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Fragrance Retention in Virgin and Bleached Caucasian Hair

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Synopsis

Human hair, when bleached, undergoes oxidation of melamine granules and its structural proteins. This work aims to compare fragrance retention in both virgin and bleached hair, taking into consideration the interactions between fragrance compounds and hair before and after chemical modification. The bleaching process of straight dark brown Caucasian hair was carried out using a 4.5% wt. hydrogen peroxide solution at pH 9.5. Fragrance raw materials were incorporated in a shampoo formulation and applied on hair by washing, followed by rinsing. Hair was then let to dry under controlled conditions of temperature and humidity and the volatiles were collected by solid-phase microextraction and quantified by Gas Chromatography Mass Spectrometry (GC-MS). The more bleached the hair, the higher is the amount of sorbed substances during shampoo washing because of a higher number of holes in the hair structure, which increases its sorption capacity. Besides that, the impairments caused by oxidative reaction of hair surfaces are responsible for the faster evaporation of fragrant compounds and this behavior was compared with the loss of moisture of untreated and bleached hair.

INTRODUCTION

Fragrances play a decisive role in cosmetics and are often the main characteristic that distinguishes a product from their competitors (1). Hair perfume perception is a very important attribute when creating shampoo fragrances. The consumers expect their hair to be perceived as clean and fresh and this impression is closely linked to the longevity of the fragrance on the hair (2). Moreover the cost of fragrance in the consumer product is not negligible, so each ingredient in the perfume has to be efficient, be cost-effective, and contribute to the overall consumer satisfaction (1). Therefore, this industry constantly strives for fragrance improvement and to make them cost-effective, longer lasting, and relevant to the values of the brand (2).

The primary purpose in bleaching human hair is to lighten the hair by partial or complete oxidative degradation of the natural color pigment. For this, severe reaction conditions are required for destruction of the chromophoric groups of hair melanin. The

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bleaching reagents most often used are aqueous solutions of hydrogen peroxide adjusted to pH values between 9 and 11. In bleaching compositions, persulfate salts are often added as accelerators (3,4).

A published work on hair fragrance persistence from Blakeway and Seu-Salerno (5) observed the high affinity of amyl salicylate to virgin hair remaining for over 24 h after deposition. According to the authors, the fragrance transference to hair occurs as a partition mechanism of organic molecules from water phase to hair keratin, a mainly nonpolar support, and the fragrance persistence on hair depends on the molecule polarity.

The present article contributes to the comprehension of the capacity of fragrance deposition on bleached hair by washing in comparison with the virgin hair and compares the fragrance liberation after drying to the desorption of moisture on hair before and after bleaching.

MATERIALS AND METHODS

SAMPLES

Caucasian virgin dark brown hair, 25-cm long, was purchased from International Hair Importers (New York, NY). The hair tresses were washed with a 2.0% w/w sodium lauryl sulfate aqueous solution and left to dry at room temperature for at least 24 h.

BLEACHING TREATMENT

Hair tresses underwent bleaching for 10, 20, and 40 min with a hydrogen peroxide solution (4.5% v/v; pH = 9.5; at 55°C), consisting of 15% (w/w) of ammonium persulfate, and 70% of distilled water. The pH was reached using a concentrated sodium hydroxide solution (6). After the bleaching treatment, the samples were rinsed with tap water for 2 min and left to dry at room temperature.

DIFFERENTIAL SCANNING CALORIMETRY (DSC) ANALYSIS

Amounts of 5–7 mg of hair, approximately 0.5-cm long, were cut and placed in pressure-resistant stainless steel capsules. Fifty microliters of deionized water was introduced. The instrument used was DSC 4000 (PerkinElmer, Waltham, MA). The temperature range was from 70°C to 180°C, with a heating rate of 5°C min^{-1}. The samples of hair were stored overnight at an environment with 20°C ± 2°C and 50% ± 5% of relative humidity to ensure constant water content. The capsules were sealed and stored for 24 h before test run.

FLUORESCENCE MICROSCOPY

Small portions of hair fibers, about 3 cm long, collected from the swatches were embedded in acrylic resin. Sectional 10-µm cuts were processed with the use of a microtome. The cuts were exposed to rhodamine B (Sigma-Aldrich, St. Louis, MO) solution (10 µg mL^{-1})

for 45 min. They were rinsed with deionized water and left to dry at 45°C for 15 min. The analyses were performed in the microscope Olympus BX53 with a USH-103OL mercury burner. Fluorescence intensity was extracted from the images using Image Pro Premier software (Media Cybernetics, Rockville, MD).

FRAGRANCE DEPOSITION

Fragrance raw material compounds ethyl 2-methylbutanoate, linalool, benzyl acetate, and Mintonat were introduced in a shampoo formula at 0.1% (w/w) of each compound. Samples of 0.6 g of virgin and bleached hair were washed individually with 120 µl of a solution of the shampoo, presented in Table I, at 50%. The wet hair tresses were gently rubbed for 1 min from the roots to the tips and rinsed with tap water for 30 s and left to dry in controlled environment (60% RU; at 22°C) for 2, 4, and 6 h.

EXTRACTION OF VOLATILES BY SOLID-PHASE MICROEXTRACTION (SPME)

Headspace sampling of the hair fragrance compounds was achieved in a sealed flat bottom vial, immediately after the tresses insertion, and with triple gray SPME fiber (CAR/PDMS/DVB; 50/30 µm diameter; 2 cm length) from Supelco (Sigma-Aldrich, Darmstadt, Germany). For the desorption of the volatile compounds, the SPME fiber was directly inserted into the chromatograph with an auto sampler.

GC-MASS ANALYSIS

GC-MS analyses were performed on an Agilent 6890 chromatograph coupled to an Agilent HP597 mass spectrometer (Agilent Technologies, Santa Clara, CA) with an electron ionization source operating at 70 eV and equipped with a fused silica capillary column ZB-1 (60 m × 0.25 mm × 0.25 µm) with 100% polydimethylsiloxane. Helium was used as the carrier gas (1.5 mL min^{-1}), and the injector temperature was set to 250°C and the detector temperature to 260°C.

Table I
Shampoo Formulation Used to Embody Fragrance at 0.1% (w/w) of Each Compound

Component	%
Water	q.s. 100.00
Dissodium ethylenediamine tetraacetic acid	0.10
Sodium lauryl sulfate	37.00
Caprylyl glycol, 1,2-hexanediol, and 1-phenylethanol	0.50
Cocamidopropyl betaine	5.00
Trideceth-9, polyethylene glycol-5, isononanoate and water	1.00
Glycol distearate, sodium lauryl sulfate	3.00
PEG-150, distearate	1.00
Cocamide diethanolamine	1.00
Sodium chlorine	0.15
Fragrance	0.40

DESORPTION OF WATER ON HAIR

Triplicates of 2 g of virgin and bleached hair were placed in Eppendorf tubes (1.5 mL) and left to hydrate in a desiccator flask with distilled water in the bottom (RU 98–99%; at 21°C) for 20 d. The hydrated hair mass was obtained in an analytical scale and placed in a desiccator with P_2O_5 (RU 0%; at 21°C) (7) and the mass variation was acquired over the time.

RESULTS AND DISCUSSION

DSC ANALYSIS

The DSC peak is associated with the temperature of denaturation of the hair keratinous materials. The area under the curve of the peak corresponds to the enthalpy of degradation of the helical structures, represented by delta H (8). It is correlated with quantity and quality of the α-helical materials of the intermediate filaments (IFs) of the hair (8–10).

The obtained values for enthalpy variation and peak temperature from DSC analysis are presented in Table II. The numbers suffered important reductions because of the bleaching process in both parameters. Both variations have been associated by Wortmann et al. (8) to the loss of helical fibrous IFs and intermediate filament-associated proteins (IFAPs) caused by the bleaching process being the first one even more affected, which explains the higher discrepancy of figures in the comparison of the virgin and the bleached hair.

FLUORESCENCE MICROSCOPY ANALYSIS

The level of damage of the hair can influence the fluorescence intensity detected as rhodamine B presents higher affinity to negative sites of the damaged hair. The bleached hair, therefore, is expected to present higher intensities than the virgin hair as the presence of sulfonic acid generated from the break of the disulfide bonds is much more expressive (3). The fluorescence intensity was calculated by the Image Analysis software in each captured image and then compared between the types of hair.

In the fluorescence microscopy analysis, the virgin hair presented values of fluorescence intensity lower than three luminance as bleached hair presented figures five times higher (Table II). These results are in accordance with the ones found by Pötsch and Moeller (11). Low fluorescence intensity was evidenced in the virgin hair in investigations with rhodamine B as well, whereas damaged hair presented higher degrees of intensity.

Table II
Results Obtained for DSC Analysis and Fluorescence Microscopy for the Virgin and the Bleached Hair

Type of hair	ΔH (J/g)	Peak T (°C)	Fluorescence intensity (Lum)
Virgin	12.4777 ± 0.3579	147.6347 ± 0.5764	1,6415 ± 1,2177
Bleached	9.4066 ± 0.3407	141.9717 ± 0.5691	16,3907 ± 1,1158

Mean ± confidence interval.

FRAGRANCE RETENTION

Hair fragrance release rate was evaluated by normalizing to 100% the peak chromatographic area of the most abundant fragrance compound. Consequently, the values obtained for the amount of fragrance transferred to hair after washing and retained after different periods of drying are expressed in percentage.

The fragrance compounds bar graph (Figure 1) shows that more compounds are trapped by bleached than by virgin hair; thus, more fragrance material is sorbed on hair by washing with shampoo. Compared with virgin hair, bleaching for 40 min showed at least 40.0% more abundance for ethyl 2-methylbutanoate, 16.9% for linalool, 20.2% for benzyl acetate, and 24.6% for Mintonat. Bleaching for 10 and 20 min analyzed immediately after washing showed amounts of fragrance materials with intermediate values, between virgin hair and hair bleached for 40 min.

Oxidative bleaching under alkaline condition causes the oxidation of melanin (in the cortex) and other hair components, generating a structural hair modification. Richena et al. (6) proved by transmission electron microscopy that gray Caucasian hair bleached with the same reactants used in this work presented hole formation in the cortex, where melanin granules used to be located. The same structural transformation can be seen in the scanning electron micrography in the work of Ruetsch et al. (12) using the hair treated with 6% hydrogen peroxide for 4 h. This hair internal morphologic transformation leads to a surface increase and consequently more fragrance compounds retention on hair washing. Therefore, the longer the hair bleaching process, the greater is the hair structural damage and, consequently, the higher is the surface of interaction between fragrance compounds and hair.

Another consequence of hair bleaching is the oxidation of cystine disulfide bond in the cortical matrix, the A-layer and the exocuticle inside cuticle cells (3,13). The chemical transformation by bleaching converts the virgin hair fiber, especially the surface from a hydrophobic material with little surface charge to a more hydrophilic, more polar, and more negatively charged surface (3). This change in the polarity of the protein groups of hair increases its capacity to interact and attach chemical compounds as water, fragrance compounds, and rhodamine B as demonstrated with the results previously mentioned. Robbins and Kelly (14) observed large amounts of cysteic acid in hydrolyzated bleached hair when analyzing amino acids in cosmetically modified hair and the quantities increased with increased bleaching. Therefore, longer bleaching processes produce larger amounts of cysteic acid and the higher is the polarity of the fiber. This explains why

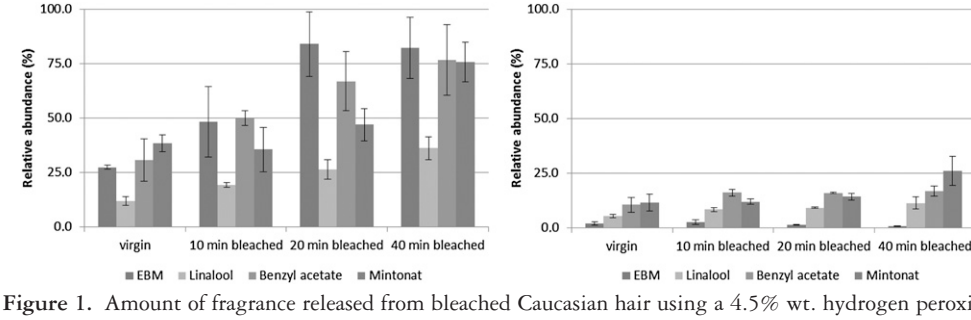

Figure 1. Amount of fragrance released from bleached Caucasian hair using a 4.5% wt. hydrogen peroxide solution in pH 9.5 at 55°C as a function of time of bleaching: 2-methylbutanoate (blue filled square), linalool (orange filled square), benzyl acetate (green filled square), and Mintonat (violet filled square).

bleached hair for 40 min showed higher capacity of fragrance deposition on hair by washing with shampoo.

The hair damages caused by the chemical bleaching must also be taken into consideration with regard to the transference of fragrance compounds to the hair interior. Chemical bleaches weakens cell membrane complex by oxidizing thioester groups between cuticle cells, leading to the breakdown of the membrane complex, and also dissolving proteins in endocuticle and in the cortex. Moreover, chemical bleaching promotes the cuticle scale lifting, cracks, and split formation (3). As a consequence, large molecules and other substances can penetrate easier from the inside to the outside of the fibers through the intercellular diffusion path in the low cross-link density regions.

The results obtained for the hair analyzed after 2 h of drying showed that analogous to the results for wet hair, the amount of fragrance of bleached hair for 40 min was higher than that of virgin hair: at least 8.8% more for ethyl 2-methylbutanoate, 1.2% for linalool, 10.3% for benzyl acetate, and 17.1% for Mintonat. Despite this, a significant reduction in this difference of amount of fragrance between virgin and bleached hair can be observed. It proves that the fragrance liberation from bleached hair is faster than that from virgin hair besides its greater capacity of embody fragrances right after washing with shampoo, as it was observed for the results of wet hair. The period of 4 h of drying follows the same tendency of reduction and this fact is very evident for the period of 6 h of drying: at least 1.8% more for linalool, 1.2% for benzyl acetate, and 5.0% for Mintonat. In both 4 and 6 h of drying, no difference in the amount of ethyl 2-methylbutanoate in virgin hair and hair bleached for 40 min can be observed.

In addition, the faster fragrance decrease rates of the bleached compared with virgin hair during drying process are assigned to hair surface damages caused by the oxidation reactions of the structural protein of hair. The damage of the barriers that prevent intercellular diffusion and penetration of substances into the fiber increases the transference of fragrance compounds from the inside to the outside of the bleached hair when compared with virgin hair.

DESORPTION OF WATER

The higher capacity of sorption of substances on bleached hair compared with virgin hair was demonstrated by water sorption tests. The results for water desorption shows that bleached hair for 40 min sorbed at least 28.1 $mg_{water}.g_{hair}^{-1}$ more water than virgin hair after 20 d at 98–99% RU (time: 0 h on abscise coordinate of the graphic in Figure 2). When submitted at 0% RU, both virgin and bleached hair show a decrease in water, and bleached hair showed a higher content of water until at least 19 h of exposition to the drying agent P_2O_5 at 21°C. After this period, no difference in water content between virgin and bleached hair can be observed.

These results correlate with fragrance uptake: bleached hair can sorb a higher amount of substances (fragrances compounds and water). The moisture decrease rate for bleached hair is higher in comparison with virgin hair and this can also be attributed to the fiber surface damage caused by bleaching that increases the ability of hair to retain and release water. This explanation is in accordance with the work of Pauling (15), which verified that the polar side chains of amino acid residues of proteins provide much of the attraction for the adsorbed water molecules.

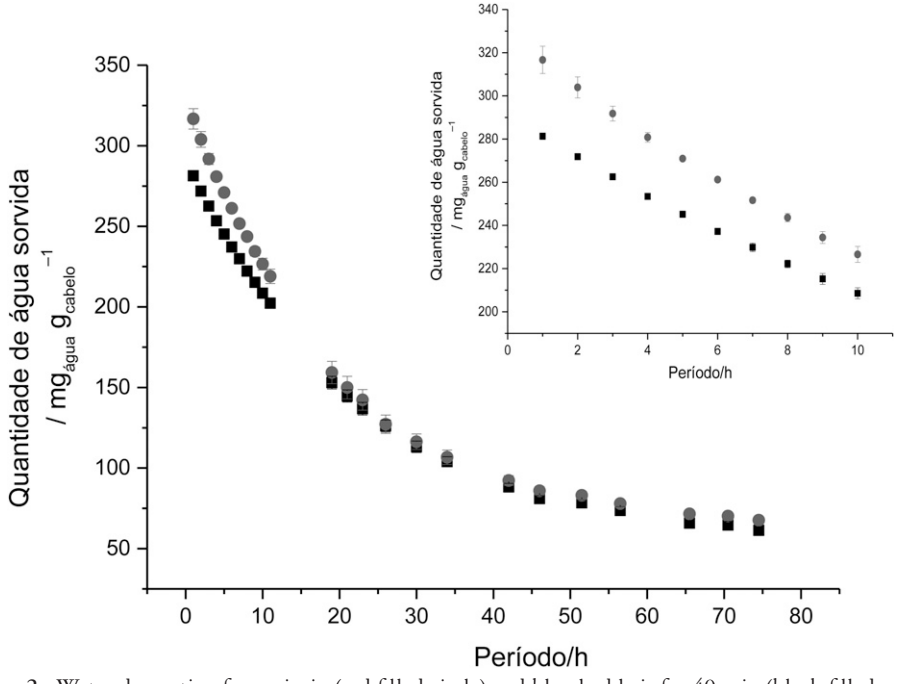

Figure 2. Water desorption from virgin (red filled circle) and bleached hair for 40 min (black filled square) as a function of period of drying (RU 0%, at 21°C).

CONCLUSIONS

The results presented herein indicate that bleaching process provokes deep damage in hair structures, altering its properties of interaction with materials. Fragrance deposition by washing with shampoo is greater on bleached hair in than on virgin hair (at least 40.0% more abundance for ethyl 2-methylbutanoate, 16.9% for linalool, 20.2% for benzyl acetate, and 24.6% for Mintonat). This behavior was attributed to the increase of interaction surface after hole formation in the cortex with dissolution of melanin granules, to the damage in hair structure that increased both intercellular and transcellular diffusion, and to the increase of affinity between hair fiber and fragrance compounds with the increase of the polar character of hair after the oxidation of cysteine groups. This last observation is in accordance with the results obtained for the deposition of rhodamine B on hair. Nevertheless, the amount of fragrance materials decreases faster in bleached hair, and this behavior also attributed to the diffusion increase caused by damages in hair structure after chemical bleaching. The results observed for the interaction of fragrance with hair are in accordance with the results obtained for the moisture desorption that was at least 28.1 $mg_{water} \cdot g_{hair}^{-1}$ greater on bleached hair but showed no difference after 19 h at RU 0% in comparison with virgin hair.

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