

Dr. Kenneth Burnell Student Research Award 2014-2015

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Effects of Heat-Induced Skin Blood Flow Changes on Skin Water Parameters

Background

Prior research¹ has investigated the relationship between skin heat-induced vasodilation and the sympathetic nervous system. It was found that despite chemically induced nerve conduction blocks to the axons of the sympathetic nerves supplying forearm skin, local heating continued to produce a vasodilation effect. This showed that withdrawal of sympathetic tone was only one mechanism involved. Other work has demonstrated that there are in fact at least two mechanisms contributing to the rise in skin blood flow during non-painful heating; a fast responding vasodilation mediated by the axon reflexes and a slower response relying on the production of nitric oxide (NO).²

This dual mechanism became evident in experiments in which the action potential conduction of the axon was blocked using an EMLA cream and NO synthase (NOS) inhibition was achieved with L-NAME. It was shown that the role of NO was about 1.75 times greater than the role of the sympathetics. Further examining the sympathetic response aspect of this theory, Charkoudien et al³ found that despite undergoing a complete regional sympathectomy for treatment of palmar hyperhidrosis, patients continued to display substantial heat-induced vasodilation. Furthermore, it has been found that NO is involved in sustaining the hyperemic response after heating, such that removal of NO by L-NAME causes a faster fall in blood flow after 30 minutes post heating.⁴ Because NO is involved with changes in vascular permeability as well as vasodilation we believe that there is a direct relationship between the amount of tissue water leaked into the interstitium and the amount of blood flow caused by local heating.

Hypothesis

We hypothesize that skin blood vessel vasodilation is associated with an increase in the amount of fluid filtered from the capillaries distal to the arteriolar vasodilation and that this subsequent increase in interstitial fluid is dependent on the amount of the blood flow increase.

Purpose

Our purpose is to test this hypothesis since the process being investigated impacts our basic understanding of skin-related physiology and has potential extensions to skin-related clinical issues. Moreover, because other research has shown that gender differences exist in the NO activity⁴, blood flow and skin water, it is important to study these issues in both genders as is presently planned.

Subjects

A total of 70 subjects will be recruited for participation in this research study. Measurements will be done in both sexes (35 male and 35 female with age-range of 18-35 years) since the literature indicates gender differences in skin blood flow and skin water content. Recruitment of subjects will be done by the co-investigators who will advertise by word of mouth within the University and by use of a flier to recruit volunteer subjects from University staff, faculty and students. Potential subjects will meet with a co-investigator, who will explain the study in detail, administer the consent form, and the research experimental measurements to be described will be done at NSU.

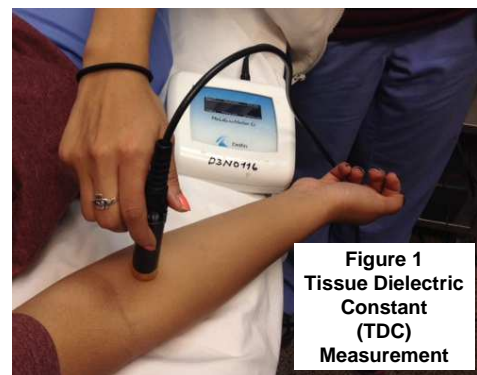
Experimental Methods, Procedures and Devices

Overview

Skin blood flow will be measured via a laser Doppler method before, during and after local heating (section A4). Skin-to-fat water content will be measured before and after heating using the method of tissue dielectric constant (TDC) as described in section A1. Stratum corneum water will be measured using electrical capacitance methods (section A2) and skin water loss will be measured by the method of transepidermal water loss (section A3). Skin temperatures will be measured with an infrared non-contact thermometer.

A1. Local Tissue Water via Tissue Dielectric Constant Method

The method is based on the principle that the tissue dielectric constant (TDC) is directly related to the amount of free and bound water contained in the measuring volume⁵⁻¹². The TDC of specific target areas is determined using a coaxial probe that makes contact with the skin for



about 10 seconds. The probe, which is connected to a control and display device, measures the TDC at a frequency of 300 MHz. At this frequency the TDC is an index of both free and bound water. The penetration depth of the measurement depends on the probe size, with larger diameter probes penetrating deeper. The output parameter is the TDC value that has a range of 1 to 80. For reference, pure water has a value of about 78.5. For this study probes with effective penetration depths of about 1.5 and 2.5 mm will be used (Fig 1). These measurements will be made on dominant arm anterior forearm site. The device to be used is the battery operated Moisture MeterD (Delfin Technologies Ltd. Kuopio FINLAND).

A2. Method for Measurement of Stratum Corneum Hydration

The method is based on the principle that the measured electrical capacitance between the skin surface to a depth of about 100 μm is an index of the relative water content of the stratum corneum (SC). The device to be used for this measurement in the present study is the hand held battery operated MoistureMeter SC also manufactured by Delfin Technologies Ltd and pictured adjacent. A measurement is taken by touching the skin surface with the end of the probe for about 10 seconds and the relative SC moisture is displayed on the device meter.

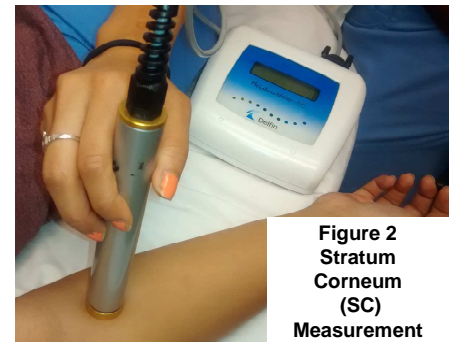


Figure 2
Stratum
Corneum
(SC)
Measurement

A3. Method for Measurement of Transepidermal Water Loss (TEWL)

This method is based on the principle that the flux ($\text{ml}/\text{m}^2/\text{min}$) of water leaving the skin can be quantified by collecting the effluent within a closed chamber and determining the change in relative humidity within the closed chamber. The device to be used for this purpose is the hand-held battery operated VapoMeter also manufactured by Delfin Technologies Ltd. The measurement procedure requires the touching of the skin with the tip of the device for about 10 seconds after which the TEWL value is automatically displayed.

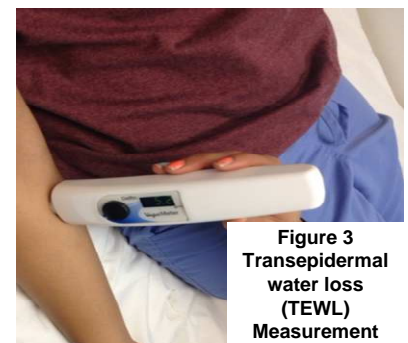
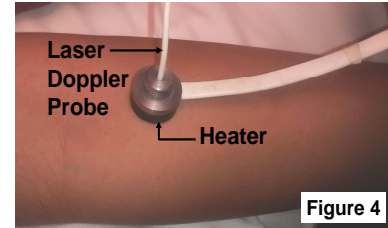


Figure 3
Transepidermal
water loss
(TEWL)
Measurement

A4. Method for Measuring Skin Blood Flow

Skin blood flow (SBF) will be measured by a laser-Doppler flowmetry system (Vasamedics BPM2, Blood Perfusion Monitor). With this method, the flux of red blood cells is detected by a small sensor taped to the skin. The sensor transmits a very low level laser light and also recovers the reflected signal that has a change in wavelength proportional to skin blood flow. The sensor is connected to a laser-Doppler monitoring and processing device via a fiber optic lead that converts the signal to blood perfusion units.



This measurement method is in standard use clinically and for research purposes and its use for various purposes has been documented in over a thousand published papers. The principal Investigator has used this technique for over fifteen years and the device is FDA 510K registered. The fiberoptic probe is inserted directly through the concentric heating element and taped to the skin as shown in Figure 4.

Protocol

After explaining the study to the subject, the consent form will be signed, and the subject will be asked if they wish to use the restroom before the study begins. Next, the subject will be asked to lie supine on an examination table and a light blanket will be draped across their body, excepting their dominant arm and head. While the subject is lying supine, a target site for the subsequent TDC, SC, SBF, TEWL and skin temperature measurements will be marked on the dominant forearm 5 cm below the antecubital fossa.

Phase 1

Six preheat data sets will be acquired at two minute intervals from the target site in the following order: the subject's skin temperature, SC, tissue dielectric constants for 1.5 mm and 2.5 mm beneath the skin, and TEWL.

Phase 2

At this point the blood flow sensor and heating element will be attached to the skin at the target site. SBF will be continuously recorded until the end of Phase 4 via an analog to digital data acquisition system. The heating element will heat the skin to approximately 34 °C and the blood flow will then be recorded once a minute for four minutes.

Phase 3

Next, the heating element will gradually increase heat applied to skin over approximately 1 minute, 30 seconds from 34 °C to 40 °C in single degree increments.

Phase 4

Once the 40 °C steady state has been achieved, over the next 12 minutes once at the end of each minute, the blood flow will also be manually recorded from the perfusion monitor.

Phase 5

The heating element will now be deactivated and removed from the subject's forearm with the SBF sensor. Similarly as in Phase 1, skin temperature, SC, tissue dielectric constants for 1.5 mm and 2.5 mm beneath the skin, and TEWL will be measured, in that order at two-minute intervals for 12 minutes.

Data Analysis

The hypothesized dependence of tissue water on induced blood flow will be tested by regression analysis. Gender differences in measured parameters will be tested with independent T-tests with a p-value <0.05 accepted as significant.

Dissemination

Investigators of this study plan to prepare an abstract presentation and present experimental findings at the Osteopathic and Medical Conference and Exposition in fall 2015. A full manuscript will be developed for submission to the Journal of Skin Research and Technology for publication along with the Journal of Clinical Physiology.

References

1. Charkoudian, N., J. H. Eisenach, et al. (2002). "Effects of chronic sympathectomy on locally mediated cutaneous vasodilation in humans." *Journal of Applied Physiology* 92: 685-690.
2. Gooding, K. M., M. M. Hannemann, et al. (2006). "Maximum skin hyperaemia induced by local heating: Possible mechanisms." *Journal of Vascular Research* 43: 270-277.
3. Pergola, P. E., D. L. J. Kellogg, et al. (1993). "Role of sympathetic nerves in the vascular effects of local temperature in human forearm skin. ." *American Journal of Physiology* 265(3): H785-792.
4. Gooding, K. M., M. M. Hannemann, et al. (2006). "Maximum skin hyperaemia induced by local heating: Possible mechanisms." *Journal of Vascular Research* 43: 270-277.