

Impact of oxidative hair coloring and bleaching on hair fiber properties

Maria Valéria Robles Velasco^{1*}, Nathalia Magnocavallo¹, Árina Colombane Câmara¹, Claudinéia Aparecida Sales de Oliveira Pinto¹, Michelli Ferrera Dario^{1,2}, Robson Miranda da Gama^{3,4}, André Rolim Baby¹

¹Faculty of Pharmaceutical Sciences, University of São Paulo, São Paulo, Brazil. ²Department of Pharmaceutical Sciences, Federal University of São Paulo (UNIFESP), Diadema, Brazil. ³University Center FMABC, Santo André, Brazil. ⁴Santo Amaro University Center, São Paulo, Brazil.

*Correspondence: mvrobles@usp.br

Abstract

Introduction: Hair coloring is a major segment of the cosmetics market, with 51% of Brazilian households using hair dyes. Given their widespread use and potential adverse effects on fiber integrity, it is important to understand the structural and chemical impacts of bleaching and coloring treatments on hair.

Objective: To evaluate and compare the effects of professional and at-home hair coloring treatments on hair fiber properties—combability, color, and protein loss—in dark-brown virgin hair strands.

Methods: Straight, virgin dark-brown hair strands (15 cm, 3.0 g) were subjected to eight treatments combining bleaching powders, hydrogen peroxide (10–40 vol), and permanent dyes ranging from blonde to dark brown. Combability was assessed using a Diastron® MTT 175. Color was evaluated by diffuse reflectance spectrophotometry, and protein loss was quantified as albumin equivalents.

Results and Conclusions: Bleaching produced the greatest color difference ($DE^* = 65.2 \pm 1.8$) and increased combing force by 46.6%, whereas treatments including coloration improved combability by up to 46.5%. All treatments caused substantial protein loss (5.1–16.6 mg albumin equivalents/g hair) compared with control strands (0.7 mg albumin equivalents/g hair). Bleaching compromised hair structural integrity, while dye formulations containing emulsifiers partially mitigated damage and improved combability; however, both processes led to considerable keratin degradation.

Keywords: hair; damage; coloring/bleaching; combability; protein loss.

Citation: Velasco MVR, Magnocavallo N, Câmara AC, Pinto CASO, Dario MF, Gama RM, Baby AR. Impact of oxidative hair coloring and bleaching on hair fiber properties. Brazilian Journal of Hair Health. 2026;3:bjhh44.

doi: <https://doi.org/10.62742/2965-7911.2026.3.bjhh44>

Received: April 9, 2026

Revised: April 15, 2026

Accepted: May 15, 2026

Published: May 19, 2026



Copyright: This content is licensed under the terms and conditions of the Creative Commons Attribution 4.0 International License (CC BY).

1. Introduction

More than 50% of Brazilian households purchase hair-coloring products, and approximately 98% of Brazilian women report using hair dye. The Brazilian hair-care market totaled US\$ 6.06 billion in 2023 and is expected to reach US\$ 7.70 billion by 2028, largely driven by permanent coloring products [1].

The hair fiber is a keratinized composite in which the cuticle and the cell membrane complex act as key diffusion and frictional barriers, while the cortex largely determines mechanical strength and contains melanin granules responsible for natural color. Oxidative treatments can disrupt surface integrity and internal structures, thereby affecting optical

properties (color change), frictional behavior (combability), and the release of proteinaceous material [2-6]. Natural pigmentation is mainly determined by the relative contributions of eumelanin and pheomelanin, which differ in hue and oxidative response. Natural hair color depends not only on the relative proportions of these pigments but also on melanin granule density/pigmentation, hair thickness, and age [7-9].

Bleaching and permanent dyeing rely on hydrogen peroxide under alkaline conditions to swell the cuticle and facilitate the diffusion of reactive species into the cortex. In bleaching systems, persulfates and high alkalinity intensify melanin oxidation and can also promote reactions with hair proteins, potentially compromising keratin integrity. When hydrogen peroxide is applied under strongly alkaline conditions (around pH 10)—typically in the presence of ammonium salts and metasilicates and combined with persulfates for prolonged processing (≈ 1 – 2 h)—the treatment becomes highly aggressive and may cause substantial fiber damage. Under such conditions, disulfide bonds may be degraded (reported up to $\sim 25\%$), which is commonly associated with reduced shine and softness, increased surface roughness, and higher combing forces [10-12].

Given the widespread use of hair dyes and bleaching products and the adverse effects associated with oxidative deterioration, it is essential to elucidate how different bleaching–coloring protocols affect both the physical properties and chemical integrity of hair fibers. Accordingly, this study examined the impact of distinct bleaching–dyeing combinations on hair-fiber properties by assessing color change (DE*), combability, and protein loss.

2. Methods

2.1. Preparation of hair strands

Histology reveals loose, 2.1. Preparation of hair strands

Straight, natural, virgin dark-brown hair strands (≥ 15 cm in length; 3.0 g) were first washed for 30 s at 22 °C with a 10% (w/w) sodium lauryl ether sulfate solution using a standardized procedure to remove natural sebum, potential cosmetic residues, and other impurities, and then rinsed under running water for 1 min [13]. The strands were subsequently dried at room temperature (22 °C).

2.2. Discoloration and coloring

Hair strands were treated with commercially available bleaching powders, hydrogen peroxide, and permanent dyes intended for professional and at-home use (Coferly® Cosmética Ltda; Appendices A and B). All procedures followed the manufacturers' instructions, using different mixing ratios to achieve four target shades ranging from blonde to dark brown. In total, eight treatments were performed in triplicate ($n = 3$ hair strands for treatment, identified as A, B and C), as summarized in Table 1. After treatment, strands were rinsed with water, dried at 22 °C, and evaluated for combability, color, and protein loss.

Table 1. Treatments applied to hair strands ($n = 3$ /each one). The qualitative composition of the commercial formulations is detailed in Appendices A and B.

Treatment Identification	Treatment
1	<u>Discoloration</u> : 20.0 g of bleaching powder and 40.0 g of hydrogen peroxide (40 vol.) and applied for 157 min.
2	<u>Discoloration</u> : 20.0 g of bleaching powder and 40.0 g of hydrogen peroxide (30 vol.) and applied for 45 min.
	<u>Coloring</u> : 10.0 g of dye (shade 10.1) and 15.0 g of hydrogen peroxide (40 vol.) and applied for 45 min.
3	<u>Discoloration</u> : 20.0 g of bleaching powder and 40.0 g of hydrogen peroxide (20 vol.) and applied for 45 min.
	<u>Coloring</u> : 10.0 g of dye (shade 8.1) and 15.0 g of hydrogen peroxide (30 vol.) and applied for 45 min.
4	<u>Discoloration</u> : 20.0 g of bleaching powder and 40.0 g of hydrogen peroxide (10 vol.) and applied for 45 min.
	<u>Coloring</u> : 10.0 g of dye (shade 7.73) and 15.0 g of hydrogen peroxide (20 vol.) and applied for 45 min.
5	<u>Coloring</u> : 10.0 g of dye (shade 12.0) and 15.0 g of hydrogen peroxide (40 vol.) and applied for 45 min.
6	<u>Coloring</u> : 10.0 g of dye (shade 10.1) and 15.0 g of hydrogen peroxide (30 vol.) and applied for 45 min.
7	<u>Coloring</u> : 10.0 g of dye (shade 8.1) and 15.0 g of hydrogen peroxide (20 vol.) and applied for 45 min.
8	<u>Coloring</u> : 10.0 g of dye (shade 7.73) and 15.0 g of hydrogen peroxide (10 vol.) and applied for 45 min.
Control	Virgin strands. No treatment.

2.3. Color evaluation

Hair fiber color was assessed by diffuse reflectance spectrophotometry, with measurements taken at the midpoint of each strand. The instrument reports color differences in the CIELAB system as three components: L^* (lightness; positive = lighter, negative = darker), a^* (red–green axis; positive = redder, negative = greener), and b^* (yellow–blue axis; positive = more yellow, negative = bluer). These components were combined to calculate the total color difference (DE^*), as shown in Equation 1 [15]. Differences among treatments were analyzed by one-way ANOVA followed by Tukey's post hoc test ($\alpha = 0.05$) using Minitab® 18.

$$\begin{aligned} DL^* &= L^*(\text{final}) - L^*(\text{initial}) \\ Da^* &= a^*(\text{final}) - a^*(\text{initial}) \\ Db^* &= b^*(\text{final}) - b^*(\text{initial}) \\ DE^* &= \{(DL^*)^2 + (Da^*)^2 + (Db^*)^2\}^{1/2} \end{aligned} \quad (1)$$

Legend: Color parameters in the CIELAB system. Legend: L^* = lightness; DL^* = lightness difference; a^* = red–green coordinate; Da^* = difference in the red–green coordinate; b^* = yellow–blue coordinate; Db^* = difference in the yellow–blue coordinate; DE^* = total color difference.

2.4. Combability test

Dry combability (total work) was measured using a Diastron® MTT 175 at room temperature (22.0 ± 2.0 °C). Prior to testing, each strand was manually combed 10 times with a plastic comb (five strokes in each side) to remove tangles and standardize the initial condition [14].

For each hair strand ($n = 3$ per treatment), total work (W) was recorded in repeated runs (≥ 5 readings) before (pre-treatment) and after (post-treatment) product application. Results were demonstrated as mean $\pm$ standard deviation, and the effect per treatment were evaluated by paired t-test ($\alpha = 0.05$) in Minitab® software.

Also, given the inherent variability of this biological material—since combability can be influenced by factors such as strand mass and other sample-specific characteristics—treatment effects were expressed as a within-strand ratio ($R = W_{\text{post}}/W_{\text{pre}}$). Values > 1 indicate increased combing work (worse combability), whereas values < 1 indicate reduced combing work (improved combability). For each treatment ($n = 3$ strands; A–C), the individual ratios (RA , RB , and RC) were reported, and the overall effect was summarized as the geometric mean ratio.

2.5. Protein Loss

Protein loss was evaluated using the Lowry method as modified by Peterson. For each strand, a 0.1 g sample was weighed, cut into ~ 3 –5 mm fragments, mixed with 15 mL of distilled water, and sonicated in an ultrasonic bath for 45 min to extract soluble proteins, peptides, and amino acids. A 2.0 mL aliquot of the extract was mixed with 2.0 mL of reagent A. After mixing and standing for 10 min, 1.0 mL of reagent B was added, and the mixture was allowed to react for 30 min. Absorbance was then measured at 750 nm using a Shimadzu® UV–Vis spectrophotometer (UV-1203), with water as the blank. Results were expressed as albumin equivalents based on an analytical curve constructed using bovine serum albumin (BSA) standard [16].

Reagent A was prepared by mixing 25.0 mL of a solution containing 0.1% (w/v) copper sulfate, 0.2% (w/v) sodium potassium tartrate, and 10% (w/v) sodium carbonate with 25.0 mL of 0.8 N sodium hydroxide and 25.0 mL of 10% (w/v) sodium dodecyl sulfate. The mixture was brought to a final volume of 100 mL with distilled water and homogenized.

Reagent B was prepared by diluting 8.4 mL of Folin–Ciocalteu phenol reagent to 50 mL with distilled water in a volumetric flask and homogenizing the solution.

The albumin analytical curve was prepared using BSA standard (96.1% purity). Aqueous solutions at different albumin concentrations were subjected to the same reaction described above, and the analytical curve was constructed by correlating albumin concentration with absorbance. Using Equation 2, the absorbance values obtained for the hair extracts were converted and reported as albumin equivalents, considering the mass of hair analyzed to express the results as mass of albumin equivalents per mass of hair.

$$y = 0.0043x + 0.1113; R^2 = 0.992 \quad (2)$$

Legend: R^2 = coefficient of determination; y = absorbance; x = protein-equivalents concentration ($\mu\text{g/mL}$).

Differences among treatments were assessed using the Kruskal–Wallis test ($\alpha = 0.05$), followed by Dunn's post hoc multiple-comparison test, using Minitab® software.

3. Results and Discussion

3.1. Color

Hair color is commonly described by (i) saturation, or “tone height,” typically graded on a 1–10 scale (1 = highest pigment concentration, black; 10 = lowest, white), and (ii) shade/nuance (“color reflections”), which refers to the primary and secondary tonal reflections present in the fiber [2, 3].

Artificial hair coloring occurs through the oxidation of primary intermediates (e.g., p-aminophenol and related derivatives) followed by sequential coupling reactions with various couplers. The final shade is influenced by the type, amount, and ratio of intermediates and couplers, as well as processing conditions such as temperature, pH, and reaction time, in addition to the diffusion rate of reaction products into the cortex [17]. Images of hair strands are shown in Figure 1, while total color variation (DE^*) values are presented in Figure 2.



Figure 1. Photographs of control (virgin) and hair strands after Treatments 1–8.

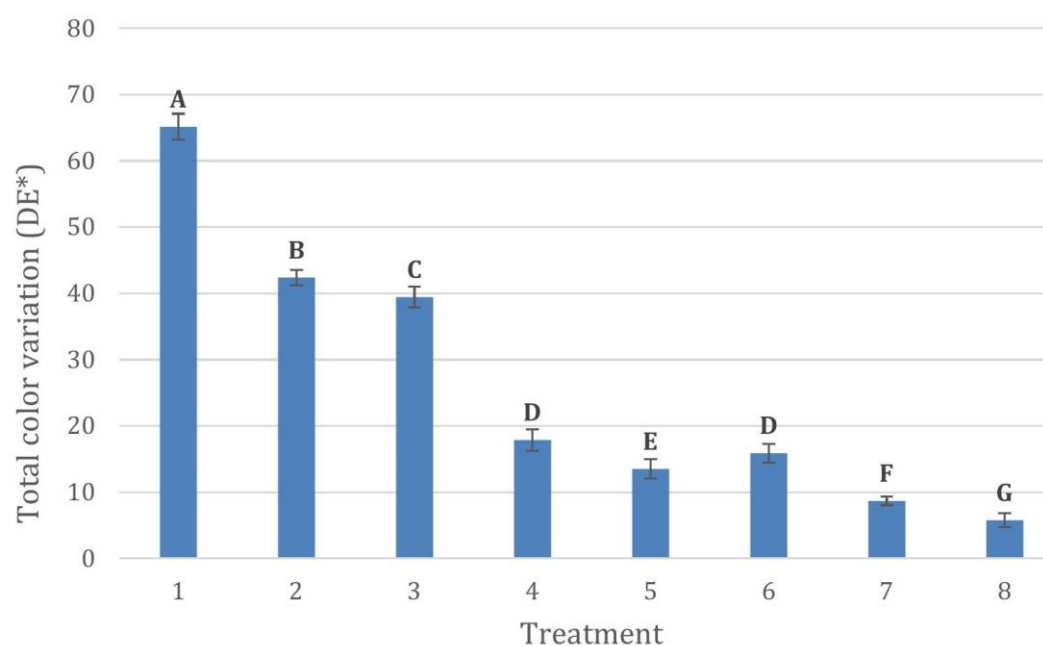


Figure 2: Total color variation (DE*; mean $\pm$ SD) between control and treated hair strands for each treatment. Means sharing the same letter are not significantly different according to Tukey's post hoc test (one-way ANOVA; $\alpha = 0.05$). Different letters indicate statistically significant differences ($p < 0.05$).

The combination of variables that determine the final color of bleached and dyed hair strands likely accounts for the differences observed in Figure 2, as expected. Notably, Treatment 1 produced the highest DE*, likely due to the higher amount of bleaching powder, the greater volume of hydrogen peroxide, and the longer application time. However, no statistically significant difference was detected ($\alpha > 0.05$) in total color variation between Treatments 4 and 6, despite the marked differences in the pigment compositions of the dyes used (shades 7.73 and 10.1, respectively). While all dyes contained p-phenylenediamine, p-aminophenol, m-aminophenol, and the coupler resorcinol, shade 12.0 additionally included 2,4-diaminophenoxyethanol HCl, whereas shade 7.73 contained 2-methyl-5-hydroxyethylaminophenol, 2-amino-3-hydroxypyridine, and the coupler 2-methylresorcinol (Appendix A). Therefore, combining bleaching with a darker dye in Treatment 4, and applying a lighter dye alone in Treatment 6, may have led to comparable DE* values.

3.2. Combability evaluation

Data in Figure 3 and Table 2 show that bleaching without subsequent coloring (Treatment 1) caused the most pronounced deterioration in combability, increasing combing work with a geometric mean ratio GM(R) = 1.466 ($\approx +46.6\%$ relative to the pre-treatment condition). A smaller increase was observed for Treatment 4 ($+12.1\%$). In contrast, all remaining treatments reduced combing work (GM(R) < 1), with the largest improvements observed for Treatments 7 and 8 ($\approx 45\text{--}47\%$ reduction) and Treatment 6 ($\approx 37\%$ reduction).

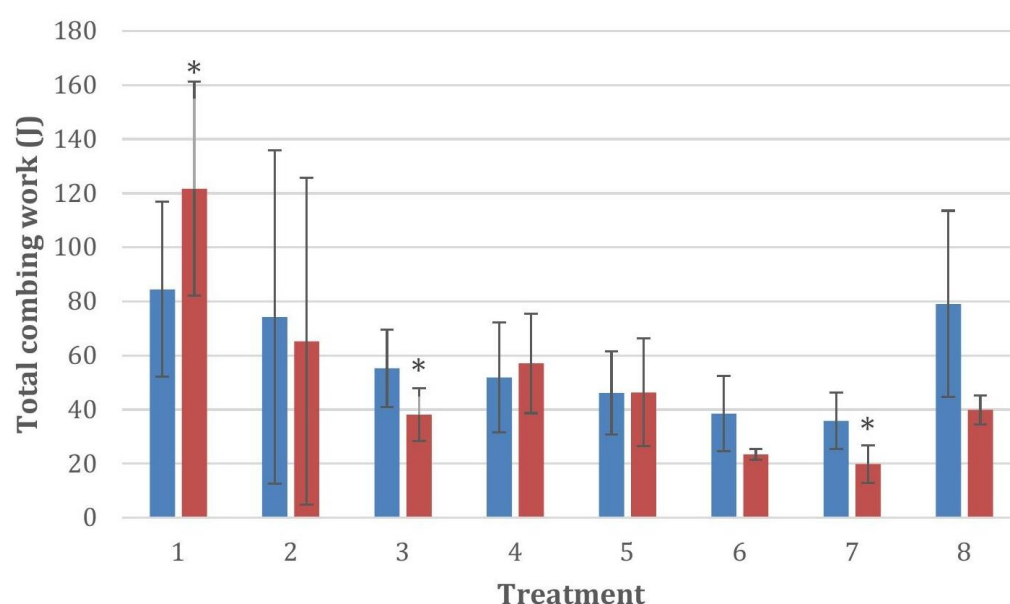


Figure 3: Total combing work (J) measured before and after each *Treatment* (mean $\pm$ SD; $n = 3$). * indicates a significant difference between pre- and post-treatment values (paired t-test, $\alpha = 0.05$).

Table 2. Change in hair-strand combing work (W/Joules) after each treatment.

Treatment	RA	RB	RC	GM (R)	% Combability variation
1	1,394	1,408	1,605	1,466	46,60%
2	0,597	0,938	1,115	0,855	-14,5%
3	0,721	0,663	0,686	0,69	-31,0%
4	1,06	1,06	1,255	1,121	12,10%
5	1,4	0,73	0,871	0,962	-3,8%
6	0,8	0,679	0,465	0,632	-36,8%
7	0,508	0,63	0,507	0,545	-45,5%
8	0,816	0,394	0,477	0,535	-46,5%

Values were expressed as the within-strand ratio $R = W_{\text{post}} / W_{\text{pre}}$. **RA**, **RB**, and **RC** correspond to the ratios **R** obtained for the three replicate strands (**A–C**). **GM (R)** is the geometric mean of RA–RC and summarizes the overall effect of each treatment. % Combing variation was calculated as $(GM(R) - 1) \times 100$; values > 0 indicate increased combing work (worse combability), whereas values < 0 indicate reduced combing work (improved combability).

The worsening in combability after bleaching may be related to the action of bleaching agents (e.g., persulfates and alkaline components) that enhance oxidant penetration and promote structural changes in the fiber, increasing friction and resistance during combing. In contrast, the improved combability observed in several coloring treatments (notably Treatments 3, 6, 7, and 8) may be associated with conditioning systems present in dye formulations, including fatty materials and emulsifiers (e.g., glyceryl stearate, lecithin, and oleic acid), which can reduce inter-fiber friction and improve strand alignment.

3.3. Protein Loss

All Treatments resulted in markedly higher protein loss than the minimal loss observed for control strands (Figure 4). Statistically significant differences were observed among treatment groups. Treatments involving coloring only (Treatments 7 and 8) showed the highest protein loss, followed by the combined bleaching-and-coloring treatments (Treatments 4, 5 and 6), which were statistically equivalent

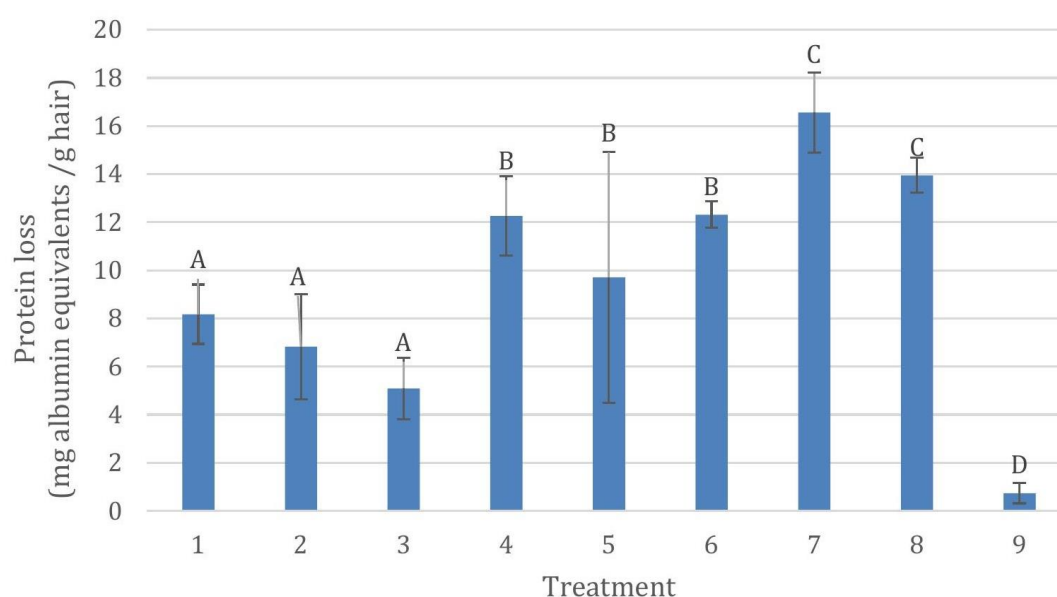


Figure 4: Means sharing the same letter are not significantly different according to Kruskal–Wallis test ($\alpha = 0.05$), followed by Dunn's post hoc multiple-comparison test.

An unexpected lack of a statistically significant difference was observed among Treatments 1, 2, and 3. This may be attributed to the use of 40 vol hydrogen peroxide with a light blonde shade (10.1) in Treatment 2 and 30 vol hydrogen peroxide with a light blonde shade (8.1) in Treatment 3, which may have produced oxidative effects comparable to bleaching alone (Treatment 1).

The combability of virgin strands and those subjected to Treatment 5 was quite similar ($GM(R) \approx 1$). However, Figure 4 shows that protein loss in Treatment 5 was significantly higher than in control. The statistical similarity among Treatments 4, 5, and 6 suggests comparable effects, consistent with the similar ΔE^* values observed in Figure 2 and supported by the same rationale.

More importantly, a pronounced difference was observed between the control and all treated strands, with the control exhibiting substantially lower albumin-equivalents protein loss. Overall, these findings indicate that bleaching, coloring, or their combination causes significant damage to virgin, straight, dark-brown hair strands.

4. Conclusions

As expected, the treatments produced a wide color gradient. In terms of the strands' apparent condition, Treatment 1 was the most damaging, causing a noticeable loss of shine and increased surface roughness, likely due to cuticle lifting and thinning of the protective layer. This effect was partially attenuated after rinsing and subsequent conditioning steps. In contrast, Treatments that included dyes tended to improve combability, which may be attributed to the presence of emulsifiers and other conditioning ingredients that enhance lubrication of the fiber. Protein-loss data, however, indicated deterioration of the keratin structure in both the cuticle and cortex after bleaching and/or coloring, which may compromise the hair's physical and chemical resistance. It should also be noted that the greater aggressiveness of Treatment 1 may be partly due to its substantially longer processing time compared with the other treatments.

Funding: None.

Institutional Review Board Statement and/or Informed Consent Statement: None.

Acknowledgments: None.

Conflicts of Interest: None.

References

1. Mordor Intelligence [Internet]. South America hair colorants market size & share analysis - growth trends & forecasts (2024-2029); 2024 [cited 2025 Dec 8]. Available from: <https://www.mordorintelligence.com/pt/industry-reports/south-america-hair-colorants-market>
2. Kirkland R. Designing the next generation of hair dyes: database reveals promising approaches to make safer, more sustainable hair color [Internet]. ScienceDaily; 2018 Jan 17 [cited 2025 Mar 10]. Available from: <https://www.sciencedaily.com/releases/2018/01/180117092345.htm>
3. Halal J. Hair structure and chemistry simplified. 5th ed. New York: Cengage Learning; 2009. Chapter 2, Hair color; p. 18-35.
4. Feughelman M. Morphology and properties of hair. In: Johnson D, editor. Hair and hair care. New York: Marcel Dekker; 1997. p. 1-12.
5. Robbins CR. Chemical and physical behavior of human hair. 5th ed. New York: Springer; 2012. Chapter 1, Morphology; p. 1-35; Chapter 6, Dyeing human hair; p. 233-262.
6. Nakamura A, Arimoto M, Takeuchi K, Fujii T. A multi-wavelength fluorescence method for evaluating effects of oxidative treatments on human hair. *J Cosmet Sci.* 2012;63(2):83-91. PMID: 22487448.
7. Brown KC. Hair coloring. In: Johnson D, editor. Hair and hair care. New York: Marcel Dekker; 1997. p. 191-8.
8. Wolfram LJ. Human hair: a unique physicochemical composite. *J Am Acad Dermatol.* 2003 Jun;48(6 Suppl):S106-14. DOI: 10.1067/mjd.2003.276. PMID: 12789162.
9. Grosvenor AJ, Deb-Choudhury S, Middlewood PG, Thomas A, Lee E, Vernon JA, Woods JL, Taylor C, Bell FI, Clerens S. The physical and chemical disruption of human hair after bleaching - studies by transmission electron microscopy and redox proteomics. *Int J Cosmet Sci.* 2018 Dec;40(6):536-548. DOI: 10.1111/ics.12495. PMID: 30229956.
10. Bedin V. Clareadores e colorantes para cabelos. *Cosmetics Online* [Internet]. 2012 May-Jun [cited 2025 Jan 30];24(3):32. Available from: https://www.cosmeticsonline.com.br/2011/ed.php?edicao=Maio/Junho_2012
11. França-Stefoni SA, Dario MF, Sá-Dias TC, Bedin V, de Almeida AJ, Baby AR, Velasco MV. Protein loss in human hair from combination straightening and coloring treatments. *J Cosmet Dermatol.* 2015 Sep;14(3):204-8. doi: 10.1111/jocd.12151. PMID: 25816718.
12. Harrison S, Sinclair R. Hair colouring, permanent styling and hair structure. *J Cosmet Dermatol.* 2003 Oct;2(3-4):180-5. doi: 10.1111/j.1473-2130.2004.00064.x. PMID: 17163924.
13. Dario MF, Pahl R, de Castro JR, de Lima FS, Kaneko TM, Pinto CA, Baby AR, Velasco MV. Efficacy of Punica granatum L. hydroalcoholic extract on properties of dyed hair exposed to UVA radiation. *J Photochem Photobiol B.* 2013 Mar;120:142-7. DOI: 10.1016/j.jphotobiol.2012.12.011. PMID: 23357210.
14. Ruetsch SB, Kamath YK. Effects of thermal treatments with a curling iron on hair fiber. *J Cosmet Sci.* 2004;55(1):13-27. PMID: 15037918.
15. Locke B, Jachowicz J. Fading of artificial hair colour and its prevention by photofilters. *J Cosmet Sci.* 2005;56(6):407-25. PMID: 16538313.
16. Gama RM. Avaliação do dano a haste capilar ocasionado por tintura oxidativa aditivada ou não de substâncias condicionadoras [dissertation on the Internet]. São Paulo: Universidade de São Paulo, Faculdade de Ciências Farmacêuticas; 2010 [cited 2025 Dec 3]. Available from: https://teses.usp.br/teses/disponiveis/9/9139/tde-19012011-145405/publico/Mestrado_Robson_Miranda_daGama.pdf
17. Oliveira RAG, Zanoni TB, Bessegato GG, Oliveira DP, Umbuzeiro GA, Zanoni MVB. A química e toxicidade dos corantes de cabelo. *Quim Nova.* 2014;37(6):1037-46. DOI: 10.5935/0100-4042.20140159.