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New Aspects of the Structure of Human Scalp Hair-II: Tubular Structure and Material Flow Property of the Medulla

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Synopsis

Asian scalp hair fibers were made thin by treatment with papain or sliced along the longitudinal axis or randomly cut by mechanical means. Optical microscopic observations of the resulting specimens indicated that (i) the medulla (M) consisted of two types of the M-surrounding cells which were linearly linked one another to form a tubular structure running through the fiber and (ii) the drum-shaped vesicles containing small proteinous granules were neatly or sparsely stored within the tube. On the other hand, H^+ and OH^- ions were able to move spontaneously from one end to another through the M tube. Large molecules such as an anthocyanin dye (from purple sweet potato) were also capable of flowing through the M tube, especially rapidly when DC voltage was applied between the two ends of the hair fiber. The possible function of the M is briefly discussed in conjunction with the tubular structure and the material flow property.

INTRODUCTION

Previously we reported the new structural model of an Asian scalp hair fiber, especially focusing on the morphology and the spatial arrangement of the cortical and cuticular cells (1); see Figure 1. Although a lot of studies have been carried out over the last several decades using the optical and electron microscopes (2–10), our experimental method, which was characterized by random scission of the fibers, led to many revisions of the traditional hair structural model (7,11).

The present investigation concerns the medulla, which exists in the middle of the fiber and has two aims. The first, is to obtain the detail structure of the medulla. Optical microscopy was fully used in the study; in other words, whereas electron microscopy works in vacuo and provides only a black-and-white photograph of the dried substance, the optical

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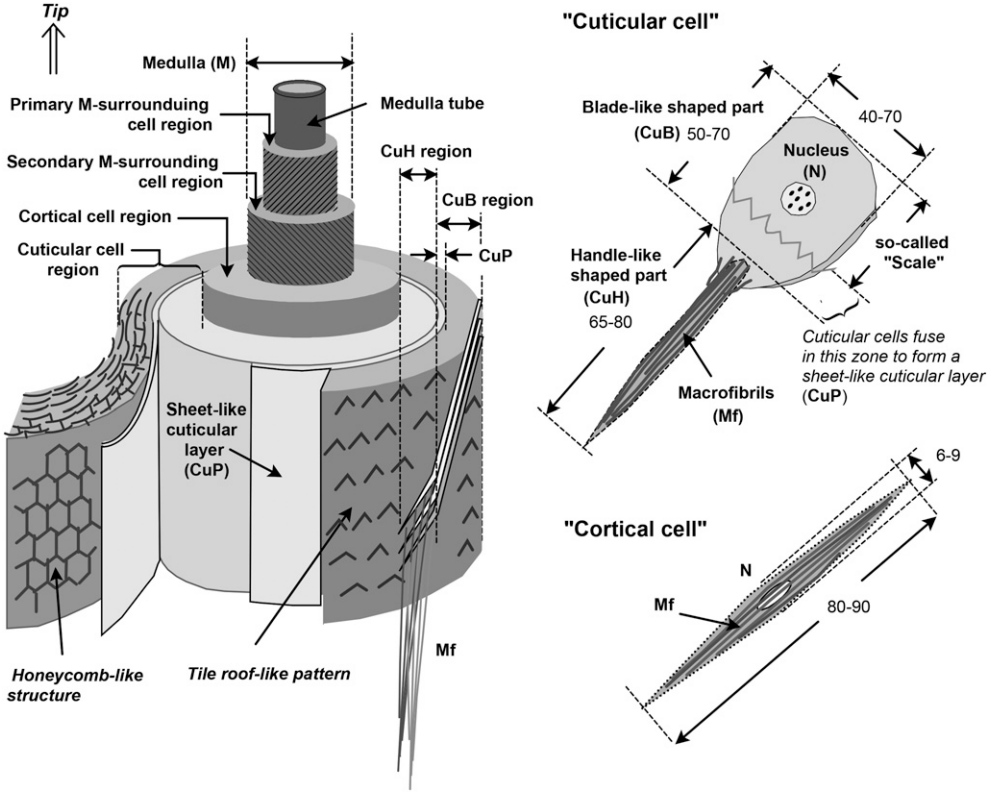

Figure 1. Schematic representation of the overall structure of a human scalp hair fiber. The previous model (1) is partly modified to match with the present observations of the M. Each of the hair parts is explained elsewhere in the text. The unit for the length is micrometer.

microscopy not only suits for wet substances but provides a see-through image with a high depth of field in semi-micro and micrometer levels, often making it possible to distinguish chromatically between two or more substances in the specimen. The second, is to examine the material flow property of the medulla. To date, the hair component has long been considered just a loosely packed assembly of the cellular disks connected in series and not performing any significant function in hair (9,11–16). The more detailed structural analysis of the medulla and its function, if any, may be very useful to evaluate the interaction of human hair with various kinds of chemical and biochemical substances including cosmetic products.

EXPERIMENTAL

HAIR SAMPLES AND REAGENTS

A Japanese girl donated her black hair fibers (jf8); cf. the Arabic numeral refers to the donor age. The fibers were cut from more than 1 cm from the scalp surface. Black hairs (cf16) were similarly collected from a girl of the Miao ethnic group living in the mountains of China's Yunnan province. White hairs, jf65 and jm67, were furnished from Japanese

female and male, respectively. Although it is not mentioned herein, various other persons also donated their scalp hairs to the present study. All of the fibers were straight, neither being stained with dyes nor subjected to any permanent setting processes. The fibers were successively washed with a 1.5% (w/w) aqueous solution of sodium dodecyl sulfate (SDS), deionized water, and 70% (v/v) ethanol and then stored at 4°C in a sealed plastic container. The reagents including an anthocyanin dye (from purple sweet potato; a mixture of cyanidin acylglycoside and peonidin acylglycoside; Kiriyachemi, Osaka, Japan) and purified papain powder from *Carica papaya* (Wako Pure Chemical Industries, Osaka, Japan) were commercially available.

MICROSCOPE OBSERVATION

The biological microscope, Olympus, Tokyo, Japan; model Vanox (AH), was used mainly for bright field, phase contrast, and polarized light observations. The inverted microscope and the stereomicroscope, Olympus model CK30 and X-Tr, respectively, were modified so as to place the observation chambers and the electrode cells (vide infra) on the microscopes. The microscopes were equipped with a digital camera, which was automatically controlled by a desktop computer to optimize for lighting, ISO levels, and focusing measures. The images in JPEG and RAW formats were developed by means of Lightroom ver. 3 and Photoshop Elements ver. 9 software (Adobe Systems Inc., San Jose, CA). Image enhancement included color level correction, noise reduction, and contrast and brightness adjustments; the purpose of the enhancement was to make the image nearly identical to that seen actually by the observers. In the case of thick specimens, the images taken at various depths of field were merged into a single deep focus picture by the use of the stacking software, Combine ZP (A. Hadley, Sheffield, United Kingdom) (17).

SPECIMEN PREPARATION FOR STRUCTURAL ANALYSIS OF MEDULLA

Enzymatic thinning of hair fibers using the papain (a representative procedure). Several strands of the hair (jf8, about 15 mm in length) were warmed in pH 7/0.067 M phosphate buffer solution (PBS) at ambient temperature for 24 h. After washing briefly with water and drying at ambient temperature, the fibers, usually 1–3 strands, were placed parallel on a glass plate (22 × 22 × 0.17 mm) and both ends of each fiber were glued to the plate with a rapid type–epoxy resin; see Figure 2(A). Next, the glass plate having the fibers was fixed by the use of the epoxy resin on the window frame (20 × 20 mm) which had been made in the bottom of a plastic culture dish (diameter, 33 mm; depth, 10 mm). After washing with PBS overnight at ambient temperature, the papain solution (20 units in 3 mL of PBS) containing its activator of SDS (12 mg) and 2-mercaptoethanol (ME) (65 μL) was put into the dish, and the digestion of the fibers was allowed to proceed at 30°C. The inverted microscope was used to observe the morphological change of the fibers.

Random mechanical cutting of enzymatically thinned hair fibers (a representative procedure). Several strands of the white fibers (jf65 about 15 mm in length) were treated by dipping in PBS at ambient temperature for 24 h, and then warmed in the PBS solution (3 mL) of papain (20 units), SDS (12 mg), and ME (65 μL) at 30°C with occasional gentle shaking. After about 3 h, a new crop (10 units) of the enzyme was added to the reaction mixture. During incubation, one strand of the fibers was periodically sucked into water (1 mL)

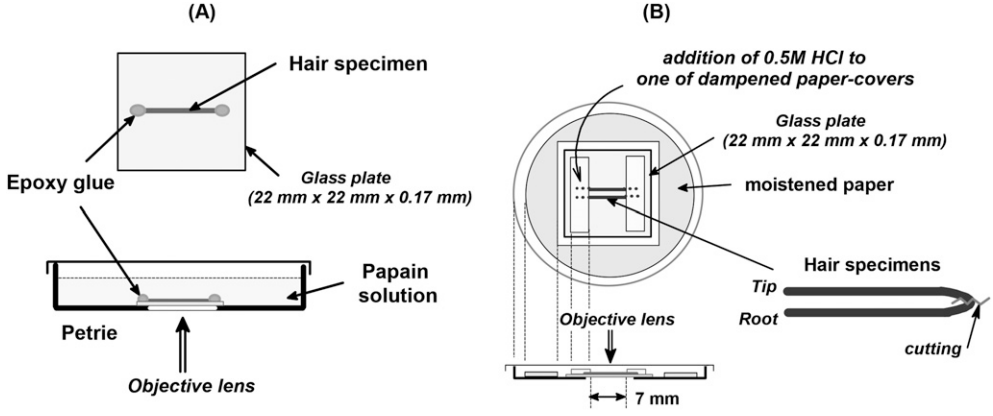

Figure 2. The observation chambers for the material flow study of the M. The type (A) was used for the inverted microscope, whereas the type (B) was mainly employed for the upright microscope.

using a pipette, then randomly cut with rapidly rotating poly(ethylene) blades in a manner similar to that mentioned previously (1). The resulting fragments were collected by means of a centrifuge, stained with a Giemsa's solution (Wako Pure Chemical Industries, Osaka, Japan) (18), washed briefly with a 50% (w/w) aqueous solution of glycerol, and then mounted to a microscope slide glass. A weight (30 g/cm^2) was placed on the cover glass (grade: No. 1; Matsunami Glass, Osaka, Japan) while sealing the edges with Canada balsam.

Mechanical slicing of a hair fiber. Several strands of hair (cf16) were dried and embedded in an epoxy resin (Poly/Bed 812) according to the protocol of PolySciences (Warrington, PA). The solidified resin block was sectioned to about 8 μm thickness by means of the microtome (American Optical, Buffalo, NY; model 820) and stained with a 0.5% (w/v) aqueous solution of gentian violet. The microscope specimen was prepared in a manner similar to that described previously.

MATERIAL FLOW PROPERTY OF MEDULLA

H^+ ion-flow through the medulla: the use of Congo red as an indicator. The hair fibers (jf8, about 25 mm in length) were kept in an aqueous solution (5 mL) of 7 M urea, 2% (w/v) SDS, and 15% (v/v) ME at about 55°C for 20 min.[1] The resulting fibers were washed briefly with water and dipped in a 0.025% (w/v) aqueous solution of Congo red (5 mL) for 1 d at ambient temperature. Three strands of the wholly stained hair were then placed, each in a U-letter shape, on the glass plate (22 × 22 × 0.17 mm) which had been glued with the epoxy resin to the window hole made in the bottom of a plastic Petrie chamber

[1] Treatments of hair and wool fibers with the medium containing ME for long time at higher temperature was previously reported (19, 20). In "H^+ ion-flow through the M: The use of Congo red as an indicator" process, the hair fibers were softened by soaking in the presence of ME for a short time of period to wholly stain the fiber body with Cong red. During the staining period, the swollen shafts were nearly brought back to the dimensions of the starting unheated wet fibers. Although keratins, the main proteins of hair, might be structurally disturbed to some degree in the dyeing process, H^+ ions was found to flow preferentially through the medulla. The "OH^--flow through the M: the use of phenolphthalein as an indicator" process, on the other hand, did not use ME in the flow study of OH^- ions.

(diameter 55 mm, bank height, 3 mm); cf. Figure 2B. Subsequently, the middle position of the U-shaped fiber was cut with razor to give the two pieces which were hence antiparallel to each other on the glass plate. Next, each of the right- and left-end sections of the paired shafts was overlaid with a dampened blotting paper (5 × 15 mm), whereas the midsection (gap about 7 mm) of the fibers was not covered with the paper. When the whole system was equilibrated to 25° ± 1°C, a glass lid (0.17 mm thickness) was temporally removed to add 0.5 M hydrochloric acid (0.05–0.1 mL) onto one of the two dampened paper covers. The color change occurring in the hair shafts was analyzed by means of the upright microscope.

OH^--flow through the medulla: The use of phenolphthalein as an indicator. A 0.5% (w/v) aqueous solution of phenolphthalein was put into a glass vial (diameter 22 mm, height 55 mm) with a screw cap till the solution level reached about 3 mm. Subsequently, the white hairs (jm67, about 40 mm in length) were vertically stood in the vial at ambient temperature, where the root side was in the indicator solution. After 7–14 d, the fibers were briefly washed with distilled water and extended on the aforementioned Petrie chamber in a parallel arrangement. Both sides of each of the fibers were then covered with the dampened papers in a manner similar to that described in "H^+ ion-flow through the medulla: the use of Congo red as an indicator." When the whole system was equilibrated to about 25° ± 1°C, one drop of 1 M NaOH was carefully placed onto one of the blotting papers. The color change occurring in the hair shafts was followed by the use of the upright biological microscope.

Flow of the anthocyanin dye through medulla: Application of a DC voltage to a hair shaft. The white hair fibers (jm67; about 30 mm length) were warmed in an aqueous solution of 8 M urea and 4% (w/v) SDS at 55°C for 2 h. One strand of the softened fibers was put through

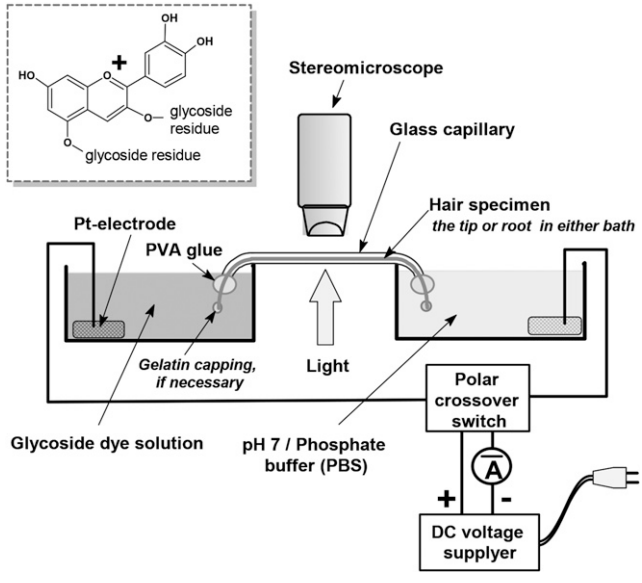

Figure 3. The instrumental setup for the DC voltage-assisted flow of the purple anthocyanin dye. The white hair fiber was put into a glass capillary, and the fiber ends were dipped into the electrode solutions as depicted in the figure. An aqueous solution of the cationic dye and the buffer solution were initially set as the positive and the minus electrode solutions, respectively; see the procedure "Flow of the anthocyanin dye through M: Application of a DC voltage to a hair shaft" for more details.

the U-shaped glass capillary (inner width 1.0 mm, wall thickness 0.1 mm, and length 25 mm) which had been filled with pure water; see Figure 3. After making each of the fiber ends protrude by about 3 mm in length from the capillary terminals, the water inside the capillary was soaked up with blotting paper, then the capillary ends were sealed with a poly(vinyl alcohol)-glue to prevent the infiltration of the electrode solutions (vide infra) into the capillary. Subsequently, the capillary with the hair shaft was placed horizontally between positive and minus electric bathes [both: 2 cm (W), 2 cm (H), and 1 cm (D)], where the former and the latter bathes had been filled with a 0.5% (w/v) aqueous solution of the cationic anthocyanin dye and a pH 7/0.067 M PBS, respectively. Initially, the root side of the hair fiber was put into the positive electric bath (anode). When the whole system equilibrated to about 25°C, DC 550 voltage, in most cases, was applied between the Pt-electrodes, using a power supply (model Crosspower 1000; ATTO, Tokyo, Japan) and an ammeter (model PC700; resolution, ±0.1 μA; Sanwa, Tokyo, Japan). By means of the stereomicroscope, a dark purple-colored zone was recognized exclusively in the medulla and became longer toward the cathode side with increasing time of the voltage application. By reversing the electrode polarity, the colored zone was quickly moved back or faded out from the anode side, which had once been the cathode side. It was also noticed that a direct current was not only varied with the hair samples but gradually increased with time; for instance, from 20 to 25 μA in one sample and from 1.3 to 2.5 μA in the other sample over a span of the experimental period (0–10 min). We consider that the current fluctuation was caused by the uncontrollable electric conductance over the wet hair surface and by the irregular inner structure of the medulla.

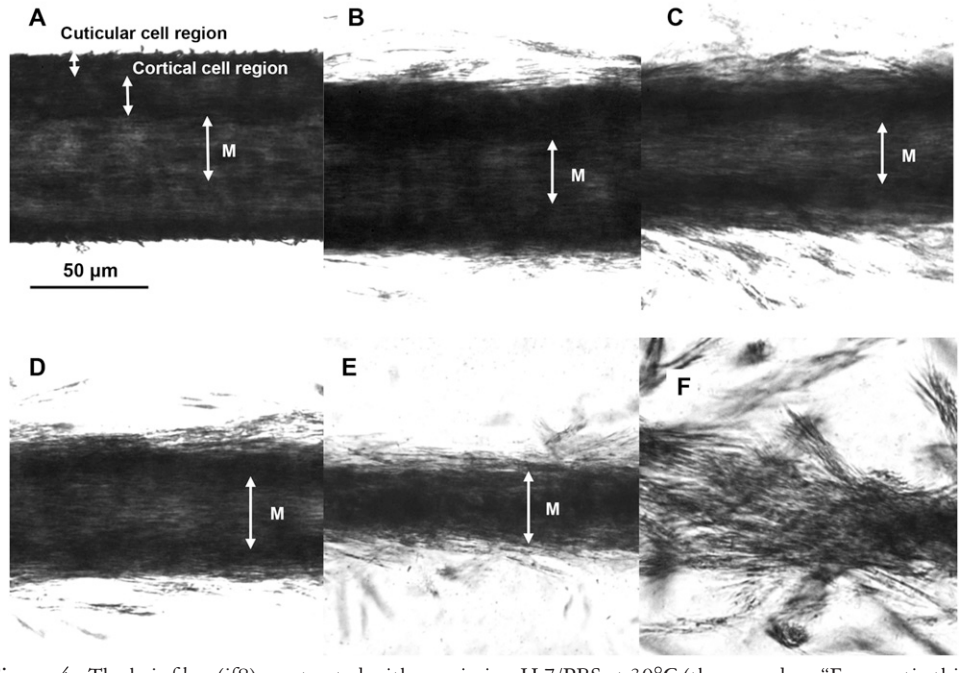

Figure 4. The hair fiber (jf8) was treated with papain in pH 7/PBS at 30°C (the procedure "Enzymatic thinning of hair fibers using the papain"). Pictures (A–F) were taken at the incubation time of 4 min, 2, 4, 6, 7, and 10 h, respectively. We consider on the basis of the diameter measurement that the dark part of the picture (E) is mainly composed of the M which consists of the M-surrounding cells and the tube; cf. Figure 6.

RESULTS AND DISCUSSION

MORPHOLOGY OF MEDULLA

Recently, we reported the following microscopic observations on the cellular components of human scalp hairs (1); see Figure 1. The cuticular cell took a trowel-like shape, consisting of a handle-shaped part (CuH) and a blade-shaped part (CuB). The CuB parts overlapped one another and partially fused together to form a layer (CuP) within the cuticular cell region. The cortical cell had a spindle-like shape and was similar in dimensions to CuH of the cuticular cell. The medulla, on the other hand, was appeared to possess a thin tubular wall. We, therefore, performed in the first phase of the present study a detailed analysis of the medulla structure, using the aforementioned advantages of optical microscopy. For this aim, the hair samples were either digested thin with papain or sliced with a microtome or randomly cut with a rotating cutter.

Figure 4 shows the time course of the structural change of the young girl's black hair fiber (jf8) by an action of papain in the pH 7/PBS at 30°C. Although the inverted microscope did not provide high-resolution images, the digestion of the hair fiber seemed to take place in a phased manner. In other words, the outmost cuticular cell region was slowly stripped off from the hair shaft during the first incubation period (about 5 h); Figure 4A–D. Then, the cortical cell region was digested within 2 h to provide the thin remainder which appeared to consist mainly of the medulla; Figure 4E. The elders' white hairs (jf65 and jm67) were digested in a similar fashion with the enzyme. The thin remainder such as the one seen in Figure 4E was useful for the structural study of the medulla. Namely, upon the mechanical cutting which was described in "Random mechanical cutting of enzymatically thinned hair fibers," two kinds of flat and long substances (width 4–8 μm, length 70–90 μm, thickness 2–3 μm) were formed from the remainder as demonstrated in Figure 5. One of them is assigned as "a primary M-surrounding cell" and the other as "a secondary M-surrounding cell." The primary M-surrounding

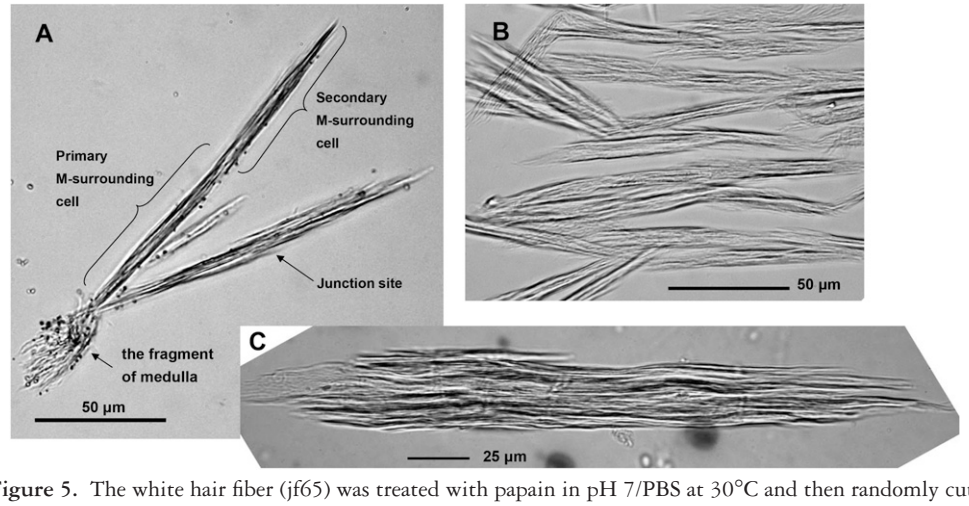

Figure 5. The white hair fiber (jf65) was treated with papain in pH 7/PBS at 30°C and then randomly cut; Giemsa staining; see the procedure "Random mechanical cutting of enzymatically thinned hair fibers." Panels (A and B) with bright field illumination; the panel (C) with oblique illumination.

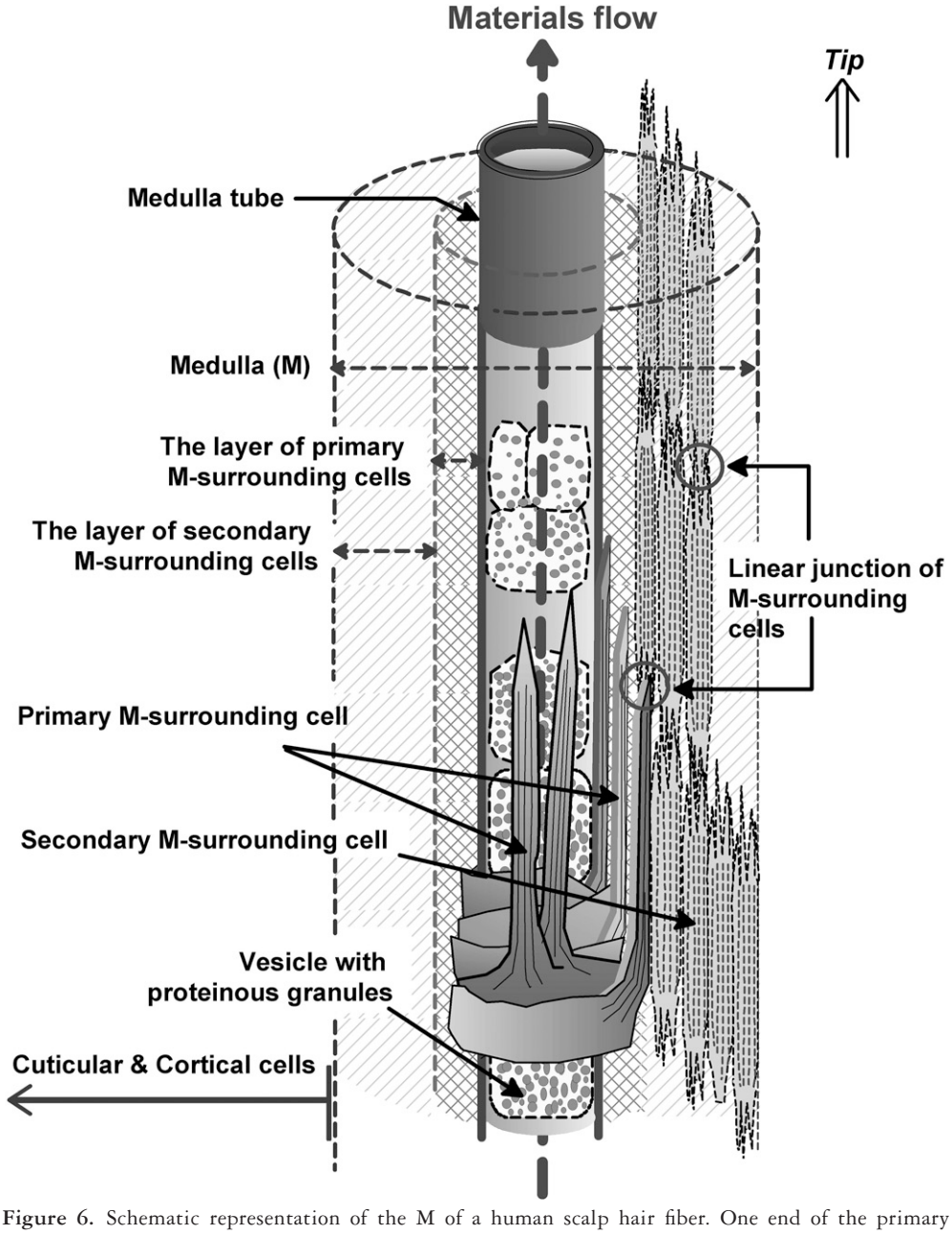

Figure 6. Schematic representation of the M of a human scalp hair fiber. One end of the primary M-surrounding cell is attached to the M tube, whereas the other end is bound to the secondary M-surrounding cell; cf. Figure 5A. The secondary M-surrounding cell also forms a linear junction with the other secondary M-surrounding cell. Hence, all of the M-surrounding cells are bound directly or indirectly to the tube wall, covering around the tube. In general, adults' M tube, unlike an elderly persons' tube, is neatly packed with the vesicles which are full of proteinous granules (0.5–1 μm in diameter). Various materials in aqueous solution may flow through the tube.

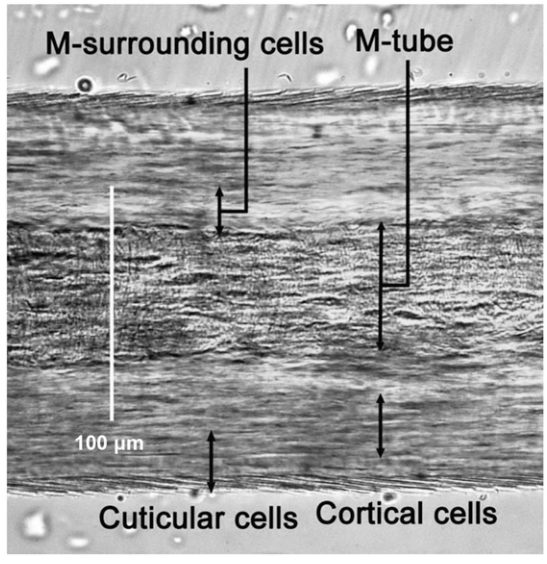

Figure 7. A polarized light microscopic picture of the white hair fiber (jm67). The medulla (M) appeared light brown for the most part. The regions of the M-surrounding cells, the cortical cells and the cuticular cells were seen in color yellow, light green and yellowish brown, respectively. The fibers were warmed in a solution of 8 M urea and 4 wt.% SDS at 55°C for 2 h, washed successively with water and ethanol, sandwiched between a slide glass and a cover glass using Canada balsam as a medium, then slightly pressed by means of a disk micrometer (CLM-DK; Mitutoyo, Kanagawa, Japan) in a manner similar to that mentioned previously (1). Upon the compression, the softened fiber (also swollen by about 1.3-fold of the original breadth) was flattened at least in the width of about 100 µm; hence, the retardation coloring due to an uneven thickness effect, if any, was minimized within the flattened area. Although the angle between two nicols were properly adjusted to distinguish between the hair components in color, the primary M-surrounding cell region was not differentiated from the secondary one. The M tube appeared to be somewhat widened and fractured by the compression.

cell was featured by the long thin flat body; see Figures 5A and 6. It appears that one end of the cell was attached on the surface of the medulla tube and the other end was linked to the secondary M-surrounding cell. In addition, a small irregular-shaped substance, like plant roots, was usually observed on the primary M-surrounding cell; see Figure 5A. It is considered that the irregular substance is a ripped off-product of the medulla tube (by the mechanical agitation), still binding partly to the primary M-surrounding cell. On the other hand, the secondary M-surrounding cell was characterized by the long, flat, and symmetric body with split ends; Figure 5B and C. The cell was connected to the primary M-surrounding cell or to the other secondary M-surrounding cells as shown in Figure 5A; see the linear junctions in Figure 6. The previous studies (9,11–16) on the hair structure did not mention the M-surrounding cells, presumably because the microscopic specimens were prepared by just cutting the hard hair fibers by means of a microtome. We found that two kinds of the M-surrounding cells were very similar to each other in the terms of the dimensions and the staining susceptibility to gentian violet, Giemsa *etc.*; therefore, these two kinds of cells are usually indistinguishable from each other, even under the polarized light illumination; for instance, see Figure 7.[2]

[2] In our previous microscopic observations, the cortical cell region was not differentiable from the M-surrounding cells; thereby, these regions were simply defined as a cortical cell region; cf. the Figure 2 in the report (1) for the cross section of a human hair fiber.

In addition to the M-surrounding cells, a large tubular structure was found to exist in the M as shown in Figures 8A and 9A and B when the hairs were treated enzymatically to remove most of the outer substances from the fibers. The M is thus regarded as the tubular substance which is doubly covered with the primary and secondary M-surrounding cells as depicted in Figure 6. We tentatively speculate that the M (tubular) wall is originated from the primary M-surrounding cells. Remarkably, many drum-shaped vesicles were arranged in a series inside the tube, and most of the vesicles were densely filled

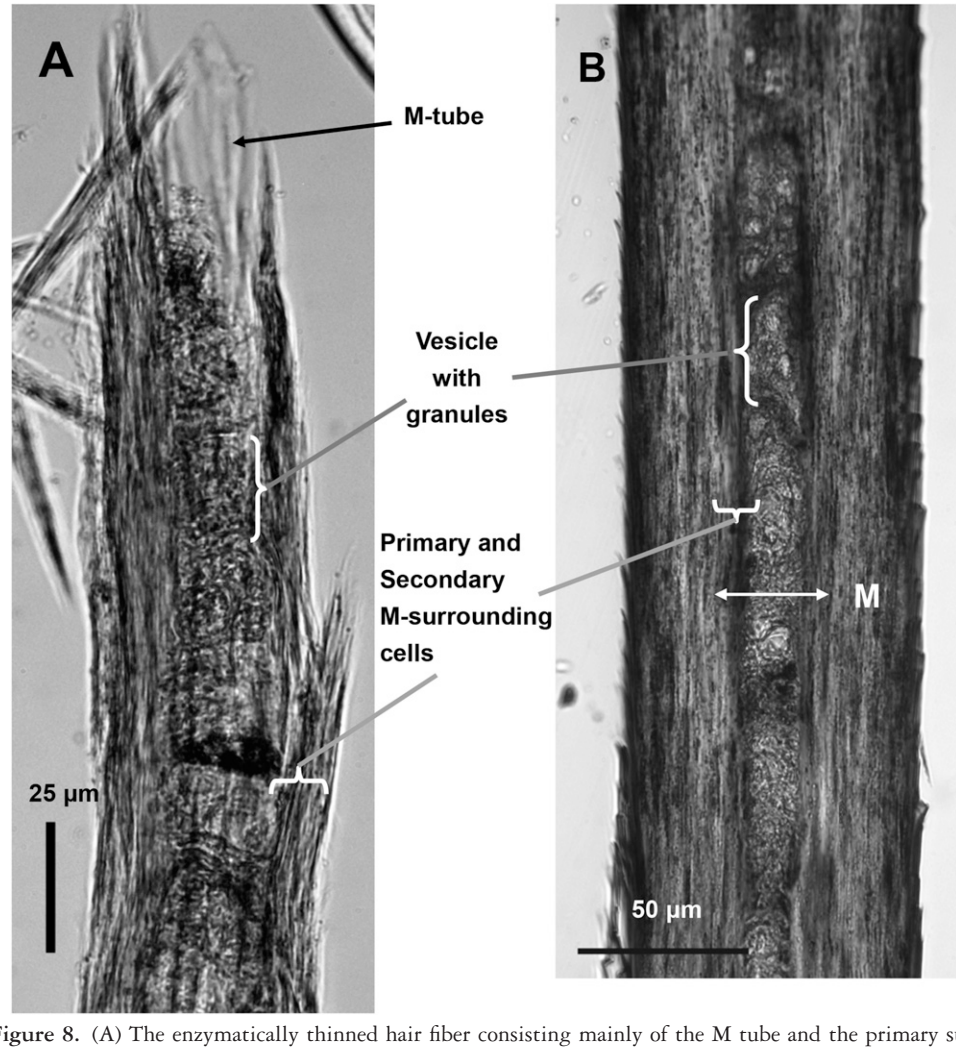

Figure 8. (A) The enzymatically thinned hair fiber consisting mainly of the M tube and the primary surrounding cells. The hair fibers (cf16) were processed with papain in pH 7/PBS in a manner similar to that described in the procedure "Random mechanical cutting of enzymatically thinned hair fibers." The resulting substance, which corresponded to that seen in the panel E of Figure 4, was washed with water, cut by means of the poly (ethylene) blades, and stained with Giemsa solution. (B) The cross-sectional specimen of the hair fiber (cf16; about 8 µm in thickness). The specimen was prepared by cutting along its longitudinal axis; gentian violet staining; see the procedure "Random mechanical cutting of enzymatically thinned hair fibers." The primary M-surrounding cell is not clearly distinguishable from the secondary one in this section of the fiber shaft. M refers to the medulla.

with the small granules (0.5–1 μm in diameter) which was strongly stainable with the protein-specific dyes such as Coomassie Brilliant Blue (CBB) and gentian violet; see Figure 9. By contrast, the tube of elderly persons was sparsely populated with the vesicles and some of the vesicles were loosely packed with the granules as typically seen in Figure 9B. It would be worth noting here that the hairs of a baby or an infant were generally very thin and

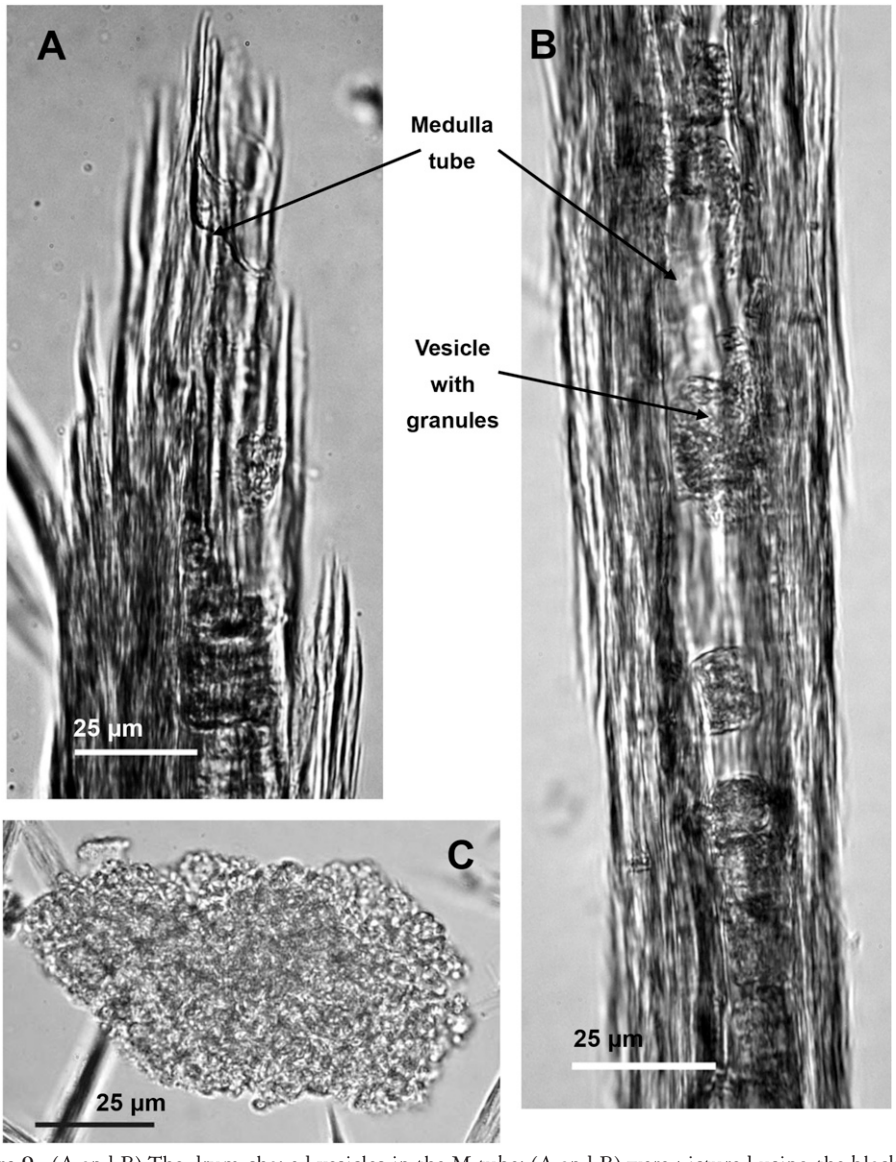

Figure 9. (A and B) The drum-shaped vesicles in the M tube; (A and B) were pictured using the black hair (cf16) and the white hair (jm67), respectively. (C) The amorphous cluster of the granules which came ou from the vesicles of the white hair (jf65). The hair samples (3–5 mm length) were warmed at 30°C in a pH 7/PBS solution of papain (about 20 units) for 3–5 h, then in the fresh enzyme solution for another 30 min. The digested hair fibers were randomly cut by means of the rapidly rotating poly(ethylene) cutter, and then centrifuged to collect the hair fragments; Giemsa staining.

the M was premature or not well developed; thereby, the inner part of their fibers, which was surrounded by the cuticular region, often appears to be packed homogeneously with the cortical cells. Accordingly, the overall look of the human hairs varies from sample to sample, especially with age and race, and has indeed been interpreted variously (13,14).

MATERIAL FLOW PROPERTY OF THE MEDULLA

The cuticular cell region of the human hair functions to protect the fiber against foreign substances and physical shocks such as heat action and mechanical friction. The cortical cell region is deformable, making it possible for hair to take an eccentric cross section and have a wavy line-shape (11,16). Although the biological function of the medulla has not been well understood, the hair component was characterized by the tubular structure as explained previously. Hence, we further investigated the possibility of the material flow through the Asian hairs to obtain the following results.

(i) The relatively large molecules such as bromocresol green (BG), Congo red, and phenolphthalein were very difficult to penetrate the cuticular cell region; cf. Figure 1. The dye molecules, however, entered the hair fiber from the medulla ends. For instance, BG traveled about 30 μm (about one-third of the hair width) in 30 min at ambient temperature from the terminal of the fiber (jm67) as typically demonstrated in Figure 10. (ii) The cuticular cell region worked well as a barrier for intrusion of even small ions into the hair fiber. In other word, H^+ ions and OH^- ions were capable of migrating preferentially through the medulla tube as displayed in Figure 11A–C and D and E, using Congo red and phenolphthalein as pH-indicators, respectively. The aforementioned barrier property of the cuticular cell region may be rationalized by the presence of CuP—the large homogeneous layer which encircles the fiber shaft (1); see Figure 1. (iii) The conduit property

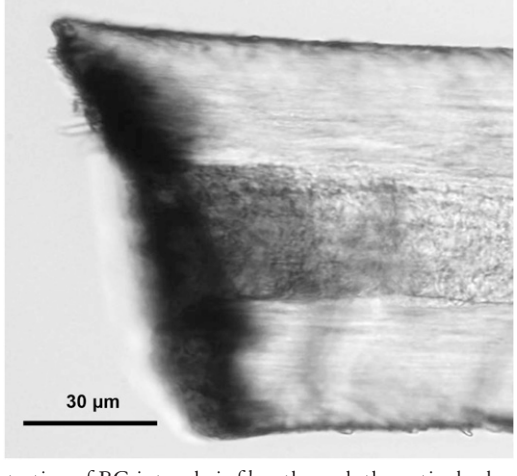

Figure 10. The slow penetration of BG into a hair fiber through the cuticular layer and the fast intrusion of the dye into the fiber from the M end. The white hair fibers (jm67) were first warmed in a mixture of 8 M urea and 4 wt.% SDS at 55°C for 2 h, then dipped in 0.5% BG solution at ambient temperature for 30 min. After brief washing with water, the hair fibers were examined with the upright microscope.

of the medulla was assisted electrically. In other words, the aqueous solution of the cationic anthocyanine dye easily flowed through the medulla in a direction from a positive electrode to a minus electrode when more than DC 500 voltage was applied between the tip and the root positions of a white hair fiber (jm67); see Figures 3 and 12. As expected, (i) the dye-flow was completely blocked and hence the medulla was not stained at all when either one of the fiber ends was capped with gelatin, and (ii) the flow was directionally reversed with the polarity of the electrodes interchanged; see Figure 12 (1–3 min vs. 3 min 40 s–5 min). Furthermore, the hair shaft itself (80 μm in diameter and 20–25% water content) showed a very high resistance of about 10^9 ohms/cm when the both ends

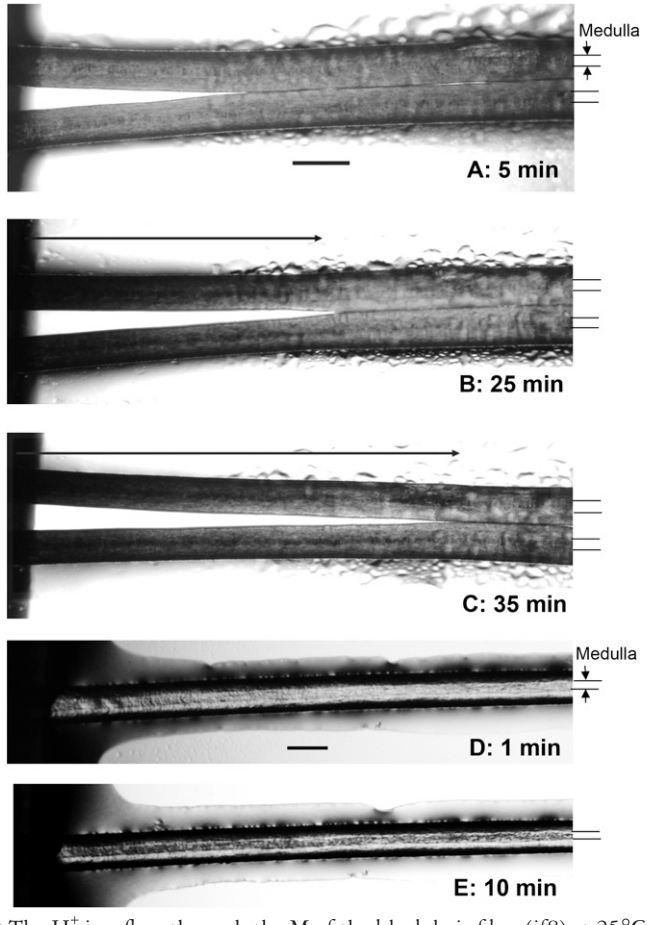

Figure 11. (A–C) The H^+ ion-flow through the M of the black hair fiber (jf8) at 25°C; bar 100 μm. The fiber was thoroughly stained with Congo red, then its left end was acidified by 0.5 M HCl; see the procedure "H^+ ion-flow through the M: The use of Congo red as an indicator" for more details. The time-lapse after acidification is printed in each picture. The M changed gradually in color from gray to reddish black toward the right hand side, as indicated by the arrow. The M tube was isotropic to the flow of H^+ ions. (D and E) The OH^- ion-flow through the M at 25°C; bar 100 μm. The white hair fibers (jm67) were stained by an aqueous solution of phenolphthalein, and then one of the two ends was alkalified by 1 M NaOH; cf. the procedure "OH^--flow through the M: the use of phenolphthalein as an indicator" of the experimental section. The color change (to red) occurred only in the M, taking less than 10 min to extend to the other end.

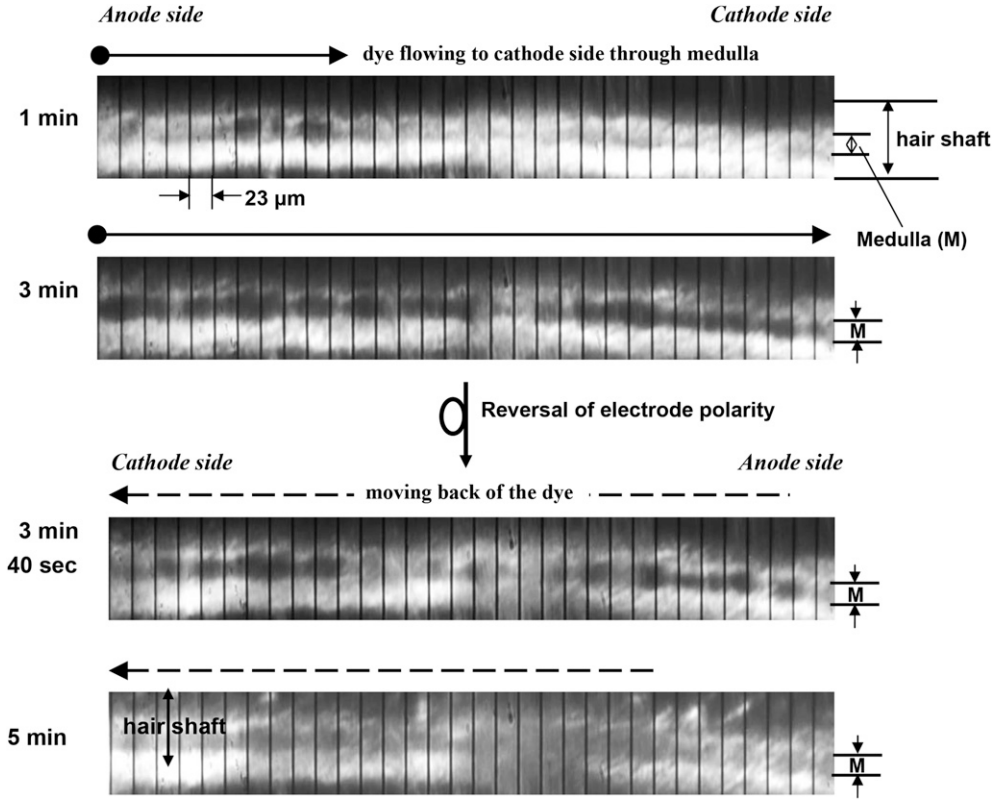

Figure 12. The DC voltage-assisted flow of the purple anthocyanin dye through the M; cf. Figure 3. One strand of the white hair (jm67) was put through the glass capillary, making one end dip into the dye solution and the other end into PBS. DC 550 voltage was then applied between the electrodes. See the procedure "Flow of the anthocyanin dye through M: Application of a DC voltage to a hair shaft" for more details. The time-lapse after application of the voltage is indicated in each of the panels. Panels of 1 and 3 min: When the buffer solution-bath was set as a minus electric side (cathode), the cationic purple dye flowed through the M, causing the length and color-density of the purple band within the M to increase with increasing time of the voltage application. Panels of 3 min 40 s and 5 min: When the polarity of the electrodes was reversed, the dye flowed back to the PBS bath to result in diminishing the purple color density in the M. The observed direct current was 23 ± 2 μA in this case.

of the hair fiber were not touching the electrode solutions. All of the aforementioned flow experiments may be taken to suggest that the medulla functions as a channel, at least in the early growing stage of the hair. The biological functions of the medulla have been under further study.

CONCLUSIONS

The medulla (M) of the Asian scalp hairs is mainly composed of the primary and secondary M-surrounding cells. The medulla is featured in its large tubular architecture (diameter, 20–30 μm). The drum-shaped vesicles, which contain many small proteinous granules, are embodied inside the tube. Various kinds of substances such as H^+-ions and dye molecules are able to flow into the hair fibers through the tube.

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