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ABSTRACT BOOK

Partnerships: Masaryk University, Faculty of Medicine, Brno; Institute of Health Information and Statistics; Czech Wound Management Association; Ministry of Health of the Czech Republic

EUROPEAN PRESSURE ULCER ADVISORY PANEL

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STINTS 1

Chairs: Amit Gefen, Pierre-Yves Rohan

Properties of corneocytes in the context of skin health; *Ana Evora, United Kingdom*

An optimised method to analyse inflammatory markers from sebum and its role in detecting skin damage; *Hemalatha Jayabal, United Kingdom*

A foam dressing is considerably advantageous over a hydrocolloid for prophylaxis of noninvasive ventilation mask-related-pressure ulcers; *Aleksei Orlov, Israel*

Can non-invasive skin parameters reflect changes at grade 1 pressure ulcer skin sites?; *Nkemjika Abiakam, United Kingdom*

Low-frequency ultrasound device for pressure ulcer diagnosis; *Elis MarinaSales de Castro, France*

STINTS 2

Chair: Peter Worsley

Contribution of the out-of-plane component in the assessment of sacral soft tissue deformations under compressive loading - Preliminary study on one subject; *Ekaterina Mukhina, France*

Human heel internal tissue displacements and strains calculated from Magnetic Resonance Imaging; *Alessio Trebbi, France*

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Characterization of skin integrity by quasi-static mechanical impedance device; *Yisha Chen, France*

10.1

Properties of corneocytes in the context of skin health

Ana Evora[1], *Nkemjika Abiakam*[2], *Shabira Abbas*[3], *Simon Johnson*[1], *Peter Worsley*[2], *Michael J. Adams*[1], *Dan Bader*[2]

1 University of Birmingham, School of Chemical Engineering, Birmingham, United Kingdom
2 University of Southampton, Southampton, United Kingdom
3 Essity AB, Stockholm, Sweden

Introduction: The integrity of the skin can be challenged by constant high normal and shear stresses, which may lead to skin damage in the form of pressure ulcers [1]. Most studies have focused on measures of biophysical markers [2] to assess skin health, overlooking the potential role of corneocytes in maintaining the integrity of the Stratum Corneum (SC). Indeed, these dead cells undergo an active maturation process, which includes the loss of corneodesmosomes and the stiffening of the cornified envelope (CE) [3]. This study was designed to evaluate the role of corneocytes in skin health.

Methods: A series of parallel studies have been conducted including the examination of skin response following the prolonged use of respiratory devices and exposure to pressure and moisture on two separate healthy cohorts. Corneocytes were collected via tape stripping from specific anatomical locations following each challenge. The ranked sum of the number of immature CEs (INV+) and the amount of desmoglein-1 (Dsg1) were evaluated using immunostaining techniques and correlated with the biophysical markers of skin health i.e., TEWL and SC hydration.

Results: Results revealed that the disruption of the barrier function following prolonged skin exposure to mechanical loads and moisture, as previously evidenced by increased TEWL and SC hydration [2], was correlated with both a relatively lower number of immature CEs and lower levels of Dsg1 (Fig. 1).

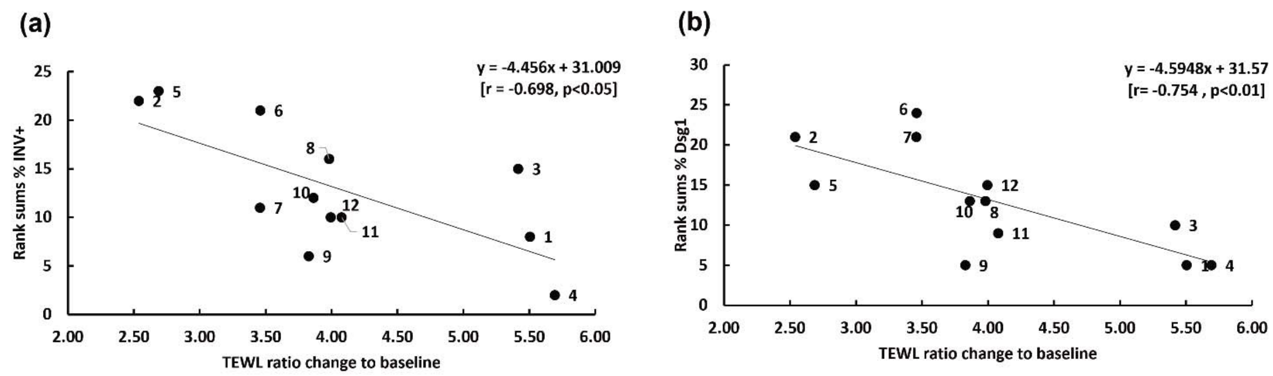

Figure 1. Relationship between the rank-sum of the percentages of (a) INV+ cells and (b) Dsg1 and the TEWL response at the sacrum after exposure to synthetic urine and loading for a total of 120 min.

Conclusions: High level of immature CEs and Dsg1 appear to provide the SC with enhanced protection against challenges from mechanical loading and moisture. This might be a direct result of these superficial cells providing a more cohesive, less easy to detach layer. However, additional evidence is required to correlate the properties of corneocytes with subject specific response to insults.

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10.2

AN OPTMISED METHOD TO ANALYSE INFLAMMATORY MARKERS FROM SEBUM AND ITS ROLE IN DETECTING SKIN DAMAGE

Hemalatha Jayabal[1], *Nkemjika Abiakam*[1], *Peter Worsley*[1], *Dan Bader*[1]

1 University of Southampton, Southampton, United Kingdom

Introduction: Inflammatory biomarkers are present in varying concentrations in a range of biofluids, with an important signaling role to maintain homeostasis. Commercial tapes have been employed to non-invasively collect these biomarkers in sebum from the skin surface to examine their concentrations in various conditions such as acne, dermatitis and pressure ulcers (PU) [1]. However, the identification of robust biomarker candidates is limited by the low abundance of specific proteins [2]. Therefore, this study aimed at developing an optimized extraction method of protein markers from skin surface and test this on a range of skin damage models.

Methods: A systematic study of chemical and mechanical approaches to optimized protein extraction were conducted employing pre-coated commercial types with synthetic sebum model. The extraction efficiency of a panel of relevant cytokines was assessed. The optimized approach will then be tested on a range of skin insult models including pressure, moisture induced damage and patients presenting with category 1 PU.

Results: The results revealed that the use of surfactant, i.e. β-dodecyl maltoside in addition to the mechanical stimuli, namely sonication and centrifugation resulted in an increased recovery of cytokines, ranging up to 80% for high-abundant cytokines, such as IL-1α and IL-1RA, and up to 50% for low-abundance cytokines, including TNF-alpha, IL-6 and IL-8.

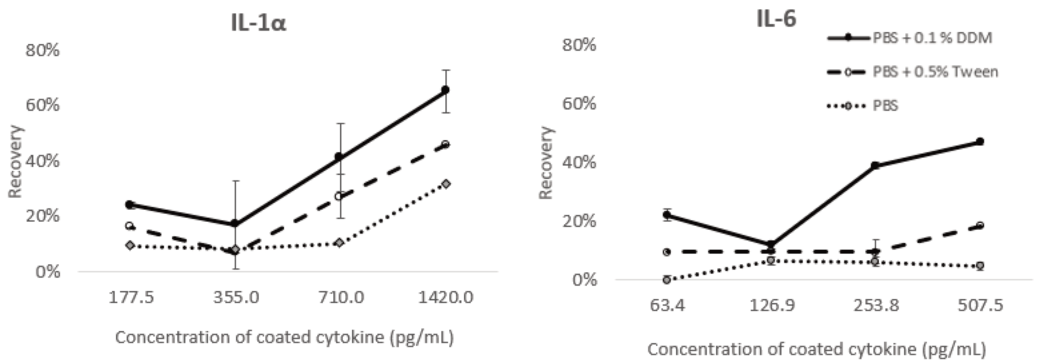

Figure 1: Percentage recovery of high-abundance (IL-1lapha) and low-abundance cytokine (IL-6) for three different extraction buffers

Conclusions: The optimized protocol will provide means to identify robust markers from skin surface that could be collected non-invasively in clinical situations involving vulnerable individuals. Indeed, the new protocol will be employed in future studies at the host laboratory involving patients with grade I PUs to identify novel predictive markers of skin health.

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10.3

A foam dressing is considerably advantageous over a hydrocolloid for prophylaxis of noninvasive ventilation mask-related-pressure ulcers

Aleksei Orlov[1], *Amit Gefen*[1]

1 Tel Aviv University, Biomedical Engeneering, Tel Aviv, Israel

Introduction: Prolonged use of continuous positive airway pressure (CPAP) masks, as often required for noninvasive ventilation during the COVID pandemic time, imposes a risk to facial soft tissue integrity and viability, as these tissues are subjected to sustained deformations caused by tightening of the stiff mask surfaces to the head. The risk of developing CPAP-related pressure ulcers/injuries (CPAP-related-PUs) can be reduced through suitable cushioning materials placed at the skin-mask interface, to spread the localised contact forces and disperse the surface and internal tissue stresses.

Methods: Using an integrated experimental-computational approach, we compared the biomechanical protective performance of a popular foam dressing material to that of a market-lead hydrocolloid dressing when applied to protect the facial skin under a CPAP mask. We measured the compressive stiffness properties of both dressing materials, and then fed those to an anatomically-realistic finite element model of the head, with an applied (simulated) CPAP mask. Through this process, we calculated the protective efficacy index (PEI) of the above materials in preventing CPAP-related-PUs, which indicates the relative contribution of the dressing type to alleviating the facial soft tissue loads with respect to the no-dressing case.

Results: We found that the greatest facial tissue stresses occur at the bridge of the nose and the cheeks, followed by the chin, which is in excellent agreement with reported clinical-epidemiological data concerning facial anatomical sites at-risk for CPAP-related-PUs. The difference in PEIs between the two material types was dramatic at the cheeks, with PEI=64% for the foam dressing with respect to a poor PEI=9% for the hydrocolloid. At the bridge of the nose that difference was lower, but still substantial, PEI=86% for the foam versus PEI=60% for the hydrocolloid. The mean PEI for the entire face was 70% for the foam dressing, and just 23% for the hydrocolloid, indicating that the foam dressing is considerably advantageous over the hydrocolloid for prophylaxis of CPAP-related-PUs.

Conclusions: The tested foam dressing demonstrated high protective efficacy at all the studied facial sites, and was considerably superior to a hydrocolloid dressing for prevention of CPAP-related PUs.

Acknowledgement: Support was received from the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No. 811965; project STINTS (Skin Tissue Integrity under Shear), and from Mölnlycke Health Care (Gothenburg, Sweden).

10.4

Can non-invasive skin parameters reflect changes at grade 1 pressure ulcer skin sites?

Nkemjika Abiakam[1], *Hemalatha Jayabal*[1], *Peter Worsley*[1], *Dan Bader*[1]

1 University of Southampton, Southampton, United Kingdom

Introduction: When the skin is exposed to prolonged mechanical forces, pressure ulcers (PUs) can occur. This is often observed in the skin of elderly individuals in acute and long-term care facilities who present with impaired mobility. Clinicians register the first signs of localised skin compromise as a category 1 PU, defined by an area of non-blanching erythema. Although there are many studies in relation to PUs, there is still a limited understanding of the temporal and spatial evolution of this condition.

Methods: A cohort of inpatients is being recruited for this longitudinal study design following ethical approval. The data from the first ten inpatients aged between 75 and 94 years old, presenting with stage 1 PU, are presented in this abstract. The PU compromised sites, either sacrum or ischial tuberosity, and a control skin site at a distance of 10 cm from the PU were assessed on two consecutive days using biophysical sensors and biochemical markers. Skin parameters were estimated involving transepidermal water loss (TEWL), Stratum Corneum (SC) hydration and inflammatory cytokines sampled from skin sebum.

Results: TEWL showed a statistically significant increase ($p < 0.001$) at the PU site compared to the healthy site on the first day of assessment (Figure 1). On day 2, the cohort presented with a similar increase in TEWL relative to the healthy site, although across the cohort individual values varied relative to the day 1 value (from -55% to 187% change). The spatial and temporal differences in skin hydration values were less significant between the two sites with values ranging from 5.8 to 83.4 AUs. Nonetheless, temporal profiles of each participant were repeatable across the assessment days.

Conclusions: Preliminary data revealed distinct temporal and spatial differences in TEWL responses between a grade 1 PU compromised site and a healthy adjacent anatomical location. Collected skin samples are to be analysed to examine whether the concentration of inflammatory biomarkers, such as IL-1α and TNF-α are related to changes evident in the biophysical parameters. Such an approach involving both biophysical parameters and biomarkers can offer the potential to identify early changes in the skin integrity of individuals at risk of developing PUs.

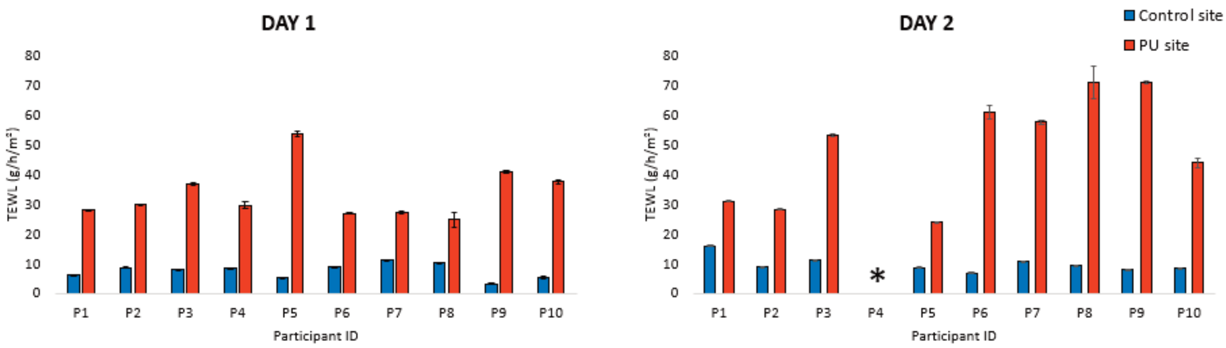

*Figure 1. Differences in TEWL responses between an anatomical PU compromise location and a 10 cm adjacent site across two days of data collection. *Missing data.*

Funding sources: This work was supported by the European Union's Horizon 2020 research and innovation programme under the Marie Skłodowska-Curie grant agreement No. 811965 (Project STINTS - Skin Tissue Integrity under Shear).

10.5

Low-frequency ultrasound device for pressure ulcer diagnosis

Elis Marina Sales de Castro[1], *Frédéric Giraud*[1], *Betty Lemaire-Semail*[1]

1 *University of Lille, L2EP, Villeneuve-d'Ascq, France*

Introduction: As a result of pressure ulcer (PU), skin mechanical properties vary [1]. In this project, a new technology for PU diagnosis is proposed, the low-frequency ultrasound (LFU) diagnostics tool. Literature indicates LFU as a complement for PU treatment, reducing pain and time for the recovery of patients [2], [3].

The LFU diagnostic tool consists of a Langevin transducer (LT) of 60kHz vibration connected to a host computer and a microcontroller. Initial results of healthy skin are presented below.

Methods: The device's set-up is presented on Figure 1. The microcontroller & LFU generation are responsible for the vibration of the LT and the assurance that the waves behave as demanded by the user. The holding structure, containing a force sensor, assures the indentation force of 0.2N during the contact. The host computer provides the interface between user and device.

The experiments taken had a ramp-like vibration over the skin, with a controlled velocity using vector control method [4]. As the vibration is the same in both contact and no-load (no contact with skin) operation, it is possible to calculate skin acoustical force with the difference of electrical effort of the device, reflected on its voltage, as shown below:

$f_r = N(v_{in\text{-}contact} - v_{no\text{-}load})$

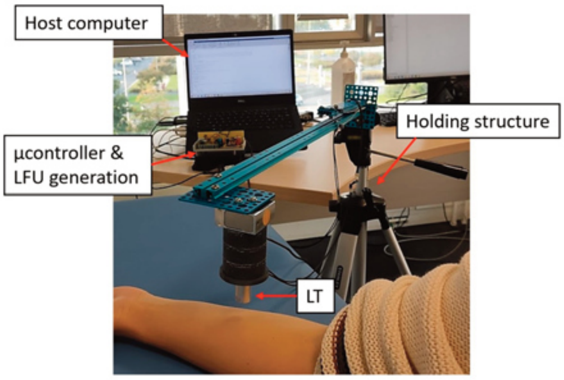

Figure 1. Set-up of the system

In the equation, $v_{in\text{-}contact}$ and $v_{no\text{-}load}$ are the voltages measured in skin tests and no-load operation, f_r is the acoustical force imposed by the skin and N is an LT intrinsic constant. Tests based on this measure were taken in 11 participants (5 female, 6 male) with ages ranging from 26 to 67 yo. Results presented in this abstract were obtained with the mechanical reaction force from skin in the morning (AM) and afternoon (PM) and for each assessment, 2 measurements were taken to guarantee the reproducibility of the measurements.

Results: The results for acoustic mechanical impedance (AMI), defined as the, $\bar{f_r}/\bar{u}$, where $\bar{u}$ is the velocity [5], are presented in Figure 2, where values of AMI range from 0.0017 to 0.0107 Ns/mm. The standard deviation within the population in different shifts is 0.0028 Ns/mm.

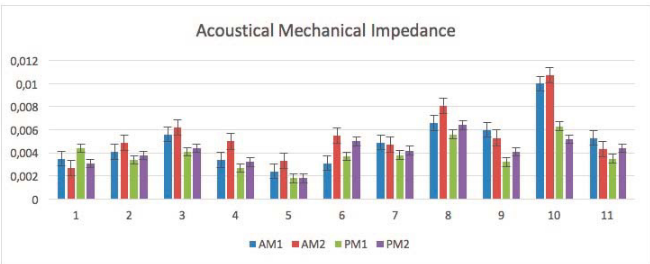

Figure 2. Results of AMI of skin

From the figure, note that the measurements in the same shift (AM or PM) present similar values.

Conclusions: The LFU diagnostic tool has shown consistent measurements for the AMI of skin. It is fair to say it can be an objective instrument to characterize skin mechanics and potentially asses early stage PU. For that, tests on damaged skin are foreseen to validate this application.

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11.1

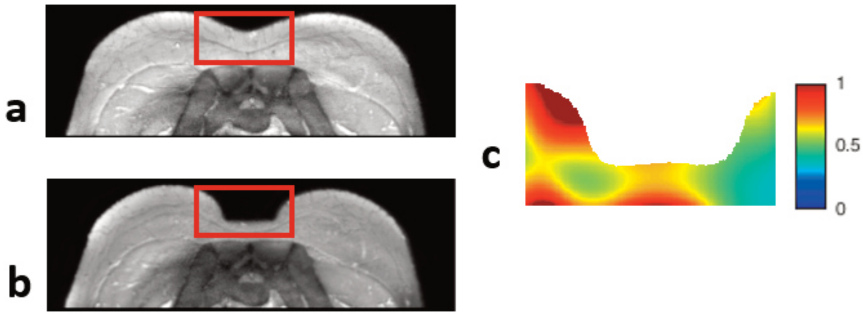
Contribution of the out-of-plane component in the assessment of sacral soft tissue deformations under compressive loading - Preliminary study on one subject.

Ekaterina Mukhina[1 2], *Pierre-Yves Rohan*[2], *Nathanael Connesson*[1], *Yohan Payan*[1]

1 *Univ. Grenoble Alpes, CNRS, Grenoble INP, TIMC-IMAG, La Tronche, France*
2 *Institut de Biomécanique Humaine Georges Charpak, Arts et Métiers ParisTech, Paris, France*

Introduction: Internal mechanical damage of the soft tissues was previously associated with the personalized risk of pressure ulcer development 1. 3D MRI is considered to be a golden standard of medical imaging for observing internal tissue deformations. 2D Ultrasound (US) images have been investigated 2 to find a more accessible alternative to MRI. However, a possible downside of using such a 2D modality is disregarding the out-of-plane tissue movements. The objective of this work was to assess the contribution of the out-of-plane component of soft tissue displacements under compressive loadings.

Methods: One healthy male volunteer (34 y.o., BMI=27.8 kg/m^2) was enrolled in the study (MAP-VS protocol N°ID RCB 2012-A00340-43). An MRI-compatible custom-made experimental setup, allowing the application of a vertical controlled load to the sacrum via an indenter, was used with different weights (0-1200 g) in a 3T MRI machine. Four load cases corresponding to the applied weight of 1200 g, 800 g, 600 g, and 400 g respectively were investigated. To evaluate the displacement fields, 3D image registrations (Elastix library) between the unloaded (Figure 1a) and loaded (Figure 1b) MRI configurations were performed. For each voxel of the vertical loading plane, the ratio of the out of plane displacement to combined in-plane displacement was evaluated (Figure 1c).



Results: The voxel-wise ratios of the out-of-plane displacement to in-plane displacement were higher than 0.5 for more than half of the voxels in the region of indentation for all investigated load cases. This ratio was also equal to or higher than 1.0 for almost half of the voxels in the region for load cases 2-4.

Conclusions: The preliminary results observed on one healthy volunteer suggest that the out-of-plane tissue displacements under compressive loads cannot be ignored. Possible next step is to investigate 3D B-mode US imaging as a way to combine the accessibility of the US technology and the advantage of three-dimension modality.

Acknowledgements: This project has received funding from the European Union's Horizon 2020 research and innovation programme under the STINTS Marie Skłodowska-Curie grant agreement No. 811965.

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11.2

Human heel internal tissue displacements and strains calculated from Magnetic Resonance Imaging

Alessio Trebbi[1], *Bethany Keenan*[2], *Mathieu Bailet*[3], *Antoine Perrier*[1], *Yohan Payan*[1]

1 *University of Grenoble, Grenoble, France*
2 *University of Cardiff, Cardiff, United Kingdom*
3 *Twinsight, Grenoble, France*

Introduction: Pressure ulcers are defined as localized areas of damaged skin and underlying soft tissues caused by sustained mechanical loads on the skin surface. They are common in the posterior heel region in bedridden patients. It is still not completely understood how external loads lead to high local internal strains and how these strains cause tissue damage. Finite Element(FE) analysis is a powerful tool to help understanding how such external loads lead to deep internal strains. However, it has been highlighted how this numerical analysis lacks proper validation(Keenan 2021). This abstract aims to describe an in vivo methodology that will be implemented for evaluating the simulations of an FE model of the human heel. This solution is based on applying various loading configurations on the heel while recording Magnetic Resonance (MR) scans.

Methods: A healthy male volunteer(aged 30 years) gave his informed consent to be scanned using a 3T MRI platform1. A T2 DESS MRI sequence with a 0.6mm isotropic voxel size was used to image the foot in a series of configurations (unloaded, loaded on hard surface, loaded on mattress, loaded with shear, and loaded with an indenter) (Figure1). The unloaded and loaded MR images were then registered using the registration toolbox2 to extract the displacement field and strain maps for the soft tissues(Trebbi 2021).

Results: The high-resolution MR acquisitions allowed a clear distinction of the tissues that compose the human heel and their displacements due to the application of the various loads(Figure2). As expected, the implementation of a mattress on the supporting surface reduced the amount of deformation and strains. Conversely, the loading configuration involving the indenter generated the highest levels of max Green Lagrange shear strain.

Figure1: MR set up with the participant's heel loaded on a soft cushion.

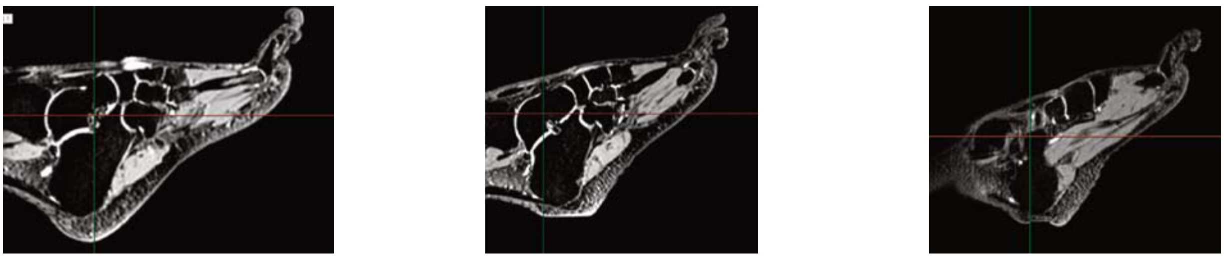

Figure2: MR images related to the configurations for A unloaded, B loaded on hard surface, C loaded with an indenter.

Conclusions: The implemented technique can give insight for several applications. First, it adds a useful tool for better understanding the propagation of deformations in the heel soft tissues that could generate pressure ulcers. Second, this procedure can be used to obtain data on the material properties of the soft tissues to define constitutive laws for FE simulations. Third, image registration offers a promising technique for evaluating FE models. Finally, these outcomes could be implemented to evaluate performances of orthotics and dressings aiming for preventing pressure injuries.

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1 3T Siemens Magnetom Prisma system

2 Registration toolbox Elastix

11.3

Molecular dynamics simulation and thermodynamical approaches to predict and model the barrier function of skin lipids

Nicola Piasentin[1], *Guoping Lian*[1], *Qiong Cai*[1]

1 University of Surrey, Chemical and process engineering, Guildford, United Kingdom

Introduction: Understanding the barrier properties of the intercellular lipids of stratum corneum (SC) is pivotal to better design of transdermal drugs and skin care products. The current state of the art is that the inter-keratinocyte space is occupied mainly by waxy acids (ceramides), free-fatty acids, and cholesterol, organized geometrically as a series of stacked bilayers in a roughly equal molar ratio1. The exposure of skin to dermatological relevant molecules can affect the lipid bilayers' features, consequently altering skin's barrier properties and structural integrity2.

Several in silico approaches have been developed to investigate the mechanisms underpinning skin barrier hallmarks. Among these, molecular dynamics (MD) simulations have been employed to predict the SC lipid properties3. Similarly, a combinations of quantum chemistry and thermodynamical calculations4 has been developed for fast barrier properties prediction. The aim of this study is to exploit both these tools to investigate and rationalize the effect of selected chemicals on SC lipid bilayer's geometrical and barrier properties.

Methods: The systems are being simulated via GROMACS with the CHARMM36 forcefield. Systems containing different concentrations of ethanol, glycerol or urea are simulated to extract their geometrical information. From this information, structural effects are quantified by measuring lipid structural parameters and the corresponding barrier properties are predicted via quantum/thermodynamical approaches.

Results: Ethanol is the molecule that majorly disturbs the lipids bilayers, inducing extraction of lipids and partitioning of ethanol molecules into the bilayers. Increasing the temperature lowers the ordering of the lipid bilayers in all cases investigated, gradually compromising the integrity in systems containing ethanol. Barrier properties are not affected by urea and glycerol, while the partitioning of ethanol eases the partitioning of other solutes through lipids.

Conclusions: Results show that ethanol disturbs the SC lipid bilayers, enhancing the partition of solutes into the lipid systems, while glycerol and urea have limited effect, suggesting that these molecules affect other SC permeation routes.

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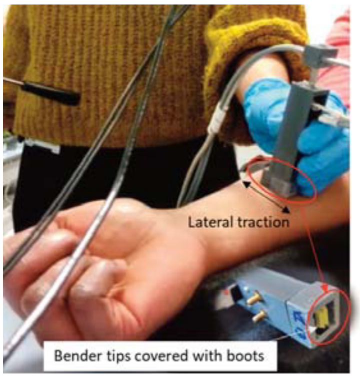
11.4

Characterization of skin integrity by quasi-static mechanical impedance device

Yisha Chen[1], *Betty Lemaire-Semail*[2], *Frédéric Giraud*[3], *Vincent Hayward*[4]

1 *Université de Lille, Villeneuve d'Ascq, France*
1 *Université de Lille, Villeneuve d'Ascq, France*
3 *Université de Lille, EEA, Villeneuve d'Ascq, France*
4 *Sorbonne Université, CNRS, Institut des Systèmes Intelligents et de Robotique, ISIR, Paris, France*

Introduction: Early detection of pressure ulcers is essential to reduce treatment costs. Biomechanical characterization of soft tissue/ skin has received increased interest [1]–[3], as they may reveal dysfunction or underlying damage. To better understand how skin biomechanics are related to the risk of pressure ulcers, we developed a portable device to characterise the quasi-static viscoelasticity of skin. To justify the feasibility of the device as a diagnostic tool, we damaged the skin with tape stripping to simulate the presence of a pressure ulcer.



Methods: To study the influence of tape stripping on skin biomechanics, 8 healthy participants (3 males, 5 females), with an average age of 34 years (27-41 years) were recruited. Skin biomechanics of the forearm were characterized using the quasi-static mechanical impedance device (Fig. 1). The device was composed of a pair of piezoelectric benders, which can deform skin laterally.

To characterize quasi-static viscoelasticity of the skin, a loading pattern of step-hold-sinusoidal was applied [4]. The skin was first stretched laterally to a baseline strain of 4.2% and held for 10 s. Then, a sinusoidal displacement was applied to the skin for 10 periods at 1 Hz, with a strain amplitude of 0.8%. Skin responses before, after insulting (tape stripping 25 times), and after 30 min of recovery were measured.

Fig. 1 Skin measurements with the quasi-static mechanical impedance device. The piezoelectric bender tips were insulated by a pair of boots.

Results: Dynamic modulus analysis was employed to deduce skin parameters. The median complex modulus of the forearm is 165 kPa (78-452 kPa). The median loss tangent value is 0.29 (0.23- 0.49).

To study the changes in biomechanics due to insulting, all the data were normalized by their baseline values. As shown in Fig. 2, tape stripping tends to lower the complex modulus, except for participant 6. While loss tangent is increased after, except for participant 8. A recovery time of 30 minutes is insufficient for most of the participants.

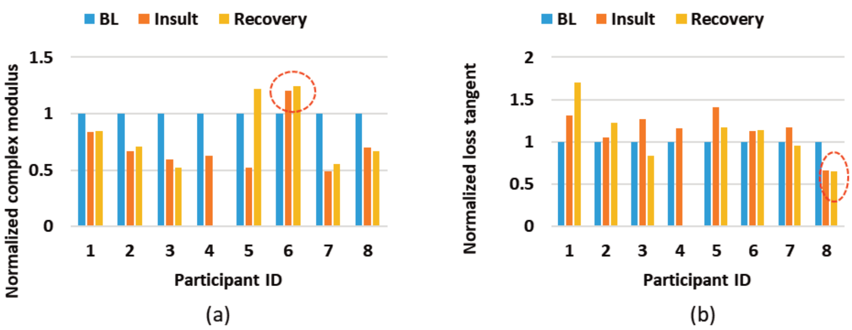


Fig. 2 Dynamic modulus analysis. (a) Normalized complex modulus. (b) Normalized loss tangent. The blue bar ("BL") represents baseline value, the orange bar ("Insult") represents data measured after tape stripping, and the yellow bar ("Recovery") represents data measured after 30 min of recovery time. Data were normalized by the baseline values. Recovery data of participant 4 are missing.

Conclusions: Preliminary results show that the quasi-static mechanical impedance device can detect the skin changes after tape stripping, with a decreased complex modulus and an increased loss tangent. Tests on a larger group of participants are required to confirm this conclusion.

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