Skin Tattoos Alter Sweat Rate and Na⁺ Concentration

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ABSTRACT

Luetkemeier, M. J., J. M. Hanisko, and K. M. Aho. Skin Tattoos Alter Sweat Rate and Na⁺ Concentration. Med. Sci. Sports Exerc., Vol. 49, No. 7, pp. 1432–1436, 2017. The popularity of tattoos has increased tremendously in the last 10 yr particularly among athletes and military personnel. The tattooing process involves permanently depositing ink under the skin at a similar depth as eccrine sweat glands (3–5 mm). Purpose: The purpose of this study was to compare the sweat rate and sweat Na⁺ concentration of tattooed versus nontattooed skin. Methods: The participants were 10 healthy men (age = 21 ± 1 yr), all with a unilateral tattoo covering a circular area at least 5.2 cm². Sweat was stimulated by iontophoresis using agar gel disks impregnated with 0.5% pilocarpine nitrate. The nontattooed skin was located contralateral to the position of the tattooed skin. The disks used to collect sweat were composed of Tygon® tubing wound into a spiral so that the sweat was pulled into the tubing by capillary action. The sweat rate was determined by weighing the disk before and after sweat collection. The sweat Na⁺ concentration was determined by flame photometry. Results: The mean sweat rate from tattooed skin was significantly less than nontattooed skin (0.18 ± 0.15 vs 0.35 ± 0.25 mg cm⁻² min⁻¹; P = 0.001). All 10 participants generated less sweat from tattooed skin than nontattooed skin and the effect size was −0.79. The mean sweat Na⁺ concentration from tattooed skin was significantly higher than nontattooed skin (69.1 ± 28.9 vs 42.6 ± 15.2 mmol L⁻¹; P = 0.02). Nine of 10 participants had higher sweat Na⁺ concentration from tattooed skin than nontattooed skin, and the effect size was 1.01. Conclusions: Tattooed skin generated less sweat and a higher Na⁺ concentration than nontattooed skin when stimulated by pilocarpine iontophoresis. Key Words: PILOCARPINE, IONTOPHORESIS, HYPOHIDROSIS, DERMIS, ECCRINE GLAND

The practice of tattooing dates back to ancient civilizations but its popularity has escalated in recent times. A report by the Pew Research Center for the People and the Press, estimated that 45 million Americans had at least one tattoo (14). Furthermore, the report indicated 40% of young adults between the ages of 26 and 40 yr have one or more tattoos compared with 10% of older adults. Tattoos are particularly popular with college and professional athletes. One anecdotal report indicated that 53% of all NBA basketball players during the 2015 to 2016 season were tattooed and that two NBA teams, the Cleveland Cavaliers and the Houston Rockets, had prevalence rates of 73% (10). Military personnel are another population with a high incidence of tattooed individuals. A survey of 1835 U.S. basic recruits and advanced training personnel indicated that 36% were already tattooed and 48% were very serious about getting a tattoo (1).

The tattooing process involves puncturing the skin with a cluster of needles at a frequency of 50 to 3000 punctures per minute. The needles are loaded with dye that is deposited 3 to 5 mm below the surface of the skin into the dermal layer. The punctures initiate an inflammatory response that includes the attraction of neutrophils and monocytes to the injured skin. Monocytes transition into macrophages and engulf some of the dye transporting it via the lymphatic system to lymph nodes. Other macrophages laden with dye remain in the dermis at the tattoo site, and together with dye filled dermal fibroblasts and unincorporated dye particles, form the basis of the permanent color (12).

The dermal layer of the skin is composed of collagen fibers, nerves, blood vessels, and glands including the eccrine sweat glands used to produce sweat for evaporative heat loss when internal heat generation exceeds thermoregulation by other means (15,16). Eccrine sweat glands actively draw fluid from the extracellular fluid into secretory coils located at the base of the glands. Eccrine sweat glands are stimulated primarily by sympathetic cholinergic nerve fibers using the neurotransmitter acetylcholine. Because the isosmotic primary sweat is extruded through the duct of the gland toward the skin surface, sodium chloride is reabsorbed causing the sweat Na⁺ concentration to diminish (5).

Because of the close proximity of the eccrine sweat glands to the location where dye permanently resides after obtaining a tattoo, it is reasonable to question whether possessing a tattoo interferes with the basic functions of the sweat gland; that is,
producing sweat and/or reabsorbing \( \text{Na}^+ \) from the sweat. This is an especially relevant concern for heavily tattooed athletes who exercise in the heat or military personnel who conduct training and combat operations in hot environments. Therefore, the purpose of this study was to determine whether tattooed skin altered sweat rate and sweat \( \text{Na}^+ \) concentration compared with nontattooed skin. An additional purpose of the study was to examine the relationship between the age of the tattoo and changes in sweat rate and \( \text{Na}^+ \) concentration between tattooed and nontattooed skin.

**METHODS**

The institutional review board of Alma College approved the experimental protocol used in this study and written informed consent was obtained from all volunteers before their participation in the study. The participants of the study consisted of 10 apparently healthy male volunteers, and all candidates met both parts of the following entry criteria. 1) The participant had a tattoo on one side of their body located on the upper back, shoulder, upper torso, upper arm, or lower arm that covered 100% of a circular area extending 5.2 cm\(^2\). 2) The participants had nontattooed skin on opposite side of their body (contralateral) at the same anatomical position as the tattoo. Table 1 provides a list of the height, weight, age, tattoo location, and tattoo age for each participant along with means ± SD where appropriate.

All measurements were conducted in the Human Performance Research Laboratory at Alma College where the temperature and relative humidity were controlled at 19°C to 21°C and 50% to 60%, respectively. Upon arrival at the laboratory, a coin toss randomly determined whether the tattooed skin or nontattooed skin was tested first. This procedure resulted in a 7:3 split meaning seven subjects tested their tattooed skin first and three subjects tested their nontattooed skin first. A subsequent \( t \)-test was performed on sweat rate and sweat \( \left[ \text{Na}^+ \right] \) and determined that this ordering was not significant to the final results. The area of skin with the highest density of ink was chosen to represent tattooed skin whereas the area of skin contralateral to the tattooed skin was chosen to represent nontattooed skin. The distance away from prominent anatomical landmarks, such as the acromion process, was used to help locate the contralateral position of the nontattooed skin. It is generally accepted that contralateral measures of sweat rate are interchangeable and appropriate for studies requiring a comparable control (7). The two skin sites were wiped clean with 70% isopropyl alcohol, rinsed with distilled water, and allowed to dry. Participants assumed a sitting position throughout both collection periods.

Sweat was induced using the Macroduct® Sweat Collection System, model 3700-sys by Wescor (Logan, UT) (2). During both tests, the subjects were instrumented with two 5.2-cm\(^2\) agar gel disks (Pilogel®, impregnated with 0.5% pilocarpine nitrate (17). Pilocarpine is an acetylcholine agonist used to promote sweating clinically as a diagnostic test for cystic fibrosis. The disks were attached to electrodes; one positive, one negative, that were used to deliver the pilocarpine into the skin using two 5-min cycles of iontophoresis. The electrodes containing the disks were attached the participant using Velcro straps and adhesive tape.

After the second cycle, the electrodes were removed and a sweat collection disk (5.2 cm\(^2\) was fitted over the skin surface where the positive electrode was before and sweat was collected for 20 min. The sweat collection disks were composed of Tygon® tubing wound into spiraling circles so that the sweat was drawn into the tubing by capillary action. The sweat rate was determined from the change in weight of the collection disk measured before and after sweat collection. After the sweat rate was measured, the sweat was blown out of the collector disk using a sterile syringe filled with 1 mL of distilled water. The diluted sweat samples were brought up to exactly 2.0 mL with distilled water and the sodium concentration was measured using a clinical flame photometer, model PFP7, by Jenway, Staffordshire, UK. The actual concentration of sodium in the sweat was determined by multiplying the concentration of the dilute sweat by the appropriate dilution factor.

The sample size of \( n = 10 \) was chosen based on a series of power analyses using a pilot study of three samples. Normal parameter estimates needed for a one sample \( t \)-test power analysis were estimated by splitting the 95% mean confidence interval from the pilot observations into five equidistant points as well as expanding the pilot standard deviation by magnitudes of 20%, 50%, and 100%. Sample size estimates ranged from 3 to 16 to achieve a power (\( 1 - \beta \)) of 0.80. Sweat rates and sweat \( \text{Na}^+ \) concentrations were compared using a two-tailed paired \( t \)-test (Microsoft Excel). Rejection of the null hypothesis was set at \( P < 0.05 \). Effect sizes were calculated using the mean differences between tattooed and nontattooed skin divided by the pooled standard deviation. Pearson product–moment correlation coefficients were calculated using standard statistical practices for assessing the relationship between the ages of the tattoo with sweat rate and sweat \( \text{Na}^+ \) concentrations.

**RESULTS**

The mean sweat rate from tattooed skin was significantly less than from nontattooed skin (0.18 ± 0.15 vs 0.35 ± 0.25 mg cm\(^{-2}\) min\(^{-1}\); \( P = 0.001 \)). Figure 1 illustrates the
mean sweat rates (±SD) from tattooed and nontattooed skin as well as the individual values for each participant. All ten participants in the study generated a lower sweat rate from their tattooed skin compared to their contralateral nontattooed skin. The mean difference in sweat rate between tattooed skin and nontattooed skin was \(-0.17 \pm 0.11 \text{ mg/cm}^2 \text{ min}^{-1}\) and the effect size (Cohen’s \(d\)) was \(-0.79\). This means that the mean sweat rate from tattooed skin was 0.79 standard deviations lower than the mean sweat rate of nontattooed skin. The mean ratio of sweat rates from tattooed and nontattooed skin (tattooed:nontattoed) was 0.53 ± 0.12. Therefore, on average, the sweat rate from tattooed skin was about half of the sweat rate from nontattooed skin. The correlation coefficient for the relationship between the age of the tattoo and the mean ratio of sweat rates was \(r = 0.05 \ (r^2 = 0.002)\) meaning that less than 1% of the variance in the ratio of sweat rates was explained by the age of the tattoo. Thus, any decrement in sweat rate due to tattooing did not appear to diminish over time.

The mean sweat Na\(^+\) concentration from tattooed skin was significantly higher than for tattooed versus nontattooed skin (69.1 ± 28.9 vs 42.6 ± 15.2 mmol L\(^{-1}\), \(P = 0.02\); see Fig. 2). Nine of 10 participants had higher sweat Na\(^+\) concentration values for tattooed skin than nontattooed skin. The mean difference in sweat Na\(^+\) concentration between tattooed skin and nontattooed skin was 26.5 ± 29.7 mmol L\(^{-1}\) and the effect size (Cohen’s \(d\)) was 1.01. This means that the mean sweat Na\(^+\) concentration from tattooed skin was 1.01 standard deviations higher than the mean sweat Na\(^+\) concentration from nontattooed skin. The mean ratio of Na\(^+\) concentrations between the tattooed and nontattooed skin (tattooed:nontattoed) was 1.73 ± 0.83. Therefore, the mean sweat Na\(^+\) concentration from tattooed skin was 1.73 times higher than that of nontattooed skin. The correlation coefficient for the relationship between tattoo age and ratio of Na\(^+\) concentrations was \(r = 0.25 \ (r^2 = 0.06)\) meaning that 6% of the variance in sweat Na\(^+\) concentration was explained by the age of the tattoo. Therefore, it is unlikely that sweat glands resume normal sodium reabsorption for years after obtaining a tattoo.

**DISCUSSION**

These data clearly demonstrate a diminished sweat rate and a higher sweat Na\(^+\) concentration from tattooed versus nontattooed skin that has undergone cholinergic stimulation. They also provide preliminary evidence that these changes to sweating are unrelated to the age of the tattoo. To the authors’ knowledge, this is the first study of its kind to document alterations in sweating function associated with tattooing so comparisons with similar research are not possible. Likewise, determination of the mechanism to explain these findings is premature. Yet, this study may provide a “proof of concept” for the development and advancement of future research.

In the present study, the stimulus for sweating was pilocarpine, which, is an acetylcholine agonist. The administration of pilocarpine by iontophoresis provided a dose-controlled stimulus delivered locally to the sweat glands and was not temperature dependent since the ambient temperature was controlled at \(-20^\circ\text{C}\). Therefore, it was highly unlikely that the differences in sweat rate or sweat Na\(^+\) seen in the present study were due to any temperature related mechanism. If the observed changes in sweating function were unrelated to temperature, then it was most likely that they were due to alterations within the sweat glands themselves. One could speculate that sweat glands from tattooed skin were less responsive to the cholinergic stimulation. This, too, is unlikely since both sweat rate and sweat [Na\(^+\)] were affected. The binding of the cholinergic stimulus, pilocarpine, signals the production of primary sweat but it is not involved with the reabsorption of sodium and chloride within the straight duct.
leading up to the skin. The primary sweat that is formed in the secretory coil is isotonic to plasma with a sodium concentration of ~140 mmol·L⁻¹. As the primary sweat leaves the secretory coil and traverses up the straight duct toward the skin, sodium and chloride ions are reabsorbed resulting in a final [Na⁺] of 70 to 80 mmol·L⁻¹. The sodium concentration of the final sweat varies according to sweat rate with higher sweat rates resulting in a higher sweat [Na⁺] (4). This is thought to be due to the reduced time that Na⁺ ions remain in the duct and accessible for reabsorption. If sweat glands from tattooed skin are less responsive to the pilocarpine, it would be reasonable to expect a diminished sweat rate but a not an elevated sweat [Na⁺]. In fact, it would be more reasonable to expect a lower sweat [Na⁺] due to the increased time that the sodium ions remain in the duct available for reabsorption but, in the present study, this was not the case. Using the same logic, it is also unlikely that differences in sweat function were due to any specific ion channel except, perhaps the Na⁺/K⁺-ATPase pumps that are operational in both the secretion of primary sweat and the alteration of final sweat.

It is also possible that sweat function was affected by trauma due to repeated punctures to the dermal layer of the skin. It is well documented that trauma from burns (9,19) and artificially induced sunburn (11) diminishes sweat rates. However, this was not the case in a study by Wing et al. (18) who examined the effects of repeatedly puncturing the skin with a stamp pad containing rows of microneedles (pretreatment) before pilocarpine iontophoresis. They reported that after pretreating the skin with microneedles, sweat production increased significantly without altering sweat osmolarity. They concluded that pretreatment with microneedles decreases skin resistance thereby improving drug deliverer by iontophoresis. It should be noted that this study dealt with the immediate effects due to any specific ion channel except, perhaps the Na⁺/K⁺-ATPase pumps that are operational in both the secretion of primary sweat and the alteration of final sweat. Another conceivable mechanism involves an obstruction of the opening of the sweat glands that reduces the amount of sweat emanating from the pore. This is thought to be the mechanism associated with antiperspirant deodorants containing aluminum chloride or aluminum zirconium chlorohydrate glycine complex. Quatrale et al. (13) reported histological evidence that application of a antiperspirant with these active ingredients caused an amorphous electron dense material to the lumen of the sweat gland duct thereby blocking the release of sweat. While the composition of tattoo ink differs from artist to artist, small quantities of aluminum and other metals are common (3). One cannot rule out the possibility of a sweat gland obstruction but it is unlikely that these effects are permanent.

A more plausible mechanism for the present findings involves an innate immune response that is triggered by the insertion of ink under the skin. The tattooing process initiates an inflammatory response known as chemotaxis, which is a multistage process that attracts neutrophils and monocytes from the blood to the injured tissue. The monocytes are transformed into functional macrophages that engulf a portion of the dye by phagocytosis. Neutrophils and macrophages are known to secrete inflammatory mediators and other toxic chemicals into the injured area that are not specific with regard to their target. Consequently, normal tissue including sweat glands may be damaged during an inflammatory response that is directed at invading pathogens.

Lea and Palowski evaluated ultrathin serial sections of human biopsy specimens, taken at 24 h, 1 month, and 1, 3, and 40 yr post-tattooing under electron microscope. They discovered that an acute inflammatory reaction characterized the tattooed area and many cells became involved in “cleaning up” the cellular debris resulting from the tattoo needles, while simultaneously taking up ink particles (8). Karanth (6) also demonstrated an increased number of Langerhans cells in frozen sections of skin biopsies taken from tattooed skin compared to control skin. Langerhans cells are dendritic cells that reside in the basal layer of the epidermis and serve as the lookouts of the adaptive immune system. They continuously scan the surface layers of skin and transport foreign antigen for presentation to responsive T lymphocytes in regional lymph nodes. As such, Langerhans cells play essential roles in the initiation of cutaneous immune responses, including immune responses to chemical allergens encountered at skin surfaces. In the same study, Karanth, found a persistently high number of Langerhans cells in tattooed skin for years after obtaining a tattoo.

Although attenuation of sweat rate observed in the present study may ultimately have relevance to thermoregulation, it is wise to interpret these initial findings as preliminary. First, the ultimate purpose of sweating is to contribute to a person’s ability to manage a heat load. In the present study, the participants were not challenged with a heat load and, thereby, it remains unknown whether similar alterations in sweating would accompany treatments involving external heating or internal metabolic heat production. Second, most individuals who obtain a tattoo will do so to only a small proportion of their overall skin surface area. In that case, one would expect that their nontattooed skin would compensate for any diminished sweating by their tattooed skin and that they would be able to manage a moderate heat load without any problem. However, it remains to be determined whether a high heat load combined with a large proportion of tattooed skin would sufficiently limit whole-body heat loss to an extent that heat balance could not be achieved resulting in a continuous rise in core temperature. This is particularly relevant with occupations (military and mining) and sporting endeavors (running and triathlon) that require high metabolic rates for prolonged periods in very hot/humid conditions, where individuals with increasing amounts of tattooed skin may be at further risk of noncompensable heat stress.

In summary, this study examined the differences in sweat rate and Na⁺ concentration between tattooed and nontattooed skin. Upon stimulation with pilocarpine, tattooed skin was associated with a lower sweat rate and a higher sweat Na⁺ concentration than nontattooed skin. These changes in sweating function appeared to be unrelated to the age of the tattoo.
Additional studies need to be conducted to determine the mechanism associated with these changes in sweat function and the extent that they may affect thermal balance.

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REFERENCES