

Morphological Damage Procedures of Hair Surface Treated with Repetitive Oxidation Coloring Agent

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ABSTRACT

Background/Objectives: People apply permanent hair dyes in various colors to change their hairstyle or appearance. Frequent dyeing damages the hair cortex and cuticle layer.

Method: In this study, a permanent coloring agent was applied to normal hair five times consecutively and the damage on the hair surface was observed with a scanning electron microscope (SEM).

Findings: In this study, the hair surface was continuously damaged as the number of permanent dyeing procedures increased. As the number of permanent dyes applied increased, the surface of the repeatedly dyed hair cracked and was lost in the cells of the cuticle. As the number of dyeing procedures to the same hair increased, the surface area of the scales making up the hair became wider and some broken cells remained attached to the surface of the hair scales.

Improvements/Applications: Damage to hair due to dyeing was confirmed by scanning electron microscopy. This study indicated that frequent dyeing should be avoided in order to prevent excessive hair damage.

Keywords: hair, hair dye, permanent colouring agent, SEM, scale, cuticle layer

Introduction

In modern society, many people vary their hairstyle to express their individuality and make themselves more attractive. Hairstyling is achieved in various ways, depending on an individual's age, personality, and social activities. Hairstyling practiced at beauty shops is divided into styling by physical procedures and methods using physicochemical reactions. Hairstyling by physical methods includes haircuts using a variety of tools, such as scissors, curlers, hairdryers, and combs. Hair styling by physicochemical reactions includes hair dyeing, bleaching, and making permanent waves^[1,2]. In recent years in Korea, with the liberalization of high school students' attire, there has been an acceptance of

the application of physicochemical reactions to the hair, such as hair dyeing and bleaching, and people as young as adolescents are interested in using these techniques to express their personality and change their appearance. In order to express an individual's appearance, not only in Korea but also throughout the world, hairstyling techniques applied to the scalp and hair can be done from adolescence to old age.

Dyeing, which changes the hair shade and color, is classified into temporary dyes, semi-permanent dyes, and permanent dyes, depending on the physical reaction between hair protein and the dye and the presence or absence of chemical reactions. Temporary dyes and semi-permanent dyes physically dye hair by the application of material to the hair and there is no chemical reaction with the hair proteins. These products have the color of the dye they are imparting^[3]. Temporary dye agents have chromophore groups which absorb light at particular wavelengths and form a hue. Most of them are azo-group-containing macromolecules which attach to the surface of the cuticle layer and exhibit a specific color^[4]. Semi-permanent dyes can be washed several times. The

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dyed hair gradually fades in color, rather than being washed away with one washing. Direct coloring is a physical reaction in which the dye material is deposited between the exposed cuticular cells on the surface of the hair. Semi-permanent dyes are used to enhance the hue of hair and to change it into gray hair but these dyes do not contain decolorant components and, therefore, cannot exhibit bright hues^[5].

Permanent dyes cause an oxidation dye reaction in which the dye precursor, which is the primary intermediate, enters the cortex inside the hair and reacts with the coupler resorcinol to form a large dye molecule. One dye precursor used in permanent dyes is *para*-phenylenediamine, which does not itself form a chromophore group. In cuticular cells and the cortex of hair, *para*-phenylenediamine is oxidized to form the quinone diimine and a chromophore group is formed in the dye molecule as reactions with resorcinol occur. These molecules continuously undergo oxidation and condensation reactions, eventually forming large dye molecules inside the hair cortex and permanently dyeing the hair.

Through this process, physical damage to the cuticle layer and cortex of the dyed hair occurs due to the alkaline and oxidant components present in the hair dye. As the hair dye procedures are repeated, the degree of damage to the entire hair fiber becomes severe, particularly damage to the cuticle layer on the surface of the hair^[6,7]. The physicochemical action artificially applied to the hair exposes the cuticular cells of the cuticle layer and the scales forming the surface of the hair separate, break, and crack but this damage cannot be observed with the naked eye. When the naked eye looks at damaged tresses, the hair simply appears as rough, dry, or crumbly.

Therefore, this study investigated the degree of damage to hair, the roughness of the hair surface and the morphological changes to the cuticle layer by repeated hair dyeing with permanent hair dye using a scanning electron microscope.

Materials and Method

Materials

Collection of Experimental Material: The normal hair of a woman in her 20s was cut to a length of 10 cm from

the scalp about 1 cm away from the scalp and was bundled into a hair tress with a forceps and fixed with silicone.

Permanent Dye Treatment: Agent 1 permanent hair dye (W company, 3/0, Japan) and agent 2 oxidant were mixed in a ratio of 1:1.5, evenly applied to the hair tress, and left at room temperature for 40 minutes. Then, the hair was washed with lukewarm water using shampoo, dried naturally, and a sample of the dyed hair was collected. To determine the degree of damage to the surface of the dyed hair with an increasing number of permanent dyes, the first, second, third, fourth, and fifth permanent dyes were applied at intervals of seven days in the same manner as above. Then, each sample was collected and used as experimental material.

Method

Observation of the External Shape Change of Hair Treated with Permanent Coloring Agents: After each hair sample was treated by the application of one to five permanent dyes, they were cut to lengths of about 1.5 cm according to the sample processing method of Chang^[8]. Pre-fixation was performed using paraformaldehyde–glutaraldehyde, (4°C, 0.4M phosphate buffer, pH 7.4) as a pre-fixing agent and post-fixation was performed using 1% osmium tetroxide (OsO₄, 4°C, 0.4M phosphate buffer, pH 7.4). After drying and dehydration with ethanol and isopropyl alcohol, the hair was naturally dried and the hair samples were placed on a stub treated with copper tape. The stub with the hair sample was platinum coated to a thickness of 20 nm by an ion deposition machine (IB-5, Eiko, Japan) and was irradiated with a scanning electron micrograph (S-4700, Hitachi, Japan) at 15 kV for observation.

Result

The surface of the hair was clearly observed in the scanning electron micrographs of normal hair used in this study. The scales, which are the exposed areas of the cuticular cells forming the cuticle layer on the surface of the hair, were observed in oval shapes and some of the scales were observed to be broken apart at the tip [Fig. 1a]. High-resolution scanning electron micrographs showed no gaps between the superimposed cuticular cells and they were closely attached to the lower cuticular cells. The surface of the scales was smooth [Fig. 1b].

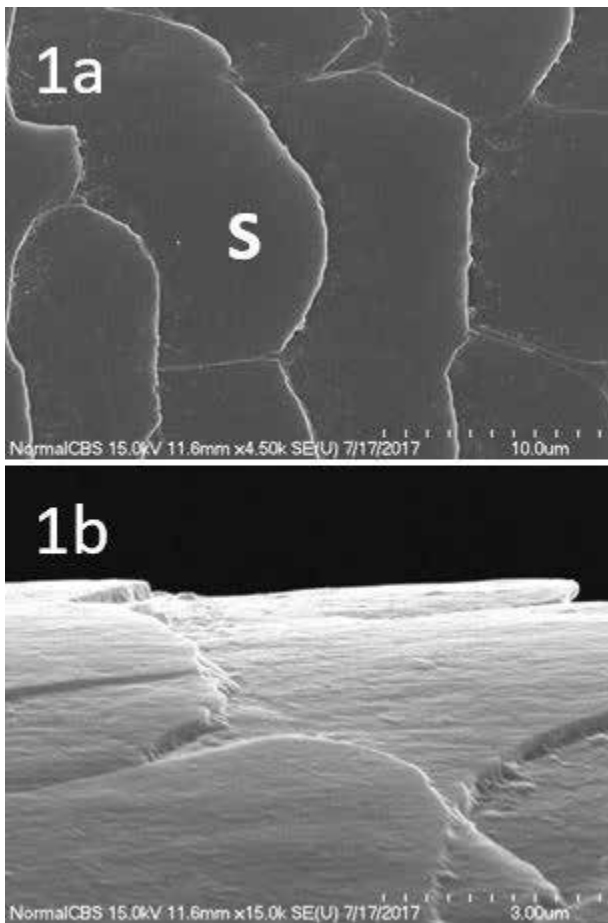


Figure 1: Scanning electron micrographs of normal hair. a: The hair surface showing round scales (S). b: High-magnification scanning electron micrograph of normal hair

In scanning electron micrographs after the first application of permanent hair dye, the hair surface was observed to be rougher than that of normal hair. The distal region of the scales was pointed and irregular. The surface of the hair scales was partially depressed and damaged by the physical action of the dyeing process and damage to the edges of the scales formed very irregular shapes [Fig. 2a].

The high-magnified images showed a fine gap of about $0.1\mu\text{m}$ between the superimposed cuticular cells of the surface. The area of the cell edge section about $5.7\mu\text{m}$ thick that was broken off appeared clear [Fig. 2b]. On the surface of the scales exposed under the part where the cuticular cells on the surface of the hair broke apart, there was a cell remnant of the endocuticle which was not separated from the surface [Fig. 2b].

The hair washed after the second permanent dye, the surface area of the scales exposed to the hair surface was

very irregular. After the primary dyeing, the separated or broken cuticular cell remnants were partially washed away [Fig. 3a].

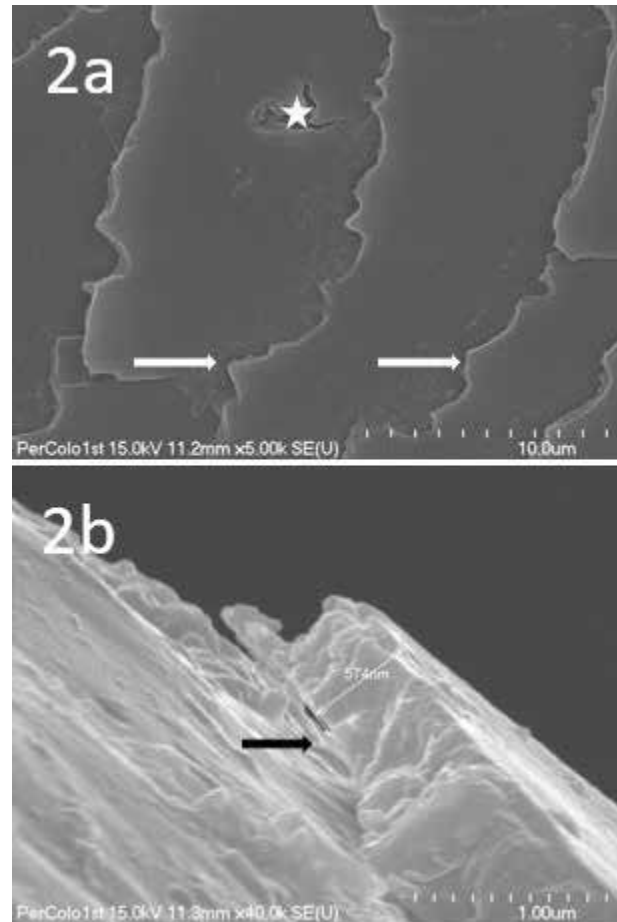


Figure 2: Scanning electron micrograph of the hair surface after the first permanent-hair dyeing. A: Irregular terminal edge of the scales due to shattering(a). b: Cracks (black arrow) developed among the cuticular cells. White arrow: edge of scale. Asterisk: depressed crack.

After the second application of permanent dye, the hair surface was rough, the surface of the cuticular cells was covered with a remnant of the endocuticle, and the edges were very irregular with sharp points [Fig. 3a]. In addition, separated cuticular cells were attached to the surface of the hair and some cell remnants which were not separated at the edge of the cell bent toward the free surface of the hair [Fig. 3]. Separation of the cuticular cells on a scanning electron micrograph at high-magnification showed that the cuticular cells were not completely separated but that the endocuticle of the cuticular cells was broken off [Fig. 3b].

The third permanent-dyed hair showed more scales separated from the surface than the second permanent-

dyed hair and a large gap between the separated scales. The surface area of the exposed scales was widened. The scales on the surface of the creviced hair were continuously separated from the overlapping scales [Fig. 4a].

Specifically, the high-magnification scanning electron micrograph confirmed that the surface of the hair was relatively smooth and that slight wrinkles had formed. In addition, the separated cells were completely detached and the surface of the underlying cuticular cells was clearly observed (Fig. 4b).

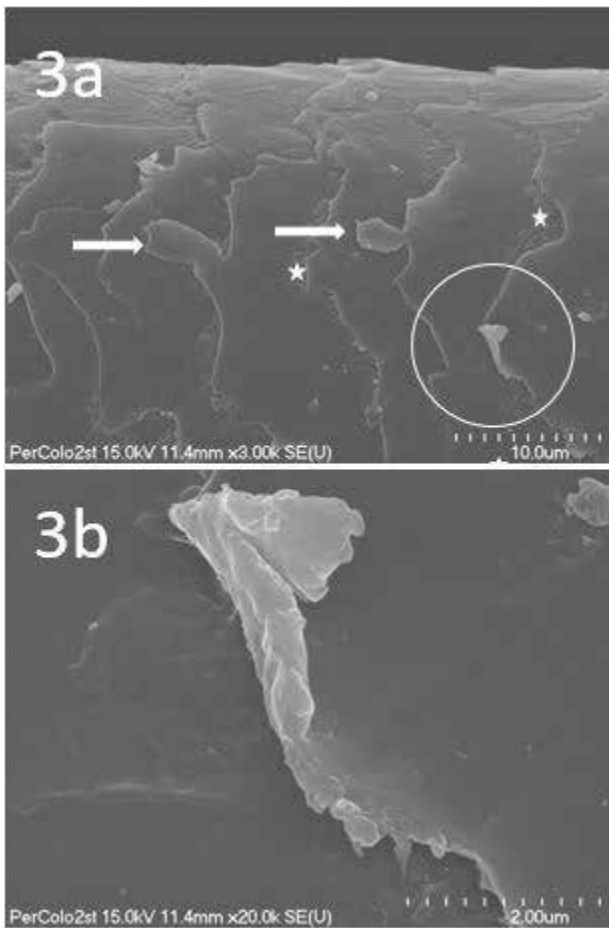


Figure 3: Scanning electron micrograph of a hair after the second permanent-hair dyeing. a: Scales on the hair surface are shattered (arrow) and the endocuticles (asterisks) are attached to the cellular membrane. Circle: separated curved. B: Separation of the endocuticle and exocuticle can be observed in the cuticular cells.

The fourth application of permanent hair dye a showed relatively smoother surface than the third permanent-dyed hair but the overlapping cuticular cells of the cuticle layer were observed to be torn off in some areas. On high-magnification scanning electron

micrographs, the cell remnants remained attached to the surface of the tears and the scale surface was cracked [Fig. 5]. At the depressed sites where the cuticular cells were severely torn, cuticular cell damage was found and the cuticular cells adjacent to the cortex were cracked [Fig. 5].

The fifth permanent-dyed hair showed fewer cell remnants attached to the surface than the fourth permanent-dyed hair, where relatively few cell remnants attached to the surface or separated cell remnants were observed [Fig. 6]. The surface of the fifth permanent-dyed hair was found to have a relatively smooth appearance, although some cell remnants were present. On the high-magnification scanning electron micrographs, the cuticular cells were physically and chemically deformed into cave shapes and some cell remnants were present but the hair surface was smooth [Fig. 6].

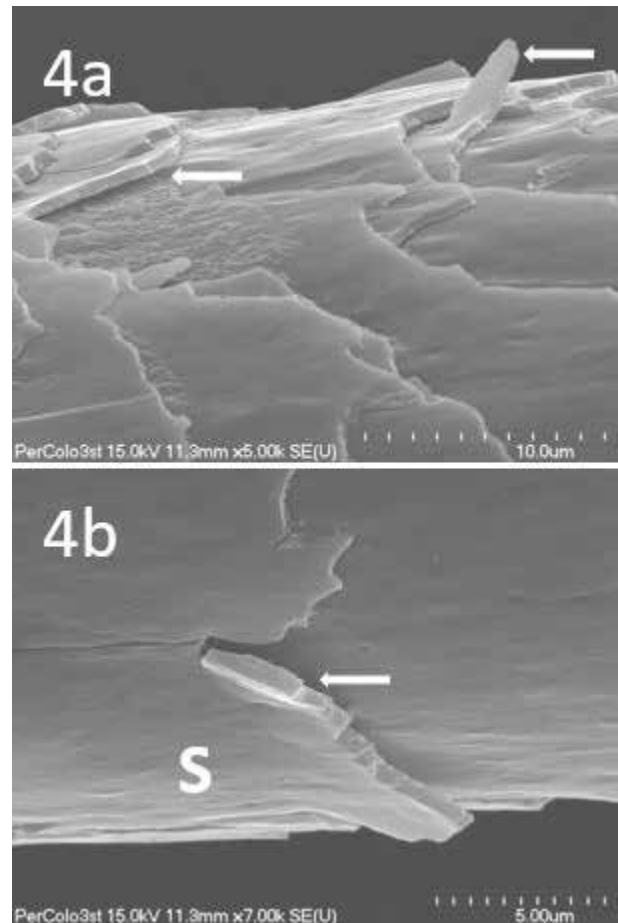


Figure 4: Scanning electron micrograph of a hair after the third permanent-hair dyeing. a: Continuous separations (arrow) of the cuticular cells can be seen. b: Magnification scanning electron micrograph of the hair surface showing separated crack (arrow). S: scale.

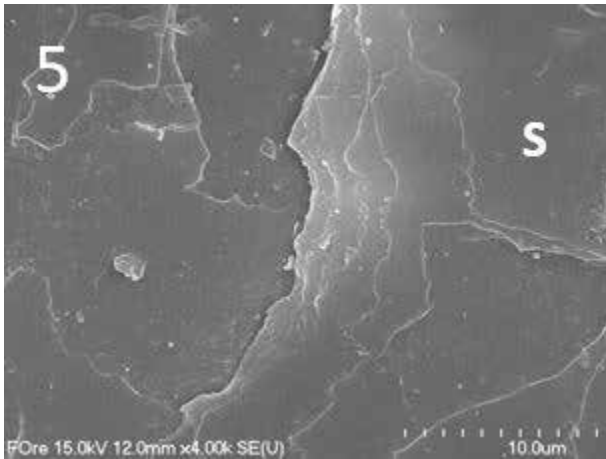


Figure 5: Magnified scanning electron micrograph of the hair surface after the fourth permanent-hair dyeing. The damaged hair surface shows depressed cracks. S: scale.

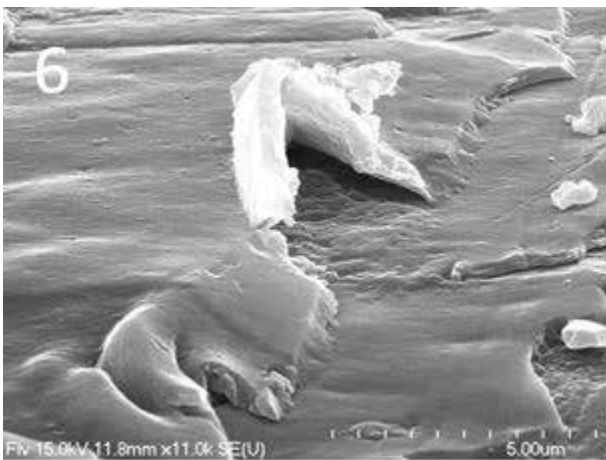


Figure 6: High-magnification scanning electron micrograph of the hair surface after the fifth permanent-hair dyeing. Swelling of the cuticular cell on the hair surface is seen.

Discussion

Scalp hair grows from the hair follicle and is physically and chemically damaged from the point of exposure to the skin surface. Damage to the hair growing on the scalp is primarily caused by weathering phenomena and secondary physicochemical actions, such as hair dyeing, discoloration, permanents and drying for hair styling. Scalp hair is damaged during frequent hair curling and combing, sun exposure, and wind-induced friction during the anagen phase of hair growth. In addition, the high salinity of sea water, the chlorine compounds used in pools, and polluted air inflict hair damage called weathering^[9-13]. In this study, normal hair was observed to have a relatively clean surface and no severe weathering phenomenon was observed.

Modern people change their natural hair color, sometimes repeatedly over many years. Dyeing to bring about a permanent change in hair color uses permanent dyes or oxidation dyes. Oxidation dyes create dye by a chemical reaction between the dye materials in the hair cortex and can produce a color brighter than the natural hair color^[14].

In this study, brown hair color was pigmented with a permanent coloring agent and the surface of the hair was observed with a scanning electron micrograph. The end of the scales on the surface of the hair appeared broken and irregular. In a scanning electron micrograph study of the outer shape of permanent-dyed hair, Lee & Chang^[8], reported that the scales of the permanent-dyed hair surface were partially separated and that these cuticular cell pieces had lower electron density. In this study, we confirmed that the outermost cuticular cells of the primary permanent-dyed hair cuticle layer were damaged and separated, in agreement with Lee & Chang^[8].

In this study, scanning electron micrographs after the application of a second permanent dye revealed that the surface area of the hair was more irregular than that dyed once with permanent dye. In addition, the separated cuticular cells did not completely fall off and the endocuticle remnants were attached to the surface of the cuticle cell membrane located below them.

The third permanent-dyed hair had more scales separated on the surface than the second permanent-dyed hair and there were many fine gaps between the underlying cuticular cells. As these gaps spread, the cuticle layer of the hair was separated continuously. Lee & Chang^[15] observed the cuticle layer of permanent-dyed hair by transmission electron microscopy and found that the cuticular cells of the surface were cleaved and separated from the lower adjacent cuticular cells. They reported that high electron density analysis showed encased 7-8nm thick fine particles between the surface and the gap in the cuticular cells. These fine particles reacted with a coupler in the dye to form large molecules before they penetrated into the cortex as the dye intermediate was oxidized. Small molecular size particles on the surface of permanent-dyed hair were not observed in the scanning electron micrographs of this study.

In this study, the fourth permanent-dyed hair was more smooth than the third permanent-dyed hair but the scales were already removed and the surface of the

exposed cuticular cells was torn off. On high-power scanning electron micrographs, the continuously-torn surface of the cuticular cells was depressed. This result was due to repeated permanent physicochemical damage to the hair surface by the continuous permanent dye procedures to the hair. The alkalinity of hair dye products swells the keratin protein of the hair cuticle layer and diffuses the dye precursors into the cortex between the swollen cuticular cells^[16].

The fifth permanent-dyed hair showed relatively fewer cell remnants separated from the surface-attached cell remnants left from the fourth dyeing. The surface of the hair was relatively smooth compared to the third and fourth dyed hair. As a result, the fifth permanent-dyed hair had relatively few cell remnants separated from the surface-attached cell remnants than the fourth permanent-dyed hair and the surface area of the scales was continuously widened. Therefore, most of the cuticular cells and cell remnants were separated and the hair surface was found to be relatively smooth.

Results of this study confirmed that the hair was continuously damaged as the number of permanent dye treatments increased. The dyeing process damaged the hair cuticle layer, causing the cut surface of the cuticular cells on the surface to collapse and fall off and the surface area of the scales became wider. With dyeing, hair was cracked but could be washed and appeared clean to the naked eye. However, when the dyeing process was continuously repeated, the cuticle layer of the hair was excessively damaged, exposing the cortex, eventually necessitating cutting the damaged hair.

Conclusion

A permanent coloring agent was tested five times on normal hair and damage on the hair surface and microstructural changes of the cuticle layer were observed with a scanning electron microscope.

The surface of the normal hair used in this study was clearly observed but the surface of the first permanent-dyed hair was rougher than normal hair. The ends of the scales of the second permanent-dyed hair surface were broken and very irregular. The third permanent-dyed hair showed more scales separated on the surface than the second permanent-dyed hair. The hair of the fourth permanent-dyed hair was smoother than that of the third permanent-dyed hair, but the overlapped cuticular cells

were observed to be partially cut off in some areas. The fifth permanent-dyed hair was found to have relatively few cell remnants separated from the cell remnants attached to the surface. The surface of the hair was smooth and the surface area of the scales was widened.

In conclusion, as the number of permanent dyes applied to the hair increased, damage to the surface of the hair increased. The hair surface was physically and chemically damaged by the combs and hair dye used during the dyeing processes. The dyed hair repeatedly cracked and was torn in the cuticle layer surrounding the cortex and the surface area of the scales which make up the hair became wider. When the broken cellular remnants were cleanly washed, however, the hair appeared cleaner to the naked eye.

Ethical Clearance: Not required

Source of Funding: The research in this study was conducted under the 2017 Hanseo University intramural research support project.

Conflict of Interest: Nil

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