its permeability. By combining these technologies, a comprehensive 360° hair reconstruction approach was achieved, encompassing deep penetration, replacement of the cell membrane complex (CMC), and surface coating. This study provides a compelling example of how natural, sustainable, and biodegradable materials offer innovative solutions for complete hair reconstruction.

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