Applying Raman Spectroscopy in Anti-Age and Scalp Research



Stephan Bielfeldt | 27.9.2023



- Confocal Raman Spectroscopy in vivo; measuring molecules in the living skin
- There are many applications to be measured best on forearm
- Shampoo effects should be measured on the scalp
- A harsh detergent must not extract a lot of MNFs and lipids from the scalp
- Collagen depletion can be measured in the dermis by Raman water profiles
- Take home messages



# In Vivo Measurement of Raman Spectra at Different Skin Depths with Confocal Laser Technology



# Characteristics of the Raman Device (2nd Generation)

- "gene2-SCA Ultimate" manufactured by RiverD International B. V., Rotterdam, Netherlands
- Two built-in lasers (671 nm and 785 nm)
- Fingerprint (FP): 400 1800 cm-1
- High Wave Number (HWN): 2500 3800 cm-1
- Movable table in two dimensions
- Adjustable pinholes: 25, 50 and 100 μm
- Resolution: 3, 5 and 10 µm
- One Fingerprint spectrum takes 5 sec.
- One high wave number spectrum takes 1 sec.





- Confocal Raman Spectroscopy in vivo; measuring molecules in the living skin
- There are many applications to be measured best on forearm
- Shampoo effects should be measured on the scalp
- A harsh detergent must not extract a lot of MNFs and lipids from the scalp
- Collagen depletion can be measured in the dermis by Raman water profiles
- Take home messages



### Water Measurement with Confocal Raman Spectroscopy High Wave Number Spectra of the Skin



Incident Laser Light of 671 nm



### Water Measurement with Confocal Raman Spectroscopy Assessment of Profiles Across SC and Viable Epidermis



Water profile of 1 subject on volar forearm (10 repetitions)

SGS proderm

7

#### Water Profiles: Example of Moisturizer and Emollient Effects Two observations: Increase of water gradient and SC swelling (volar forearm)



#### Penetration of Caffeine 2% into Stratum Corneum Application on 3 areas (volar forearm) for 1, 2, 3 hours, n = 3



9

#### Permeation (Depletion) of Caffeine 2% into Viable Epidermis Application on 1 area (volar forearm) measured after 1, 2 and 3 hours, n = 3



In water

In water + penetration enhancer



- Confocal Raman Spectroscopy in vivo; measuring molecules in the living skin
- There are many applications to be measured best on forearm
- Shampoo effects should be measured on the scalp
- A harsh detergent must not extract a lot of MNFs and lipids from the scalp
- Collagen depletion can be measured in the dermis by Raman water profiles
- Take home messages



### Washout of NMF and Lipids from Scalp by Shampooing



Shampooing on a test area

Head holder

Scalp measurements



# Which Skin Components Contribute to the Raman Spectrum in the Stratum Corneum?

- Keratin (ca. 70% of dry wt.)
- Water (20-70% of total wt.)
- NMF (20-30% of dry wt.)
  - Free Amino Acids 40%
  - PCA 12%
  - Lactate 12%
  - Urea 7%
- Lipids
  - Cholesterol
  - Ceramides / Free Fatty Acids



Caspers, P.J. In vivo skin characterization by confocal Raman microspectroscopy. (2003)



#### From Filaggrin to Natural Moisturization Factor (NMF) Right side: The typical distribution of total NMF in SC as measured with CRS



## Raman Fingerprint Spectrum of Skin: Video of a Measurement





### Raman Fingerprint Spectrum of Skin: A Multitude of Moisturizer Molecules Quantified from one Measurement

Single Fingerprint spectra of main skin components





Raman Shift [cm<sup>-1</sup>]



Raman Shift [cm<sup>-1</sup>]



- Confocal Raman Spectroscopy in vivo; measuring molecules in the living skin
- There are many applications to be measured best on forearm
- Shampoo effects should be measured on the scalp
- A harsh detergent must not extract a lot of MNFs and lipids from the scalp
- Collagen depletion can be measured in the dermis by Raman water profiles
- Take home messages



#### Experiment 1: Wash Out of NMF and Lipids: n = 3; 3 Washes Washing procedure: 1 ml of 10 % LES shampoo, 1 min washing each, 30 sec rinse



Measurement at baseline and 30 min after the last washing

SGS proderm

LES = Shampoo solution with 13.4 % Lauryl Ether Sulfate

#### Experiment 2: Wash Out of Lipids from Scalp by Normal Shampooing The formulations contain both 13.4 % WAS



# Experiment 2: SDS Extracts also Less Water Soluble Molecules Like NMFs than Decyl Glucoside!



SDS = Sodium Dodecyl Sulfate



# NMF Content after Washing on the Forearm with SDS at Different Concentrations, N = 2



SDS = Sodium Dodecyl Sulfate



## Penetration of SDS 2 % and 13.4 % into Forearm Skin, N = 2



SDS = Sodium Dodecyl Sulfate





- Confocal Raman Spectroscopy in vivo; measuring molecules in the living skin
- There are many applications to be measured best on forearm
- Shampoo effects should be measured on the scalp
- A harsh detergent must not extract a lot of MNFs and lipids from the scalp
- Collagen depletion can be measured in the dermis by Raman water profiles
- Take home messages



#### Water Measurement in the Dermis to Estimate the Fiber Density On photoaged dorsal skin the water content increases with age



Sun protected inner arm skin

Sun exposed outer arm skin

Is this the loss of collagen fibers?



# LC-OCT – Imaging of Epidermis, Papillary and Reticular Dermis Resolution (x, y, z) ~ 1 $\mu$ m



#### 3D-Image; size of 1.3 x 0.5 x 0.5 mm



Assessment of 60 optical reflection curves



### Measurement of Optical Attenuation (OA) to Assess Fiber Density in the Dermis



Optical Attenuation: The negative slope of the LC-OCT signal in the upper dermis Young skin => high fibre reflection => low Optical Attenuation Aged skin => low fibre reflection => high Optical Attenuation



# Correlation of Dermal Optical Attenuation of Dorsal and Volar Dermis with Age



# Correlation of Dermal Optical Attenuation of Dorsal and Volar Dermis with Water Content and Skin Elasticity (Poster Presented on IFSCC 2023)



## Take Home Messages

A highly dosed harsh detergent must not extract a lot of MNFs and lipids from the scalp.
=> Measurements under practical conditions are recommended.

- Raman water content in the dermis is increasing with age on sun exposed skin.
- Water content is increasing because fibers are replaced by water.
- Low fiber density correlates with low skin elasticity.
- In vivo Confocal Raman Spectroscopy is a method of many talents and the portfolio of methods rapidly increases.

