Na⁺ secretion rate increases proportionally more than the Na⁺ reabsorption rate with increases in sweat rate

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The formation of sweat in the human eccrine gland is the result of a two-step process. The first step involves the secretion of an isosmotic precursor sweat by the proximal secretory coil. Next, ductal water reabsorption is small (13) because dehydration is known to affect [Na⁺]secretion because of a reduction in ductal Na⁺ reabsorption. Water was allowed ad libitum during each of the five exercise trials. The intensity for the five exercise bouts in the heat was set to 50, 60, 70, 80, and 90% of age-predicted maximum heart rate. Forearm sweat samples and capillary blood samples were collected during each of the five 30-min exercise bouts. The sweat and blood samples were analyzed for Na⁺ concentration in sweat and serum, which were used to calculate the rate of Na⁺ secretion and Na⁺ reabsorption. The mean correlation between sweat rate and Na⁺ concentration in sweat was found to be r = 0.73. Within the sweat rate range of the present study, both Na⁺ secretion rate and Na⁺ reabsorption rate increased linearly; however, the Na⁺ secretion rate increased almost twice as fast (slope = 141 vs. 80). Thus the rate at which Na⁺ escaped reabsorption increased with increases in sweat rate and was significantly (P < 0.05) correlated to the Na⁺ concentration in sweat (mean r = 0.90). Such results strongly suggest that the physiological mechanism responsible for the previously reported linear increase in Na⁺ concentration in sweat seen with increases in sweat rate is that the Na⁺ secretion rate increases proportionally more than the Na⁺ reabsorption rate. Such data should help to elucidate the physiological mechanism responsible for the previously reported linear relationship between increases in sweat rate and [Na⁺]sweat.

METHODS

The subjects for the study were 10 healthy volunteers (4 men and 6 women). They had a mean ± SE age, height, weight, and body surface area of 25 ± 1 yr, 168.7 ± 2.4 cm, 66.5 ± 3.6 kg, and 1.76 ± 0.06 m², respectively. The subjects were recruited from the San Diego area using posted flyers and via word of mouth. They were all recreationally active; however, none was formally heat acclimated. They were instructed to refrain from initiating new exercise programs before each exercise bout in an environmental chamber set at 35°C and 40% relative humidity. The intensity for the five exercise bouts in the heat was set to approximate 50, 60, 70, 80, and 90% of age-predicted maximum heart rate. Forearm sweat samples and capillary blood samples were collected during each of the five 30-min exercise bouts. The sweat and blood samples were analyzed for Na⁺ concentration in sweat and serum, which were used to calculate the rate of Na⁺ secretion and Na⁺ reabsorption. The mean correlation between sweat rate and Na⁺ concentration in sweat was found to be r = 0.73. Within the sweat rate range of the present study, both Na⁺ secretion rate and Na⁺ reabsorption rate increased linearly; however, the Na⁺ secretion rate increased almost twice as fast (slope = 141 vs. 80). Thus the rate at which Na⁺ escaped reabsorption increased with increases in sweat rate and was significantly (P < 0.05) correlated to the Na⁺ concentration in sweat (mean r = 0.90). Such results strongly suggest that the physiological mechanism responsible for the previously reported linear increase in Na⁺ concentration in sweat seen with increases in sweat rate is that the Na⁺ secretion rate increases proportionally more than the Na⁺ reabsorption rate. Such data should help to elucidate the physiological mechanism responsible for the previously reported linear relationship between increases in sweat rate and [Na⁺]sweat.
rate (220 minus age). The absolute workloads were not critical and were simply designed to produce various sweat rates, ranging from very low to very high, from each of the subjects. Heart rate was continuously measured during exercise using a Polar heart rate monitor. The order of the five exercise bouts was randomly assigned for all subjects.

Forearm sweat samples were collected for 20 min (from minute 10 to minute 30) during each of the five 30-min exercise bouts using macrground sweat collectors (Wescor, Logan, UT). This protocol was designed to allow the subjects to start sweating before the initiation of sweat collection and has been successfully used in the past (25). The collectors were placed on the flexor surface of the proximal forearm and were held in place by a Velcro strap that prevented leakage and sample contamination. The collection site was identified so that the macrground sweat collectors could be placed at the same location for all trials. The skin was cleaned with deionized water and dried immediately before securing each collector. Forearm sweat rate, for the approximate 5-cm² area under the macrground sweat collector, was calculated by the securing each collector. Forearm sweat rate, for the approximate macrground could be placed at the same location for all trials. The skin sample contamination. The collection site was identified so that the macrground collectors were placed on the flexor surface of the proximal forearm designed to allow the subjects to start sweating before the initiation of macroduct sweat collectors (Wescor, Logan, UT). This protocol was measured ANOVA was used to compare mean Na⁺ reabsorption rate, expressed in nanomoles per square centimeter per minute, was calculated according to the following formula developed by Sato (23):

\[ \text{Na}^+ \text{ reabsorption rate} = \left[ (\text{sweat rate} \times [\text{Na}^+]_{\text{sweat}}) - (\text{sweat rate} \times [\text{Na}^+]_{\text{serum}}) \right] / \text{change in volume within the sample collection tubing} \]

Last, the rate at which Na⁺ escaped reabsorption was calculated as the difference between the Na⁺ secretion rate minus the Na⁺ reabsorption rate.

For the previously mentioned dependent variables, Pearson product-moment correlations were calculated for each subject and the mean r was the average of the 10 values. A two × five repeated-measures ANOVA was used to compare mean Na⁺ secretion vs. reabsorption data, while a one-way repeated-measures ANOVA was used to analyze the mean relative Na⁺ reabsorption rate data. Post hoc analyses were conducted using dependent r-tests and were adjusted using the Bonferroni correction. Overall significance was set at the P < 0.05 level.

RESULTS

In agreement with numerous past studies (1, 2, 4, 9, 11, 14, 32) the results showed that within the range studied there was a significant linear relationship between sweat rate and [Na⁺]sweat. There were no differences in responses between men and women; thus all data were grouped together. As can be seen in Fig. 1 the mean r was 0.73. At the lowest sweat rate, the mean ± SE [Na⁺]sweat was 19 ± 5 mmol/l, whereas at the highest sweat rate it increased to 59 ± 10 mmol/l. These values are similar in magnitude to other studies that sampled sweat locally (1, 2, 4). Figure 2 shows that the rate at which Na⁺ was secreted into the coil and the Na⁺ reabsorption rate during passage of sweat down the distal duct of the eccrine gland both increased linearly with increases in sweat rate. However, the Na⁺ secretion rate increased significantly faster, as evidenced by a significant interaction (P < 0.05) and by having a slope that was almost twice as great (141 vs. 80) as the Na⁺ reabsorption rate. Conversely, as shown in Fig. 3, the mean relative Na⁺ reabsorption rate, expressed as a percentage of Na⁺ secreted during precursor sweat formation, decreased significantly with increases in sweat rate. Specifically, 86 ± 3% of the secreted Na⁺ was reabsorbed at the lowest sweat rate, and this decreased to 65 ± 6% at the highest sweat rate.

Conceptually, the difference between the two lines presented in Fig. 2 is the rate at which Na⁺ escaped reabsorption during passage along the distal duct. In other words, it represents the amount of Na⁺ not reabsorbed that ultimately would appear as Na⁺ in the sweat. Figure 4 illustrates this relationship and shows that the rate at which Na⁺ escaped reabsorption was highly correlated (mean r = 0.90) with [Na⁺]sweat.

DISCUSSION

Numerous studies (1, 2, 4, 9, 11, 14, 32) have previously reported that as sweat rate increases so does the [Na⁺]sweat. However, the physiological mechanism responsible for this phenomenon is currently unknown. Conceptually, [Na⁺]sweat should be the difference between the amount of Na⁺ secreted during precursor sweat formation in the proximal coil minus the rate of ductal Na⁺ reabsorption. The data from the present study are believed to be the first to measure these two parameters in vivo over a wide range of different sweat rates and are presented in Fig. 2. As can be seen, within the range of sweat rates reported in the current study the rate of both Na⁺ secretion and Na⁺ reabsorption increased in a linear fashion. Such findings support the work of others (3, 5, 25) who have indirectly determined ductal Na⁺ reabsorption by calculating the maximum free water clearance, which is proportional to maximum water-free Na⁺ reabsorption. These studies reported that there was a significant linear relationship between maximum free water clearance and maximum sweat rate (3, 5, 25). However, in the current study, the rate of increase in Na⁺ secretion was proportionally greater, as evi-
demonstrated by having a slope that was almost double that of Na⁺ reabsorption (141 vs. 80). Such findings do not support Taylor’s (30) contention that “Elevated flow will increase sodium and chloride sweat content, possibly because of reduced ductal reabsorption at higher flow rates.” Clearly the absolute rate of Na⁺ reabsorption was not reduced with increases in sweat rate in the present study. Furthermore, several previous investigators (2, 4, 27) have hypothesized that Na⁺ reabsorption would become saturated at high sweat rates, thus causing the observed increase in [Na⁺]sweat. Again, the data presented in Fig. 2 do not support such a hypothesis as Na⁺ reabsorption did not demonstrate a plateau effect with increases in sweat rate up to 0.82 mg·cm⁻²·min⁻¹ or ~1 l/h.

It could be argued that possibly the sweat rates attained with the present study were not large enough and thus failed to elicit a plateau effect in Na⁺ reabsorption. However, the mean forearm sweat rate produced by the most intense workload (i.e., 90% of maximal heart rate) in the present study (0.82 mg·cm⁻²·min⁻¹) was similar in magnitude to previous results that were produced by exercise in the heat or maximal pharmacological stimulation (5, 8, 10, 18, 19). For example, Gonzalez et al. (10) reported a mean forearm sweat rate of 0.75 mg·cm⁻²·min⁻¹ in six healthy men during exercise in a hot (40°C) humid (90% relative humidity) environment that produced a mean esophageal temperature of 38.9°C. In addition, Crandall et al. (8) reported that pharmacological-induced maximal forearm sweat rate was 0.63 mg·cm⁻²·min⁻¹. Thus lower than expected forearm sweat rates do not seem to be the likely cause for not observing a plateau in the Na⁺ reabsorption vs. sweat rate relationship data presented in Fig. 2.

Rather the present data suggest that within the range of sweat rates studied, the absolute rate of Na⁺ reabsorption from the distal duct increases continuously with increases in sweat rate. However, when expressed as a relative percentage of Na⁺ secretion, it decreases with increases in sweat rate. Specifically, Fig. 2 shows that the absolute rate of Na⁺ reabsorption at the highest sweat rate was almost three times that seen during the lowest sweat rate (77 vs. 29 nmol·cm⁻²·min⁻¹). However, as can be seen in Fig. 3, at the lowest sweat rate ~86 ± 3% of the Na⁺ secreted during precursor sweat formation were able to be reabsorbed during passage along the distal duct of the eccrine gland. This value was significantly reduced to 65 ± 6% at the highest sweat rate. Such data clearly show that the faster the precursor sweat travels along the distal duct (i.e., the greater the sweat rate) the smaller the percentage of total Na⁺ secreted that can be reabsorbed. This decrease in relative Na⁺ reabsorption with increases in sweat rate could be the result of a number of factors, including decreased contact time of the fluid column with the luminal membrane of the distal duct, a decreased lumen-intracellular diffusion gradient, and/or saturation of various transport proteins. An alternate mechanism may involve the fact that decreases in the cytoplasmic pH have been shown (6) to reduce the activity of the amiloride-sensitive epithelium Na⁺ channel (ENaC), which is the primary luminal path used for ductal Na⁺ reabsorption. Because glycolysis appears to be the predominate way that
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ecrine sweat glands resynthesize ATP (23), it is not unreasonable to hypothesize that the cytosolic pH could decrease with increases in sweat rate, thus reducing ENaC activity. Such a scenario could explain the decreased relative Na\(^+\) reabsorption vs. sweat rate relationship seen in the present study. Interestingly, the mean relative Na\(^+\) reabsorption data presented in Fig. 3 are quantitatively similar to the in vitro results presented by Mangos (13) collected using isolated human distal ducts. He reported that following microperfusion at a rate approximately equivalent to a sweat rate of 0.5 mg cm\(^{-2}\) min\(^{-1}\) with an isotonic solution containing 145 mmol/l of Na\(^+\), that the mean relative Na\(^+\) reabsorption was 77%. This agrees quite favorably with the results of the present study presented in Fig. 3.

Theoretically, the difference between the rate of Na\(^+\) secretion and the rate of Na\(^+\) escaped reabsorption, or the rate at which Na\(^+\) escaped reabsorption should be highly related to the [Na\(^+\)]\(_{\text{sweat}}\). As can be seen in Fig. 4, these two variables were linearly related and had a mean r of 0.90. Thus the more Na\(^+\) that escaped reabsorption during passage along the distal duct the higher the mean [Na\(^+\)]\(_{\text{sweat}}\). Interestingly, as can be seen by examining the x- and y-axis, the rate of Na\(^+\) escaping reabsorption increased about sixfold, whereas the [Na\(^+\)]\(_{\text{sweat}}\) only increased about twofold. At first glance this seems illogical; however, it must be remembered that during the same time the sweat rate also increased threefold. Thus a sixfold increase in Na\(^+\) escaping reabsorption should only increase the [Na\(^+\)]\(_{\text{sweat}}\) about twofold, if the sweat rate simultaneously increased threefold.

There are three assumptions that were made in the present study that need to be further addressed. First, it was assumed that the measured [Na\(^+\)]\(_{\text{serum}}\) provides a valid measure of the precursor [Na\(^+\)]\(_{\text{sweat}}\). The support for this assumption is based on the fact that numerous previous studies, using three different techniques, have all shown that precursor sweat is isotonic to serum. Specifically, Sato and Dobson (25) using the backward extrapolation technique reported that the mean ± SE precursor [Na\(^+\)]\(_{\text{sweat}}\) in the forearm of 14 healthy volunteers was 140 ± 1.8 mmol/l. Additionally, precursor sweat samples collected directly from the secretory coil were shown to be isotonic to serum (24), having a mean Na\(^+\) concentration of 142 mmol/l. Finally, sweat samples collected following the application of amiloride, which selectively inhibits Na\(^+\) reabsorption from the distal duct, were isotonic to serum (20). Taken together these three studies clearly support the validity of using [Na\(^+\)]\(_{\text{serum}}\) as a surrogate for precursor [Na\(^+\)]\(_{\text{sweat}}\).

The second assumption made in the present study was that following reabsorption from the distal duct, Na\(^+\) have little, if any, backdiffusion. Theoretically, this seems defensible since in the human eccrine sweat gland the ENaC is only expressed in the luminal membrane, whereas the ouabain-sensitive Na\(^+\)-K\(^+\) ATPase is localized to the basolateral membrane (21, 23). Furthermore, the one study (13) that has directly measured Na\(^+\) backdiffusion using radioactive \(^{22}\)Na reported that at sweat rates comparable with those reported in the present study only 8% of the reabsorbed Na\(^+\) reentered the distal duct via backdiffusion, presumably via a paracellular pathway.

The third assumption was that leaching of Na\(^+\) from the stratum corneum into the collected sweat sample was minimal. Because it has been previously shown (7, 9, 19) that K\(^+\) concentration in sweat stays relatively constant, regardless of sweat rate, Weschler (31) has recommended comparing K\(^+\) concentration in sweat vs. serum as an indicator of potential leaching. In the present study, 18 randomly selected paired sweat and serum samples were analyzed for K\(^+\) concentration using flame photometry. The mean K\(^+\) concentrations in sweat and serum were both 5.1 mmol/l. These data strongly suggest that leaching of Na\(^+\) was negligible in the current study.

In conclusion, the results of the present study provide insight into the physiological mechanism responsible of the well documented increase in [Na\(^+\)]\(_{\text{sweat}}\) with increasing sweat rates. The results show that within the sweat rate range of the current study the absolute rates of both Na\(^+\) secretion and Na\(^+\) reabsorption increase linearly with increases in sweat rate. However, Na\(^+\) secretion increases proportionally faster, thus, the rate at which Na\(^+\) escape reabsorption during passage along the distal duct of the sweat gland increases at higher sweat rates. Lastly, the rate of Na\(^+\) escaping reabsorption was linearly related to the [Na\(^+\)]\(_{\text{sweat}}\). Such data strongly support that with increasing sweat rates, the rate of Na\(^+\) secretion during precursor sweat formation increases faster than Na\(^+\) reabsorption in the distal duct. This difference allows for a greater percentage of the secreted Na\(^+\) to escape reabsorption, thus causing [Na\(^+\)]\(_{\text{sweat}}\) to increase.

REFERENCES


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