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Hair coloring by melanin precursors: A novel system for imparting durable yet reversible color effects

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Synopsis

This paper describes an approach to hair coloration which is different from any currently available system. It is based on formation within the hair of deposits of a melanin pigment, very similar in character to that responsible for the color of natural hair. The hair coloring process consists of two steps: a shampoo during which a dye initiator (Cu II) is introduced into the hair structure and, following a rinse, treatment with an aqueous solution of a colorless melanin precursor, 5,6-dihydroxindole. The coloring process is usually complete within five to ten minutes.

The melanin deposits are formed in the peripheral regions of hair and thus are readily available for color modification or even total removal.

INTRODUCTION

While the texture and geometry lie at the roots of hair styling, there is little doubt that the essence of hair's beauty manifests itself in the color. This has been well recognized as much in the distant past as it is now. It is truly remarkable how, using the melanin pigment (a substance without an identifiable chromophore) as its primary colorant, nature via the unique combination of chemistry and physics has been able to generate hundreds if not thousands of shades ranging from the Scandinavian blondes through Scottish red-heads to the intense black hair of Africans and Orientals. Still, the need for color enhancement or, indeed, its change, exists and is the driving force of the hair coloring market as reflected by the variety of products available to the consumer.

Setting aside the diversity of claims and application techniques, hair coloring products fall into two general categories: those which are based on materials that are inherently colored and those that use colorless precursors which only develop their hair coloring characteristics on interaction with an oxidant. Dyes of the first category are used in temporary (or shampoo removable) coloring products and semi-permanent formulations (color stable to several shampooings). The second category is the mainstay of the so-called permanent or oxidative hair color. Their importance lies not only in color durability and versatility in producing color change, but also in that the natural hair color can be modified, almost at will, to any desirable hue or shade, whether lighter or darker than the original. This is accomplished through a combination of bleaching of the natural melanin pigment and simultaneous color formation. Such shade manipulation is clearly not available in the temporary and semi-permanent products, the function of which is primarily the building up of color intensity. However, while the semipermanent colorants lack the versatility of oxidative dyes as well as their durability, they are generally recognized as being gentler to the hair since no peroxide is required in the process.

In each hair coloring category a sizeable number of dyes (or precursors) is required to attain a viable palette of shades. These dyes differ not only in their chromophoric characteristics but also in their affinity to hair, water solubility, and overall photostability. In color impartation, a delicate balance of constituent dyes is essential to obtain uniform and desirable results. However, subsequent exposure of the dyed hair to shampooing, sunlight, perspiration, and simple wear and tear, often highlights the differences in dye properties which can result in unpredictable color changes.

As the natural color of hair is not based on a mixture of dyes, hair care regimens have virtually no effect on hair color, and the latter shows only on-shade fading on prolonged exposure to light (1). It is primarily this wear characteristic of the natural hair pigment that prompted us to explore the feasibility of using melanin as a hair colorant for use under cosmetic conditions.

MATERIALS AND METHODS

MELANIN PRECURSORS

5,6-Dihydroxyindole, DHI, was prepared by reductive cyclization of the dinitrostyrene as described elsewhere (2,3). The purity of the compound was >99% as determined by tlc analysis and Proton NMR.

METAL SALTS

Reagent quality metal salts were obtained from Fluka Chemical, Switzerland. Unless otherwise specified, the metal salt compositions were prepared by dissolution of the appropriate salt (0.08 M) in a commercial shampoo. If required, the pH of the shampoo was adjusted with either hydrochloric acid to pH 5 or monoethanolamine to pH 9.

HAIR TRESSES

These were prepared from intact and bleached human hair purchased from DeMeo Brothers, New York. Approximately two grams of hair were used for each tress. The hair was shampooed prior to use, rinsed well with water, and air dried.

HAIR DYEING

Hair coloring involving formation of melanin from DHI was carried out in two consecutive steps. First the hair tresses were shampooed with a metal salt-containing shampoo (lathering time was five minutes), rinsed with tap water, and then treated for a specified time with an aqueous composition containing DHI. Both the shampoo and the dyeing steps were carried out at a 1:1 liquor ratio. After coloring, the dyed tresses were thoroughly rinsed under running tap water and air dried. Commercial hair coloring products were used according to instructions supplied in the kits.

COLOR EVALUATION OF HAIR

Color values and spectral reflectance data were obtained using the LabScan LS-5100 spectrocolorimeter (Hunter Associates Laboratory, Inc., Reston, VA). Two measurements (one of each side) were obtained per tress and averaged. The tress configuration and the design of the hair tress holder allowed for the measurement of the same area of the tress before and after treatment.

Results are expressed in terms of the Hunter L, a, and b color scale where L is a measure of lightness and varies from 100 for perfect white to 0 for black. The chromicity dimensions a and b are color designations, with 'a' being a measure of redness when the value is positive and greenness when negative, while 'b' is a measure of yellowness when positive and blue when negative.

In some tables the total color change (TCC) is used. TCC is calculated from the L, a, and b values according to the equation,

$$\mathrm{TCC} = \sqrt{(L_i - L_f)^2 + (a_i - a_f)^2 + (b_i - b_f)^2}$$

where subscript i denotes the initial value and subscript f the final value of the color parameters.

LIGHTFASTNESS

This was evaluated by exposing the tresses in an Atlas Fade-O-Meter (Atlas Electric Device Co., Chicago, IL) equipped with a sunshine carbon arc lamp. The lamp is centrally located in a 19½-inch specimen rack which rotates at approximately three revolutions per minute during the exposure period. An area of 2 cm × 3.5 cm of each tress was exposed for 10 hours at approximately 65% relative humidity in all light-fading studies. At the conclusion of exposure, the hair tresses were evaluated for color changes.

WATER BLEEDING

The dyed tresses were immersed in a stirred bath containing 2% of a commercial shampoo (active ingredient, sodium lauryl sarcossinate, 6%) at 50°C for 15 minutes and then thoroughly rinsed under running tap water and air dried. Color measurements were taken before and after the water bleeding test.

SHAMPOO STABILITY

The colored tresses were hand shampooed with undiluted commercial shampoo (active ingredient, TEA lauryl ether sulfate, 5%) for 30 seconds, rinsed, and towel dried. This cycle was repeated four more times. LabScan readings were taken before and after the shampoo treatment.

ACID PERSPIRATION

A synthetic perspiration composition was prepared from sodium chloride, lactic acid, histidine hydrochloride, and sodium dihydrogen phosphate, adjusted to pH 3.5 with

HCl. The colored tresses were immersed for five hours at 50°C in this solution. The change in color depth was determined on the LabScan.

HAIR WAVING

Commercially available products were used and process instructions followed, with the exception that the hair was treated in straight, rather than in curled, configuration.

RESULTS AND DISCUSSION

In vivo the formation of melanin takes place within the melanocytes and it is their secretory products (melanosomes) that are responsible for the color of skin and hair. Melanosomes appear as discrete black granules which are injected into the corticular cells located at the base of the hair follicle where the keratinocytes differentiate. From there the melanosomes are carried upward with the flux of the keratin cells. Note in Figure 1 the distribution of the melanosomes throughout the fibrils of the hair cortex. The color of the emerging hair is a complex function of the chemical characteristics and the quantity of the embedded pigment as well as of its particle size. As long as the hair is in the growing phase, the melanin supply continues unimpeded. In the resting phase (telogen) the melanocytes are dormant and resume their activity once the growth (anagen) phase is reinitiated. The failure of the pigment cell apparatus to produce melanin at this stage manifests itself in hair greying.

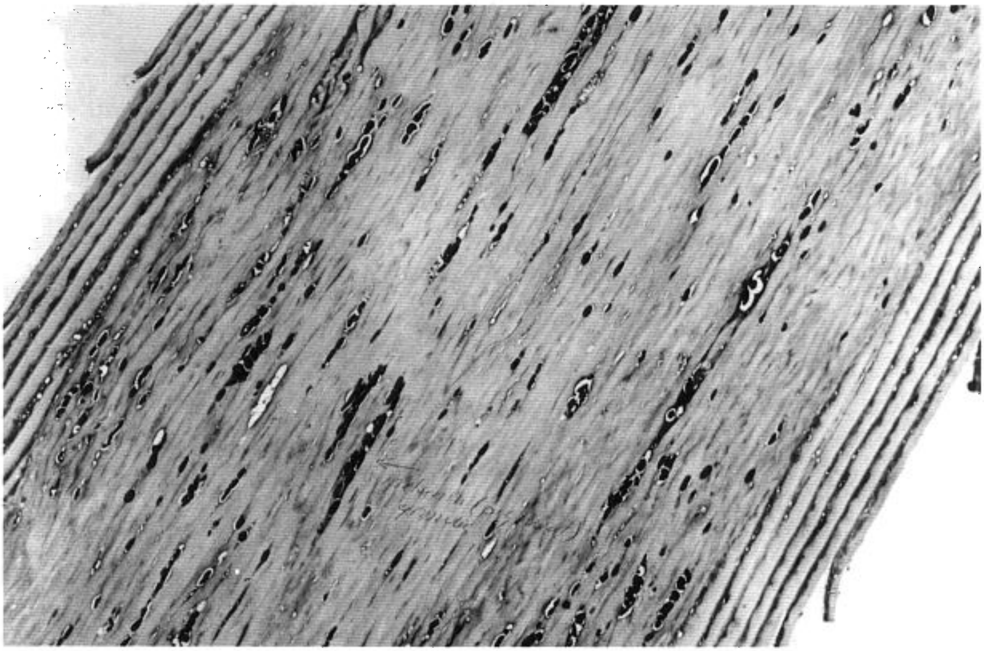

Figure 1. Longitudinal cross section of brown hair showing melanosomes (black granules, arrow) in the keratin matrix of the hair cortex.

The pioneering work of Raper and Mason (4) deciphered the basic pathway of melanogenesis (Figure 2), the early stages of which involve conversion of tyrosine (A) to L-DOPA (B), which is then oxidized to DOPAquinone. Both processes are under catalytic control by the same copper-containing enzyme, tyrosinase. The subsequent steps of the melanogenesis, that is the formation of the putative melanin precursor 5,6-dihydroxyindole (DHI) and its conversion to melanin, are thought to occur spontaneously under oxidative conditions without requiring specific enzymatic assistance. In view of the latter, it is thus not surprising that synthetic melanins prepared directly from precursors such as DOPA or DHI exhibit similar patterns of chemical reactivity and are virtually spectroscopically indistinguishable from their natural analogues (5).

Adaptation of the indigenous melanogenesis route into a process that can be exploited for hair coloring under cosmetically acceptable conditions requires significant shortcuts in the pathway. This can be attained by using a melanin precursor, such as DHI, as the principal active ingredient. Although conventional oxidants may be used to bring about the conversion of DHI to melanin, we find that metal salts are significantly more effective. This approach is of particular importance, as many salts have great affinity for keratin (6) and thus a system of hair-anchored catalysis can be readily envisaged. In the course of preliminary experiments, a basic procedure for development of melanin color in the hair was established. It involves treatment of hair with an aqueous solution of a metal salt, a rinse, and subsequent exposure of the hair to aqueous DHI. Under these conditions, melanogenesis within the hair is complete in a few minutes.

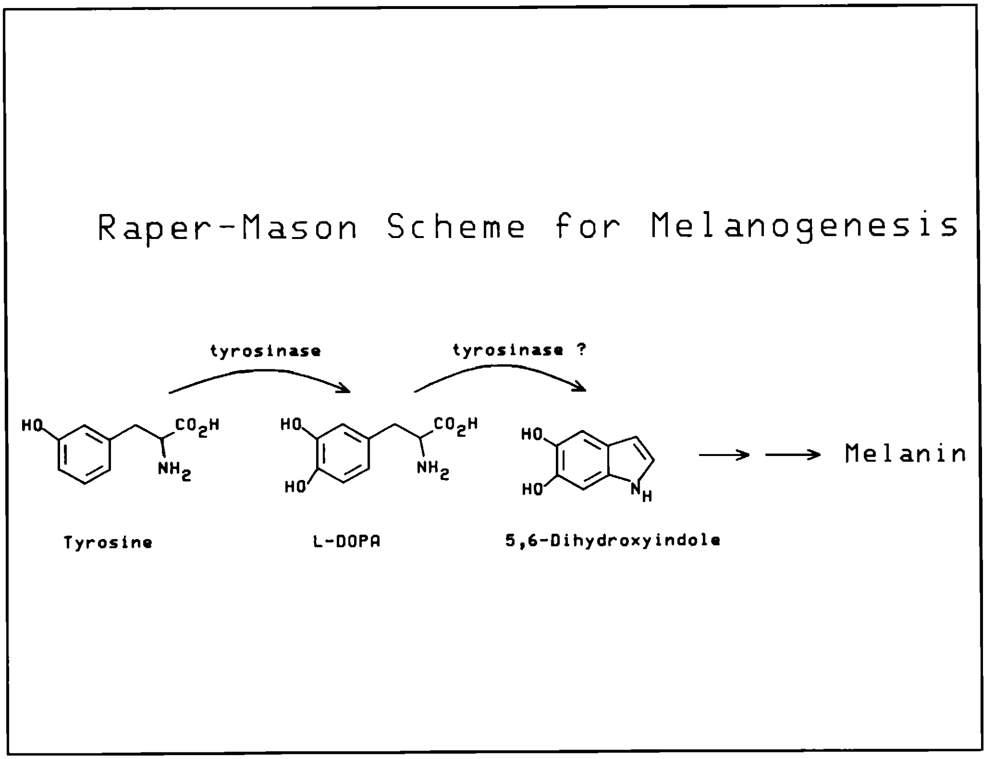

Figure 2. The Raper-Mason scheme for biochemical formation of melanin from tyrosine.

PARAMETERS OF COLOR FORMATION IN HAIR

Metal catalysis. The rate of color development with DHI as the dye precursor depends on the nature of the metal ion used in the pretreatment: Cu(II), Fe(II), and Ni(II) are effective in promoting fast melanogenesis, whereas Mg(II), Ti(IV), and Al(III) give rise to much slower coloration. Usually when the melanin formation is fast, the color developed in hair is dark grey to black. Slower melanogenesis leads to a color shift towards brown. Accordingly, grey hair pretreated with Cu(II) is dyed black, whereas the color is medium brown with a Ti(IV) pretreatment. Thus, by the use of the appropriate metal ion, some color modulation through the brown shades can be attained. We do not have satisfactory explanation at this time why and how the different metal ions lead to these color effects. Conversion of DHI to eumelanin by Cu(II) has been well documented (7,8) and this metal ion has been our primary choice in studying the melanization of DHI on hair. Also, salts of copper bind to keratin very readily (6) so that even a brief (5 min) treatment of hair with dilute solutions of copper salts (0.04 M at pH 9.0) deposits sufficient quantity of Cu(II) to dye the hair uniformly black after exposure to DHI. Under such conditions the quantity of Cu(II) taken up by the fiber is close to 1000 ppm. Longer treatment times, although leading to higher uptake of Cu(II), have virtually no effect on the attained color. Table I illustrates the effect of pretreatment of hair with different concentrations of cupric salts on subsequent color development with DHI. The melanization of DHI is significant even for the lowest concentration of Cu(II) as judged by a large decrease in the L value. As the eumelanin pigment dispersion is relatively achromatic, its deposition should also affect the a and b values. However, the former is close to 0 for the untreated grey hair, and it remains virtually unchanged. The latter, however, does decrease substantially and even at intermediate concentrations of Cu(II) reaches the level typical for black hair.

Color development. As a result of fiber-anchored catalysis, the conversion of DHI to melanin takes place preferentially in the hair. Unlike the auto-oxidation of DHI, which is favored by alkaline conditions, we find that the metal-induced melaninization process is less pH-sensitive (Table II). The DHI concentration required for effective coloring is in the range of 0.02 to 0.13 M. A study of the color intensity on bleached, blended grey, and brown hair as a function of the DHI concentration revealed that above 0.07 molar the L value for the dyed tresses reached a relatively constant value (Figure 3).

The color develops quickly and there is little change in both the shade and intensity after ten minutes of treatment with DHI. However, one has to bear in mind the L

Table I

Color Parameters (L, a, and b) for Cu(II)-Catalyzed Melanization of DHI on Blended Grey Hair

[Cu(II)]M	L	a	b
0.000	33.4	−0.35	5.33
0.001	18.5	0.15	2.12
0.010	15.4	0.21	1.05
0.025	15.5	0.13	0.55
0.040	16.0	0.15	0.85
0.050	14.8	−0.10	0.95
0.100	14.7	−0.15	0.72

[DHI] = 0.07 M; pH = 9.5; color development time = 15 minutes.

Table II
Color Parameter (L) for Cu(II)-Catalyzed Melanization of DHI on Blended Grey Hair at Various pH Values

pH	L
2.0	15.9
4.0	13.9
9.0	13.2
11.0	13.4

[Cu(II)] = 0.04 M, [DHI] = 0.07 M, color development time 15 minutes

values denote merely the intensity of reflectance and these do not necessarily correlate with the quantity of coloring agent deposited within the hair. Nevertheless, the shade intensity imparted by the DHI system is comparable to that obtained from commercial oxidative dye formulations (Table III). It should be pointed out that although in the experiments reported here the color-forming reactions were performed promptly after the metal pretreatment, we have found that the catalyst efficacy is not subject to aging, i.e., the hair can be pretreated with metal salts days or weeks before exposure to DHI to impart the desired color range.

PROPERTIES OF COLORED HAIR

Location of the melanin dye. Microscopic examination of the dyed hair reveals that the melanin is deposited primarily in the periphery of the hair fiber (Figure 4). This is not

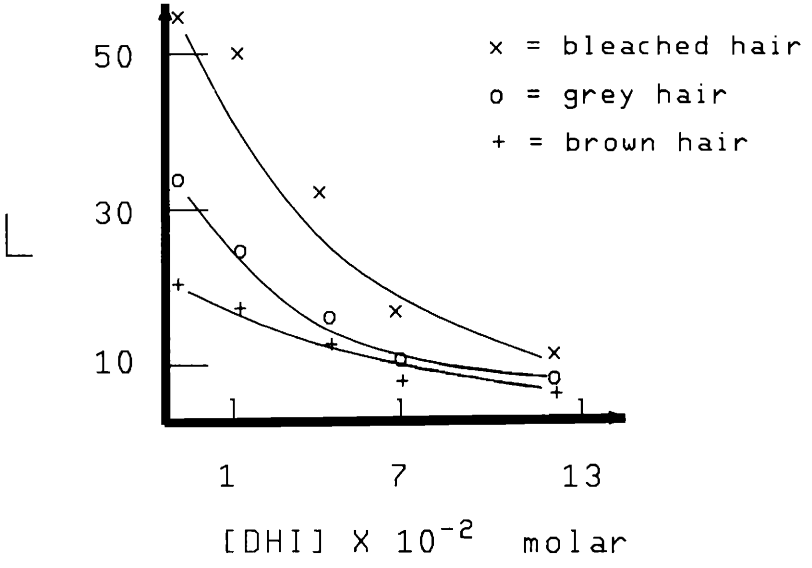

Figure 3. A plot of L values for bleached, blended grey, and brown hair dyed with various concentrations of DHI.

Table III
Color Parameters (L, a, b) for Grey Hair Dyed With Various Commercial Products and DHI

Product	L	a	b
Permanent[a]	14	0.7	2.1
Semi-permanent[b]	26	0.2	4.2
0.07 M DHI	13	0.1	0.1
0.013 M DHI	12	0.3	−0.2

[a] Clairol Nice N'Easy #122 Natural Black.
[b] Clairol Loving Care #83 Natural Black.

surprising in view of the short contact time for both the metal catalyst and the melanin precursor exposure. It is, however, instructive to see that even in this case, the intensity of imparted color is high and matches either the color of natural hair or conventionally dyed fibers (Table III).

Color stability. Evaluations, summarized in Table IV, suggest that, in general, the response of melanin color to a variety of post treatments is intermediate between those of permanent and semipermanent colorants. Bearing in mind the widely recognized chemical and physical inertness of melanin, it may appear surprising that the color is not more durable. It is important to make two observations regarding this matter. First, the color deposits, as shown above, are in the fiber periphery and are readily accessible to other reagents, unlike the melanosomes in natural hair which are embedded within the fiber cortex. Second, the melanin deposits, while undoubtedly particulate (TEM

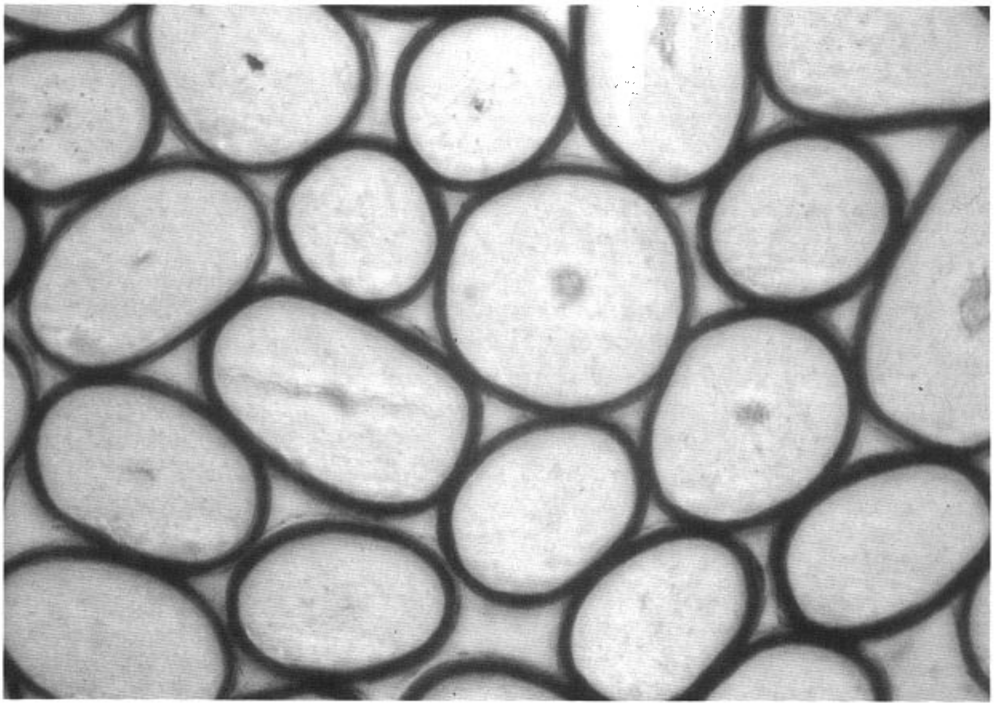

Figure 4. Optical micrograph of virgin white hair dyed with DHI. Note ring dyeing only.

Table IV
Effects of Post-Treatments on Color Stability of Hair Dyed With Different Coloring Systems

	Total color change		
Treatment	DHI	Permanent	Semi-permanent
Water bleed	3.35	3.11	10.7
Five shampoos	2.63	3.25	13.23
Acid perspiration	5.89	0.95	10.8
Fade-O-Meter 10 hours	3.20	0.41	2.07
Wave 1[a]	7.70	2.44	11.12
Wave 2[a]	8.08	2.86	9.93
3% H_2O_2	17.2	2.25	3.38
Relaxer[b]	3.26	1.16	4.74

[a] Wave 1 is Kindness Extra Curly Perm for color-treated hair. Wave 2 is Kindness Body Wave for natural hair.

[b] Relaxer is Revlon Realistic Relaxer.

evidence points to this), consist only of small particles and thus are much more readily subject to chemical modification than the larger natural melanosomes.

Color modulation and removal. The stability of the color imparted to hair by current products is a drawback if a color change or its total removal is needed. However, an interesting and potentially useful aspect of the melanin dyeing system is that the chemical nature of the colorant (melanin) and its cuticular location can be exploited for changing or removal of the imparted color. The agent most useful for this function, not surprisingly, is hydrogen peroxide. The degree of color modification (apparent both in the intensity and its tonality) is a function of peroxide concentration and contact time on the dyed hair. While the application of 3% H_2O_2 can, in a few minutes, remove essentially all of the deposited dye, lower concentrations are able to change the black color to lighter and warmer shades of brown and provide a method of extending the shade range. Since the peroxide treatment times are short, only the synthetic melanin located in the cuticle is bleached and the reagent does not reach the natural melanin imbedded deeper within the fiber. Thus, hairs can rapidly be dyed black, modified by several shades, and if desired, bleached back to principally the initial shade.

The effects of peroxide concentration on color changes are shown in Table V. As

Table V
Effect of Hydrogen Peroxide Concentrations on the Color Parameters (L, a, b) by Bleaching DHI-Dyed Hair

$[H_2O_2]\%$	L	a	b
DHI-dyed	17.3	0.0	0.4
0.1	20.6	0.0	2.6
0.5	26.3	0.9	5.5
1.0	27.3	0.5	5.8
3.0	32.8	0.7	7.9
6.0	33.1	0.8	7.8
Undyed grey	33.0	−0.5	9.1

pH = 10; bleaching time 10 minutes.

bleaching progresses, the imparted hair color changes its chromaticity, which allows for controlled modification to various hues of brown.

With prolonged time, the initial hair color is virtually recovered, except in the case of very light grey or blond hair where some degree of yellowness is evident.

CONCLUDING REMARKS

The melanin coloring methodology can be viewed broadly as one that embraces all currently available hair coloring systems, permanent, semi-permanent, and temporary. With some limitations, it can be any one. As we can only add color, the obvious drawback is the lack of shades lighter than the untreated hair. While the process of color impartation is simple, the available color palette is at present limited. Further exploration in this area is clearly necessary.

ACKNOWLEDGMENTS

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