

Confocal in-vivo technology to measure horny layer thickness

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Introduction:

The outermost layer of the skin, the stratum corneum (SC), plays a key role in mechanical and chemical skin protection and serves as the main barrier to water loss of the human body. SC thickness measurements are important to characterize barrier function and for localization of topically applied cosmetics products and drugs in the skin in-vivo.



Figure 1: Confocal Raman spectroscopy (CRS) and Line-field optical confocal tomography (LC-OCT) measurements.

Objectives:

Aim of this project was to compare SC thickness results assessed by confocal Raman spectroscopy (CRS), compared with those obtained in parallel by line-field confocal optical coherence tomography (LC-OCT) measurements. Determination of SC thickness by CRS was validated and proven to be comparable to confocal laser scanning microscopy measurements before, but with devices showing a lower spatial resolution than the LC-OCT and the new more advanced Raman spectroscopy device [1, 3]. Therefore, our aim was to check the previous Raman-results.

Materials & Methods:

LC-OCT: Three 3D images were assessed (lateral, 1.2 mm x 0.5 mm with a depth of about 150 μm ; resolution 1.3 μm). The horizontal profiles were used to determine the SC thickness in each image block.

CRS: A confocal Raman system ("gene2-SCA Ultimate") was used to record the Raman signal in the high wave number range of the Raman spectrum (2600–3800 cm^{-1}). (laser wavelength: 671 nm; pinhole 25 μm ; integration time: 1 second; depth up to 40 μm ; resolution 3 μm). The obtained water profiles were used to calculate the SC thickness [3].

Panel: female and male participants with healthy skin; 18 - 65 years of age

Test area: Volar forearms (2 x 2 cm), identical for both measurement methods.

Results:

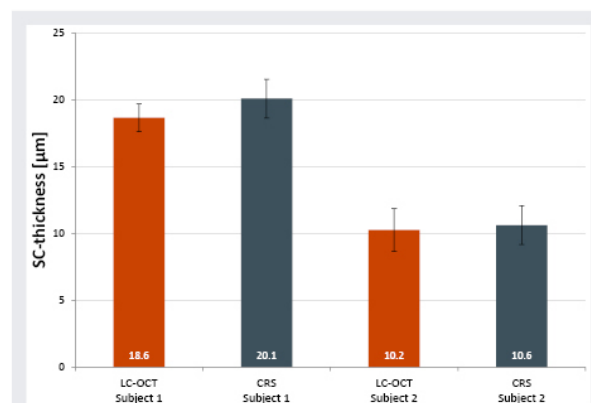


Figure 2: Stratum Corneum thickness (mean values and 95% confidence interval) on volar forearms assessed by LC-OCT and CRS for 2 male participants; paired t-test: not significant differences were found ($p = 0.107$ for Subject 1; $p = 0.690$ for Subject 2).

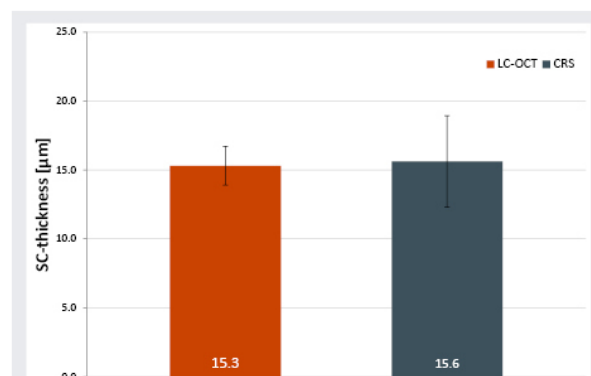


Figure 3: Stratum Corneum thickness (mean values and standard deviation) on volar forearms of female participants assessed by LC-OCT and CRS ($n = 96$ for both panels); t-test (group comparison), p -value = 0.390 (not significant).

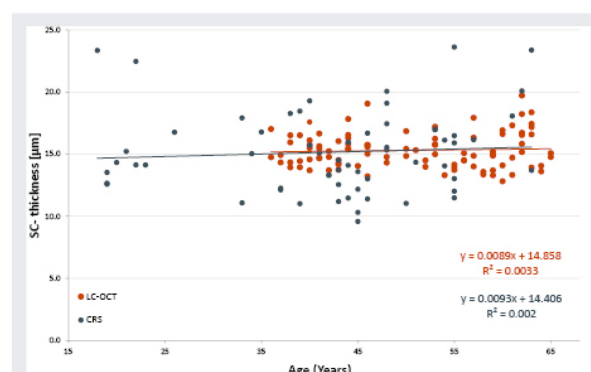


Figure 4: Stratum corneum thickness on volar forearms for female participants assessed by LC-OCT (orange color) and CRS (grey color) drawn over the subjects age ($n = 96$ for both panels). For both methods an almost identical line equation and no age correlation (R^2) of SC thickness was detected.

Conclusions:

- SC thickness assessed with LC-OCT and CRS revealed comparable results (no significant differences) for the comparison on the same two subjects and averaged over 2 different panels with a large sample size of 96 subjects.
- CRS shows a higher variation. This might be caused by the slightly lower spatial resolution of CRS.
- For SC thickness no correlation was observed with age for both methods
- For LC-OCT and CRS, SC thickness was found to be thinner on volar forearms (approx. 15 μm) compared to the findings of Böhling et al (approx. 19 μm [3]).
- Our new findings are in line with recent publications of SC thickness on volar forearm with LC OCT [2]. The findings of Böhling [3] can be explained best by the lower spatial resolution of the devices used of approx. 5 μm .
- **We conclude that the SC thickness on human adult volar forearm skin (18 to 65 years) is 15 μm at average and independent of age.**

References:

- (1) Ogien J, Levecq O, Azimani H, Dubois A. Dual-mode line-field confocal optical coherence tomography for ultrahigh-resolution vertical and horizontal section imaging of human skin in vivo. *Biomed Opt Express*. 2020 Feb 10;11(3):1327-1335
- (2) Monnier, J. et al. In vivo characterization of healthy human skin with a novel, non-invasive imaging technique: Line-field confocal optical coherence tomography. *J. Eur. Acad. Dermatol.* 34, 2914–2921 (2020).
- (3) Böhling, A., Bielfeldt, S., Himmelmann, A., Keskin, M. & Wilhelm, K.-P. Comparison of the stratum corneum thickness measured in vivo with confocal Raman spectroscopy and confocal reflectance microscopy. *Skin Res. Technol.* 20, 50–57 (2014).