

Study of the migration of small molecules through the hair fiber to the scalp

Vargas Calle, Angela Cristina^{1*}; Zeferino, Ana Raquel ¹; Facchini, Gustavo ¹; Eberlin, Samara ¹; De Freitas Carli, Barbara ¹; Pinheiro, Adriano¹

Introduction

Human hair is a mainly keratin structure that packages itself in a highly complex way. This high complexity leaves free spaces (vacancies) that allow substances to permeate the hair fiber. The permeation is influenced by different factors including pH of the environment, hair porosity, and quality of the hair. The study of the permeation of molecules through the hair fiber to the scalp is important for the development of cosmetic products with actives that could potentially have efficacy not just on the fiber but on the scalp. On (aqui não seria In) this study, scalp explants from rhytidectomy with hair shafts of 6 cm were cut in pieces with 6mm diameter. The hair tips were submersed in an aqueous solution of fluorescent dye eosin at 3 different times. After that, the skin explants were included on TOC (o que é TOC?) compound and cut with cryotome. The cuts were analyzed by fluorescence microscopy to understand if the dye was able to migrate to the scalp. The results show migration of the fluorescent dye in all the times tested. The presence of fluorescent molecules on the skin cuts means that the actives absorbed on hair fiber are capable of achieving the hair bulb and in contact time higher than 30 min they can even reach the inner structures of the scalp.

Materials & Methods

Skin explants from elective rhytidectomy surgery, from a single donor, were cleaned and cut into circular sections measuring 6cm in diameter. Three sections were kept in culture medium and the hairs from each of the skin segments were cut to 6 cm in length. The ends of the wires (1cm) were submerged in a solution of the fluorescent dye yellow eosin (C.I 45380) with water at pH 6 (selected from previous experiments performed in hair fibers) by 20min, 30min and 1h respectively. The experimental scheme is showed on Figure 1.



Figure 1. Experimental scheme

After the exposure time, the hair fibers were removed from the fluorescent dye and the skin was included in O.C.T compound Tissue-Tek and histological sections were made on a Leica CM18670 cryostat at a temperature of -30°C, with a thickness of 8 µm and using a steel knife.

After cutting, the cell nucleus was marked with DAPI dye to facilitate visualization. The sections were then taken to an Olympus BX53 fluorescence microscope coupled to a DP74 digital camera and observations were made with a 10X objective. The images obtained were analyzed with the ImageJ 1.47v program to quantify the fluorescence intensity of the eosin dye.

To verify if there were differences between the measurements, statistical comparisons were performed on the software GraphPad Prism, version 8.3.4, using ANOVA one-way test followed by Tukey's post-test. Statistically significant differences between samples were checked with a p-value ≤0.05. (diferente espaçamento entre linhas)

Conclusions

The results of the experiments shows that the hair fiber is capable of absorb eosin dye from aqueous solution from the tip to the scalp; here 6cm fiber was tested, but the results let to think that the dye could travel even highest distances. The diffusion of eosin on hair is pH dependent, then probably the amount of dye that is transferred to the scalp could change with the pH of the hair or the solution. At this experiment the pH of the eosin solution was maintained at pH6 and no product was applied on the fiber; at this pH the hair fiber is in their isoinic point (pH 5,6 – 6,2) [6], minimizing the ionic interactions between the eosin and the hair.

Results & Discussion

Some of the images obtained from cross-sections of the scalp are shown in Figure 2. Here it is possible to observe the hair follicle structure marked in blue and some red points referent to the fluorescent dye eosin that migrates from the tip of the hair to the hair follicle.

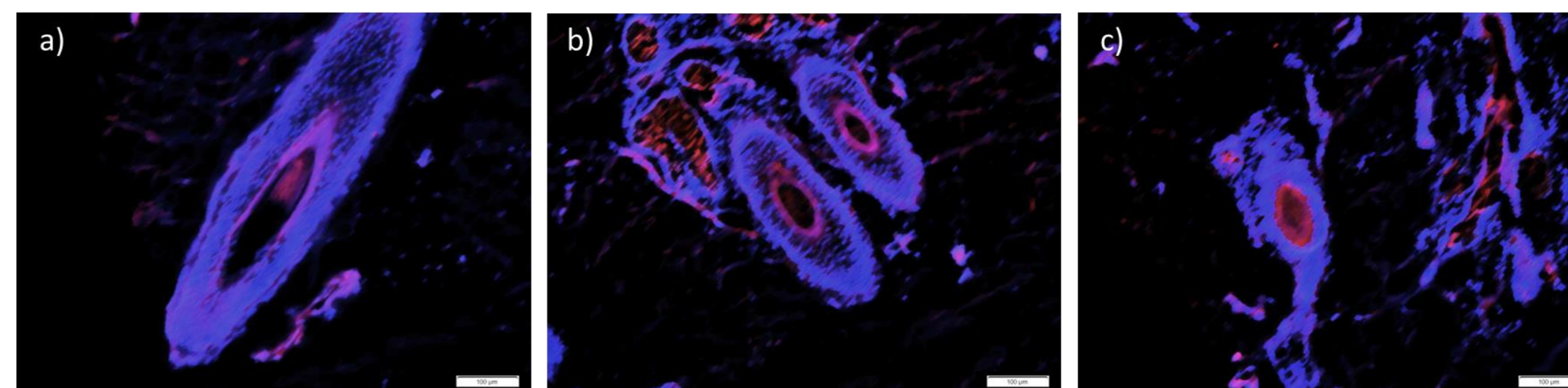


Figure 2. Scalp transversal cuts. Images of follicles (blue) after the ends of the hairs were immersed in eosin (red) dye by a) 20min b) 30min c) 60min.

Within 20min, it is possible to observe the eosin dye more located in the hair shaft, with little migration to other structures. On the other hand, the images of the follicles whose hair were exposed to eosin during 30 min showed the fluorescent dye not only on the hair shaft but, on the follicle structure. Increasing the exposure time to 60min, it is possible to visualize the eosin not only in the hair shaft and follicle, but in other structures of the skin.

The images were then analyzed with the software ImageJ, in order to quantify the fluorescence intensity between the groups; the values of maximum intensity and medium intensity were plotted. Figure 3, surmise (é essa a palavra?) the results. Same colors mean no statistically significant differences between the groups. Não entendi, 20 e 60 minutos que tem a mesma cor na coluna não teve diferença estatística? E 30 min, teve?

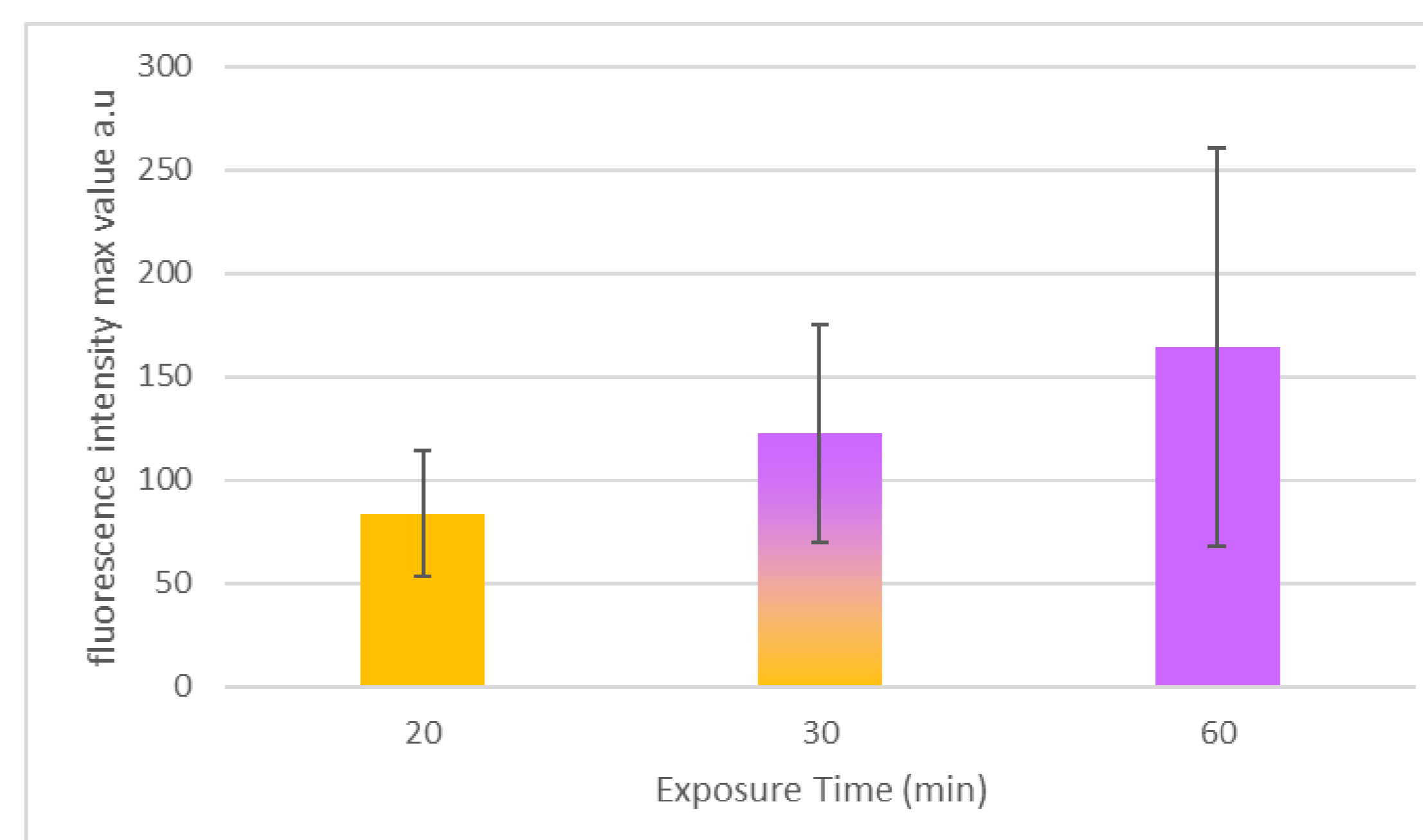


Figure 3. Maximum eosin fluorescence intensity. 10 images analyzed per group. Different colors mean statistically significant differences.

The results showed the presence of eosin dye in the scalp after exposure of the ends of the hair strands at all times studied; that is, after 20 minutes the dye had already managed to migrate to the hair follicle, confirming the hypothesis that small particles can be transported through the hair strand to the scalp. Also is possible to observe that the standard deviation increases with exposure time; this shows that the dye is more dispersed on the scalp surface with some points with high intensity (more concentrated).

References

1. Pötsch L, & Moeller M R. (1996). On pathways for small molecules into and out of human hair fibers. Journal of forensic sciences, 41(1):121–125.
2. Velasco M V R, et al. (2009). Hair fiber characteristics and methods to evaluate hair physical and mechanical properties. Brazilian Journal of Pharmaceutical Sciences, 45(1), 153–162. <https://doi.org/10.1590/S1984-82502009000100019>
3. Wortmann F. J, et al. (2023). pH-equilibration of human hair: Kinetics and pH-dependence of the partition ratios for H⁺ - and OH⁻ -ions based on a Freundlich isotherm. Biophysical chemistry, 297:107010. <https://doi.org/10.1016/j.bpc.2023.107010>
4. Barba C, et al. (2010). Water absorption/desorption of human hair and nails. Thermochimica Acta. 503:33-39. DOI 10.1016/j.tca.2010.03.004.
5. Chandrashekara M N, & Ranganathaiah C. (2009). Diffusion of permanent liquid dye molecules in human hair investigated by positron lifetime spectroscopy. Colloids and surfaces. B, Biointerfaces, 69(1):129–134. <https://doi.org/10.1016/j.colsurfb.2008.11.014>
6. Han S K, Kamath Y K, & Weigmann H D. (1985). Diffusion of semipermanent dyestuffs in human hair. Journal of the society of cosmetic chemists, 36:269-278