SIT | Skin Investigation and Technology Hamburg GmbH

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Effects of cigarette smoke on the skin in comparison to UV radiation: Parallels and differences

- Usage of the smoke chamber model for anti-pollution claim support -

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Introduction

- The human skin is the outermost barrier and therefore an important defense system for the human body
- Continuous increase in air pollution affects the human skin
- Cigarette smoke (CS) and ultraviolet radiation (UVR) are very common environmental factors that are related to the development of various dermal diseases
- CS and UVR exposure contributes to the generation of ROS which damages the natural skin barrier and leads to the release of pro-inflammatory mediators such as IL-8 and matrix metalloproteinases (MMP-1 and MMP-3)
- UVR weakens the skin‘s immune system by affecting epidermal Langerhans cells

- Valacchi et al., Ann. N. Y. Acad. Sci. (2012) ISSN 0077-8923 (Cutaneous responses to environmental stressors)
- Puri et al., Indian J Dermatol Venereol Leprol 217; 83:415-423 (Effects of air pollution on the skin: A review)
- Bickers and Athar, Journal of Investigative Dermatology (2006) 126, 2565-2575 (Oxidative stress in the pathogenesis of skin disease)
- Clausen and Grabbe, Journal of Investigative Dermatology (2015) 135, 1218-1220 (Multifaceted contributions of epidermal Langerhans cells to cutaneous carcinogenesis)
- Achachi et al., Journal of Investigative Dermatology (2015) 135, 2058-2067 (UV radiation induces the epidermal recruitment of dendritic cells that compensate for the depletion of Langerhans cells in human skin)

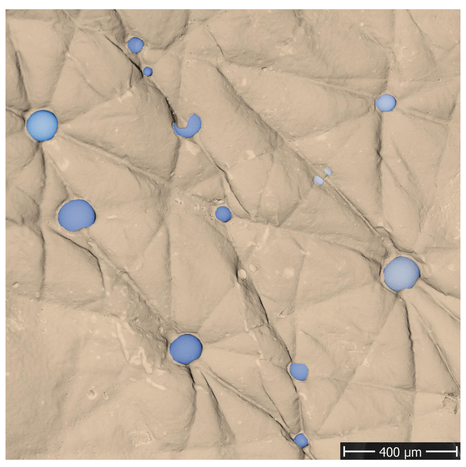
Methods

A clinical study was conducted to obtain epidermis and skin fluids from UVA-exposed as well as CS-exposed and unexposed control skin of 12 healthy male and female Caucasian volunteers

- Cigarette smoke was generated by a self-designed smoking machine
- Suction blisters were generated on the skin by using custom-made plexiglass suction chambers
  →IL-8 and MMP-1 level in the interstitial fluid
  →Analysis of Langerhans cell migration (blister roofs)
- Lipbarvis® technology (LBV)
  →Before – and after comparisons of the lipid lamellae (LBV-TEM)
  →Determination of skin lipids (LBV-LIP)
- Measurement of the TEWL values was performed with the DermaLab®

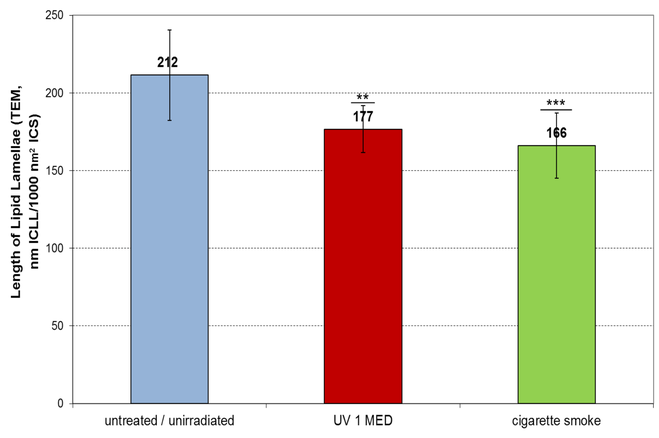
Small particles adsorb onto the skin surface during CS exposure and have an impact on the epidermal barrier lipid lamellae in the intracellular space of the stratum corneum

Scanning Electron Microscopy investigation of silicon replicas



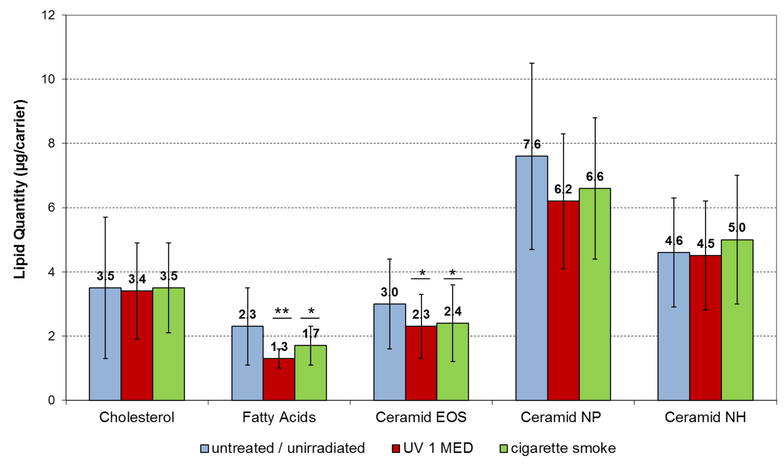
Exposure of the skin with cigarette smoke (CS) for 30 minutes. Small particles, adsorbed onto the skin surface during CS exposure, were visualized by investigation of silicon replicas via Scanning Electron Microscpy (SEM).

Investigation of epidermal barrier lipid lamellae by using the Lipbarvis® technology (LBV-TEM)



Investigation of the epidermal skin barrier 24 hours after exposure to pollutants; Skin samples in the range of 3-5 cell layers were generated by using a particularly gentle adhesive / carrier system and examined by transmission electron microscopy; length of lipid lamellae was determined and related to the intracellular space; ANOVA for repeated measures with post hoc pairwise comparisons by Fisher LSD Test, **p<0.001, ***p<0.0001.

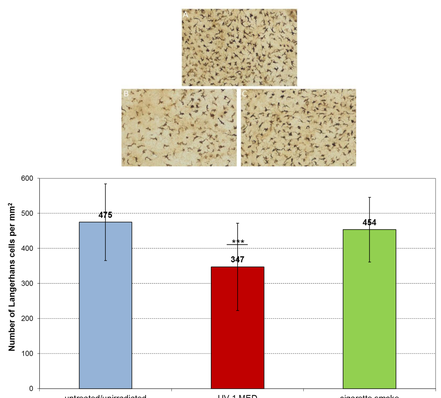
Determination of skin lipids via High Performance Thin Layer Chromatography (LBV-LIP)



Determination of skin lipids 24 hours after exposure with pollutants; Skin samples in the range of 3-5 cell layers were generated by using a particularly gentle adhesive / carrier system and analysed using the High Performance Thin Layer Chromatography (HPTLC); quantitative determination of skin lipids; ANOVA for repeated measures with post hoc pairwise comparisons by Fisher LSD Test for normally distributed data or Friedman ANOVA with pairwise comparisons by Wilcoxon Test for paired samples for not normally distributed data, *p<0.05, **p<0.001.

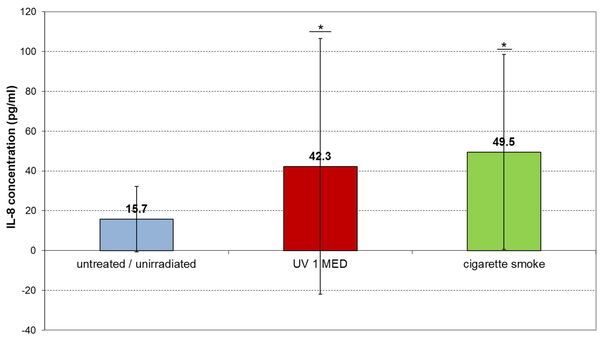
Likewise UVR, CS causes an increased release of pro-inflammatory mediators, is involved in collagen degradation and tends to have an effect on the immune system

Investigation of Langerhans cell migration using blister roofs



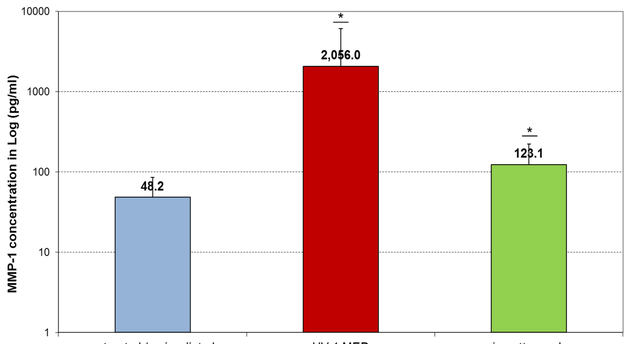
Langerhans cell staining (A-C) and total number of Langerhans cells (below) 48 hours after exposure to pollutants; ultraviolet radiation of human skin (B) and exposure of human skin to cigarette smoke (C), untreated/unirradiated skin was used as a control (A); harvest of blister roofs after 48h; analysis of Langerhans cell migration; ANOVA for repeated measures with post hoc pairwise comparisons by Fisher LSD Test, ***p<0.0001

Analysis of IL-8 via Enzyme-Linked Immunosorbent Assay



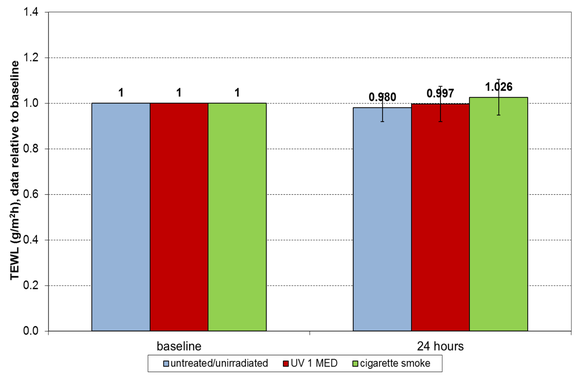
Interstitial fluid analysis of suction blisters 24 hours after exposure to pollutants; cytokine concentration of Interleukin-8 (pg/ml); Friedman ANOVA with pairwise comparisons by Wilcoxon Test for paired samples for not normally distributed data, *p<0.05.

Analysis of MMP-1 via Enzyme-Linked Immunosorbent Assay



Interstitial fluid analysis of suction blisters 24 hours after exposure to pollutants; concentration of matrix metalloproteinase-1 (pg/ml); Friedman ANOVA with pairwise comparisons by Wilcoxon Test for paired samples for not normally distributed data, *p<0.05.

Trans-Epidermal Water Loss measurement via DermaLab®



Trans-Epidermal Water Loss (TEWL) measurement at baseline and 24 hours after exposure to pollutants; TEWL was slightly increased 24 hours after exposure to pollutants; analysis of data relative to baseline.

Summary

- CS exposure and UVR lead to an epidermal Langerhans cell depletion and a significantly increased IL-8 and MMP-1 release shown by using the Suction Blister method
- CS and UVR verifiably damage the epidermal skin barrier shown by TEWL measurements (tendency) and by the Lipbarvis® technology
- The association between CS and the incidence of psoriasis could be explained by the reduction of free fatty acids and ceramides EOS and NP as well as the resulting reduction in lipid lamellae length and therefore the loss of barrier function
- Together with the smoke chamber model, the Suction Blister method with the analysis of IL-8 and MMP-1 and the LBV method are suitable to detect pollution effects and are thus suitable for anti-pollution claim support
- Taking into account the LBV results, the anti-pollution claim support could also be supplemented by methods for the determination of lipid peroxidation, such as the determination of the parameter 8-isoprostane