Review

Wiley Online Library

ELECTROANALYSIS

SPECIAL ISSUE

DOI: 10.1002/elan.201600018

Non-invasive Analyte Access and Sensing through Eccrine Sweat: Challenges and Outlook circa 2016

Jason Heikenfeld*[a, b]

Abstract: Despite the many ergonomic advantages of eccrine perspiration (sweat) compared to other possible biofluids (particularly in "wearable" devices), sweat remains an underrepresented source of biomarker analytes compared to the established biofluids blood, urine, and saliva. Upon closer comparison to other non-invasive biofluids, the advantages may even extend beyond ergonomics: sweat might provide superior analyte information. A number of challenges, however, have historically kept sweat from its place in the pantheon of clinical samples. These challenges include very low sample volumes (nL to μL), unknown concentration due to evaporation, filtration and dilution of large analytes, mixing of old and new sweat, and the potential for contamination from the skin surface. More recently, rapid progress in "wearable" sweat sampling and sensing devices has resolved several of the historical challenges. However, this recent progress has also been limited to high concentration analytes (μM to mM) sampled at high sweat rates (>1 nL/min/gland, e.g. athletics). Progress will be much more challenging as sweat biosensing moves towards use with sedentary users (low sweat rates or not sweating at all) and/or towards low concentration analytes (pM to nM). Addressing these unresolved challenges will require significant advances in sweat stimulation, sample collection efficiency, compact sensors, and likely more. Fortunately, none of the remaining challenges appear to be fundamentally blocking, and scientific and engineering innovations have the opportunity to enable broader application of sweat biosensing technology.

Keywords: Eccrine · perspiration · sweat · sensors · electrochemical · wearable · microfluidics

1 Introduction

Although wearable technologies have recently seen a water-shed moment in their commercialization, the introduction of new capabilities has not kept pace with rapidly rising expectations in the medical, industrial, and consumer markets. This is partly because today's wearable technologies remain heavily reliant on decades-old techniques for accessing information from the body. Many wearable technologies, for example, simply electrically or optically interrogate the body from outside skin, which severely limits the specificity of their detection. The identification and quantification of most biomarkers (ions, molecules, proteins, etc.), is instead only possible through a probe that relies on direct chemical interaction with the analyte. Creating such chemical sensors in a wearable format remains a significant challenge, but, as revealed in this special issue of Electroanalysis, appears promising. However, even if you can make such sensors [1], there remains a second, perhaps even greater challenge: how does one reliably and non-invasively extract biomarker analytes from the body? It has been recently postulated by several groups that this second challenge can be resolved by sampling and sensing of analytes in eccrine sweat (Fig. 1), as supported by emergent theory [2] and recent technological demonstrations [1–6]. This article reviews the status of wearable biosensing of analytes in eccrine sweat, including both challenges and outlook circa 2016. Eccrine sweat has many inherent advantages over other *non-invasive* biofluids (tears, saliva, urine), but also has numerous and unique challenges. This article may serve useful for those considering exploration of sweat biosensing, but also raises many issues that are relevant to biosensing with other non-invasive or minimally invasive techniques.

2 Sweat: Pros and Cons as an Analyte Source

Eccrine sweat glands are found at many locations of the body at densities of >100 glands/cm^2 [2], therefore enabling a multitude of viable sampling sites. Thus, the argument for using a wearable device to sample and sense sweat is rather straightforward. The finer arguments in favor of or against eccrine sweat, are best understood in the context of alternate methods of analyte sampling. For example, one-time sampling and sensing of eccrine sweat is *not* analytically superior to invasive biofluid extraction (blood, interstitial fluid, etc.). Rather, to understand the advantages and challenges of eccrine sweat, is to also understand sweat in the context of repeated or continuous sampling, and in the context of other *non-invasive* biofluids (saliva, tears, urine). Within these contexts, major

[a] J. Heikenfeld
Professor, Univ. Cincinnati, Depts. of Elect. Engineering and Computing Systems, Materials Engin., Biomed. Engin
*e-mail: heikenjc@ucmail.uc.edu

[b] J. Heikenfeld
Chief Science Officer, Eccrine Systems Inc., Cincinnati, Ohio, USA

© 2016 Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim

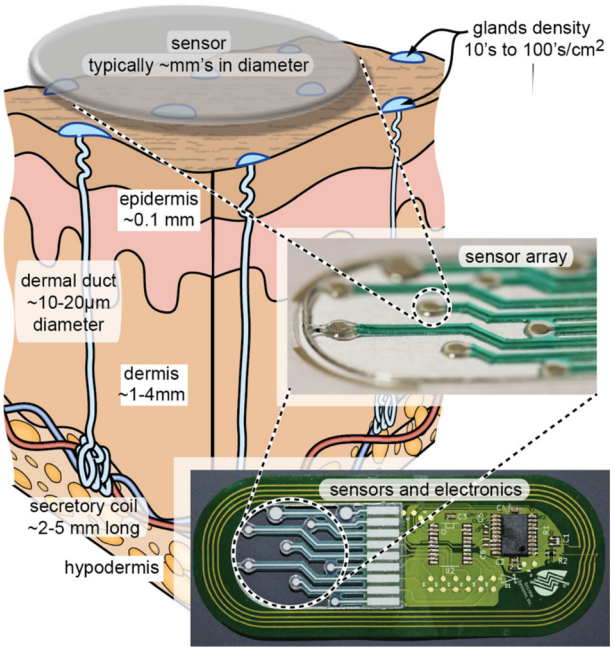


Fig. 1. Diagram and example technology [2,4] for sensing analytes in sweat as it is secreted onto skin. Multi-analyte RFID patch photos provided courtesy of Eccrine Systems Inc.

advantages and challenges of eccrine sweat are briefly summarized below, followed by brief qualification of challenges as technological (likely directly resolvable with enough effort) or fundamental (only resolvable through indirect methods or 'work-arounds').

Advantages *of sweat for non-invasive biofluid access:*

- Most sampling sites and surface area outside the body
- Continuous access (e.g. urine requires a catheter)
- Can be stimulated on-demand with local iontophoresis
- Ease of placement and comfort (not in mouth or eye)
- Is not a digestive fluid that can degrade some analytes
- Can be sampled at very low fluid generation rates
- Can be sampled without external contamination
- Can be sensed quickly before analytes degrade.

Some additional detail is provided for the last three of these advantages. First, low fluid generation rates can, for some larger size analytes, allow greater time for concentration equilibration with blood or improved correlations with blood by comparing ratios of analytes [2,7] (see Fig. 5 and related discussion). Second, once adequately sealed onto a cleaned skin surface, sensors can be shielded from external fluids and contamination sources. Elimination of external contamination also improves sample security (e.g., illicit drug testing [8]). Third, many biomarker analytes can degrade in as little as tens of minutes due to microbes, proteinases and other enzymes [9]. For sweat, wearable sensors can be brought extremely near to the skin and sample the new sweat quickly before it degrades [2,9].

Challenges *of sweat for non-invasive biofluid access:*

- Underdeveloped compared to most other biofluids
- Irregular sampling without iontophoretic stimulation
- Skin surface presents a contamination source
- Old sweat can mix with and contaminate new sweat
- Very low sample production rates (1's nL/min/mm^2)
- The most widely-used sensing methods are not available in forms that are continuous and/or compact
- *Large pH changes (4.5 to 7.0) are possible, which can skew concentrations of weak acids and bases*
- *Most large analytes (e.g. proteins) are diluted (filtered)*
- *Some analyte concentrations only represent skin and/or the metabolism of the eccrine sweat gland itself*
- *Active analyte channels in the eccrine gland can skew the concentrations of some analytes relative to the concentrations seen in blood.*

The first six challenges listed here are all at least in part addressable through technological innovation. These first six challenges will be discussed in greater detail in Section 3. The last four *italicized* challenges are fundamental (physiology), and if not impossible to overcome, require solutions that are indirect and complex (analyte proxies, sample preparation before sensing, etc.). In addition, there are also several *historical* disadvantages presented in the next section which were not covered in this section, because they are now easily resolved.

3 Technology circa 2016 – Quickly Addressing the Easy Challenges

There are several additional *historical* challenges for sweat, which are nicely summarized by Luque de Castro et al. [10] in the following excerpt:

"The main limitations of sweat as a clinical sample are the difficulty to produce enough sweat for analysis, sample evaporation, lack of appropriate sampling devices, need for a trained staff, and errors in the results owing to the presence of pilocarpine. In dealing with quantitative measurements, the main drawback is normalization of the sampled volume."

With exception to the challenge of pilocarpine interference, simply using an integrated wearable device or patch, which places sensors or sweat collection right against skin (Figs. 1, 2), resolves all the other issues cited

Jason Heikenfeld is a professor and device scientist at the Univ. of Cincinnati. He is a Senior member of the Institute for Electrical and Electronics Engineers, a Senior member of the Society for Information Display, a life-member of SPIE, a member of ASEE, and a Fellow of the National Academy of Inventors. In addition to his scholarly work, Prof. Heikenfeld is an award-winning educator, and has led the creation of programs and coursework that foster innovation, entrepreneurship, and an understanding of the profound impact that technology has on society.



© 2016 Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim

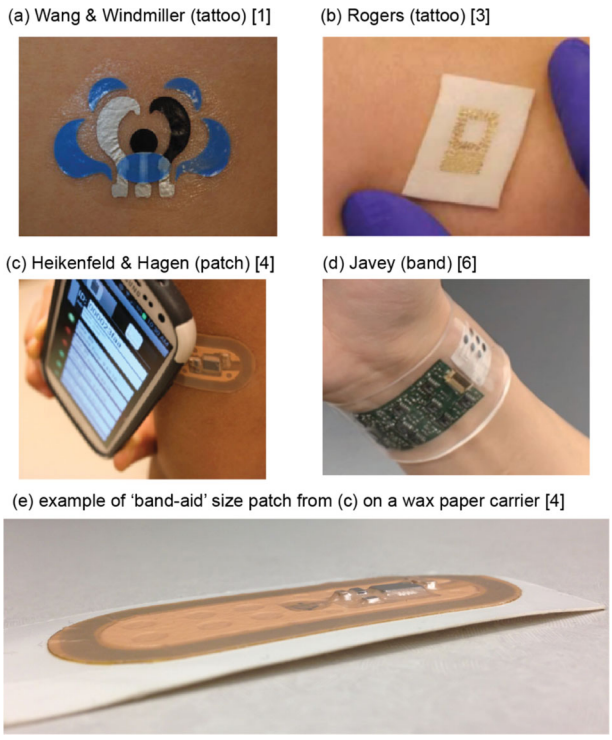


Fig. 2. Circa 2016 – bringing sensors directly onto skin [1,3,4,6] solves numerous historical challenges [10]. In all the examples provided in the figure, technology is mechanically compliant to skin, which is essential as a first step to reduce the sweat volume between skin and sensors.

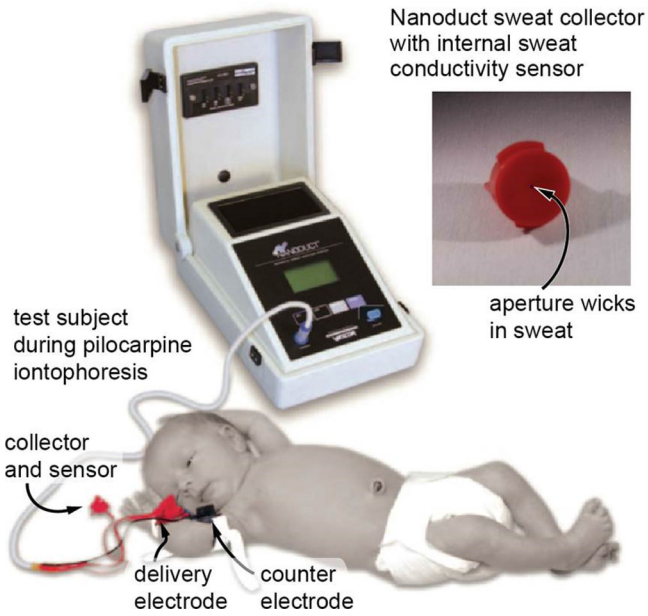


Fig. 3. Wescor Nanoduct sweat stimulation and chloride sensing system for infant cystic fibrosis testing [11].

by Luque de Castro et al. [10]: (1) you need much less sample (see next section for calculations); (2) sample evaporation is blocked by the sensor or patch; (3) the device or patch is the appropriate sampling device; (4) the device largely replaces the need for highly trained staff; (5) and lastly, normalization of sample volume is no longer relevant as sensors continuously receive new sweat. The sample volume can also be normalized by continuously determining the flow rate of new sweat to the device. This flow rate of sweat can be directly measured (e.g. flow sensors inside a microfluidic channel) or indirectly measured (e.g. measure sodium or chloride ion concentrations which are proportional to sweat generation rate [2]). One challenge cited by Luque de Castro et al. [10] was not addressed in our discussion, "*and errors in the results owing to the presence of pilocarpine,*" and will be addressed in the next section.

Hence, circa 2016, many of the historical challenges clinically perceived for sweat can be resolved by quickly sampling and sensing sweat as it secreted onto the skin surface (Fig. 1, 2). An example commercial validation has existed for several years. The Wescor Nanoduct sweat Cl^- measurement system iontophoretically stimulates sweating, and then continuously collects and senses sweat (Fig. 3). The device measures sweat conductivity which is then used to back-calculate sweat Cl^- content (ionic strength of sweat is dominated by NaCl and KCl). A higher than usual Cl^- concentration is a positive predictor for infant Cystic Fibrosis [11].

Thus far, this section of the review has suggested that historical challenges are resolved by simply placing sensors very near to the skin surface. There are several other caveats that should be considered. Ideally, the sensor or sampling component should not be frequently sliding over the skin surface, which could cause entrapment of old or contaminated sweat. Preferably, placement on skin should maintain tight contact to minimize the amount of dead volume between sensors and the skin surface. If you can minimize the amount of dead volume between the device and skin (Fig. 4a; [2]), the amount of time sweat sits 'stagnant' on skin is reduced, which will decrease analyte contamination coming from the dead stratum corneum [12] (see next section for more detailed discussion). Furthermore, if you minimize the dead volume, you need less sample volume per reading, and your time for obtaining a new sample is reduced (Fig. 4b, [2], and also see next section). Importantly, a reduced dead volume will also increase the sweat flow rate across the sensors, ideally to a velocity that is greater than the back-diffusion velocity of solutes in older sweat (Fig. 4c, [2]). As a result the challenge that 'old sweat can mix with and contaminate new sweat' can be resolved.

Now that we understand the challenges that are at least partially addressable with technology circa 2016, we turn the discussion to challenges that remain largely unresolved in the current literature.

4 Technology Outlook – the Big Challenges

Technology circa 2016 or earlier [1–11, and references therein] may be adequate for initial applications such as

© 2016 Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim

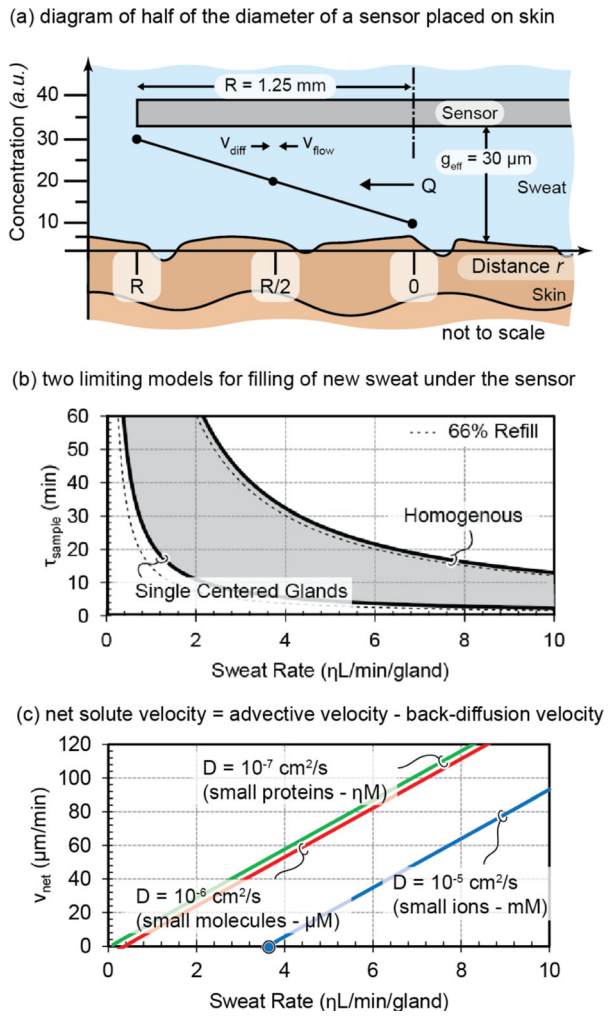


Fig. 4. Effects of (a) sensor gap from skin and of sweat gland generation rate on (b) time required for filling of a new sweat sample under a sensor and (c) preventing mixing of old and new sweat solutes (adapted from [2]).

monitoring electrolyte loss in athletes [13], or for cystic fibrosis testing [11], but the majority of untapped opportunity for sweat biosensing lies beyond just simply measuring Na^+, K^+, and Cl^-. This section is broken into several subsections, each detailing a significant unresolved challenge.

● *Available sample volumes are very low (1's–100's nL)*

Assume a skin roughness peak-to-valley height of ~60 µm [14,15] and a 10 mm^2 sweat sensor placed on skin. The resulting open ('dead') volume between skin and the sensor can be theoretically approximated at 300 nL. Assume a moderate to high sweat generation rate of 1 nL/min/gland and gland density of 100 glands/cm^2 (10 glands under the sensor). It would take 30 minutes to obtain a viable sample to fill the volume based on a single-centered filling model (Fig. 4b) [2]. Even worse, the real filling method would involve multiple glands, and therefore would be more homogeneous and require more than one hour (Fig. 4b) [2]. With hair, skin defects, or debris, this open volume could be even larger. Fortunately, skin does become more compliant (smoother) when wet and under prolonged contact pressure. Also, another mitigating factor is for applications such as athletics, which can have much higher sweat generation rates (up to ~5 nL/min/gland). However, for other applications obtaining such high sweat generation rates may be impractical. Therein lies a central challenge for sweat biosensing: *the 10's of µL frontier enabled by point of care advances such as i-STAT cartridges, is still ~1000X too large to satisfy many applications of sweat biosensing.* Advanced material and microfluidic techniques are clearly needed between sensors and skin, if fast sampling and sensing is to be enabled at lower sweat rates (<1 nL/min/gland). Considering that many small molecule analytes diffuse immediately into sweat and that the eccrine duct requires only a few minutes to provide a fresh new sample [2], in some cases sampling resolutions of <2 minutes could be theoretically possible with technological innovation.

● *Irregular sampling without iontophoretic stimulation*

Recall the challenging sample volume requirement of the previous section, which was calculated for 1 nL/min/gland (a moderate to high sweat rate). Now consider yourself, likely in a sedentary state while reading this very article, and therefore unlikely to be sweating (unless this article is unexpectedly intriguing). Fortunately, even for sedentary individuals sweat events do occur naturally multiple times per day (during sleep, when you step into a hot car in the summer, often when you eat a meal, due to anxiety, etc.). However, many of these events are infrequent and will generate sweat at only ~0.1's nL/min/gland or less. This challenge places further emphasis on reducing the needed sweat sample volume, such that more natural sweat events during daily life are also *viable* sweat sampling and sensing events.

An alternative approach to resolving irregular sweat sampling is continuous or repeated sweat stimulation. There exists an FDA-approved approach for sweat stimulation using iontophoresis of pilocarpine (e.g. Wescor Nanoduct Fig. 3, [11]). This is only a one-time sampling and sensing protocol, which is unfortunate because one of the key potential advantages for sweat is continuous or repeated sampling and sensing (Sect. 2).

For regular or continuous *stimulation of sweat*, there are two things that might help: (1) reducing the sampling volume (see previous section), such that lower sweat generation rates are viable (0.1's nL/min/gland), which extends the effective duration of adequate sweat flow due to a dose of pilocarpine; (2) validation and approval of sweat stimulants (cholinergic agonists) that metabolize more slowly than pilocarpine. There are several cholinergic agonists with slow metabolic conversion (insensitivity to acetylcholinesterase) that are already in use for ophthalmic and urinary retention applications [16], but which have not been pursued or approved for sweat stimulation. If both reduced sample volume and slower metabolizing stimulants are achieved, this author speculates that con-

© 2016 Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim

tinuous sampling should be possible for 24 hours, or maybe even more.

- *Reduce or eliminate skin-surface contamination*

Before talking about analyte contamination from the skin-surface, recall that the previous two challenges addressed in this section both pointed toward sweat sampling and sensing at lower sweat generation rates. Consider now, the additional and competing challenge that the lower the sweat generation rate, generally the greater the influence of contamination coming from the skin surface [12]. Using a first order approximation, consider *ex-vivo* contamination on the skin surface to be something that dissipates over time (finite source, dissolves, and is advectively transported away by sweat). Next consider *in-vivo* contamination to be at a constant flux, from sources such as dead skin cells, lipophilic analytes diffusing through the stratum corneum, or even include microbes as a constant source of contamination. The contamination effect of all these sources will be diluted and diminished at higher sweat generation rates (nL/min/gland). Again, this is at odds with resolving the previous two challenges of this section, which instead concluded on a need for sampling and sensing at lower sweat generation rates.

Previous clinical techniques may provide clues for mitigating contamination. Sato and colleagues published prolifically on the physiology of eccrine sweat glands, including a technique for higher purity sweat collection [12]. In this technique the skin surface is coated with a thin layer of oil through which sweat can easily penetrate due to the hydraulic pressure of sweat generation [2]. This oil film allows sweat through, but blocks diffusion of hydrophilic contaminants from skin areas further away from the sweat ducts. However, this is just a clinical method for sampling sweat. If this method were adapted as-is to a wearable device (such as those in Fig. 2) the sensors and/or microfluidics could become coated with oil and rendered inoperable.

- *Many requisite sensor modalities are not available in compact and continuous sensing modalities*

This need is particularly well suited to the audience of this special issue of *Electroanalysis*. Sweat's greatest opportunity is likely in repeated or continuous sensing, which for biosensors is the least developed because most biosensors were developed for biofluids in which a one-time sampling/sensing protocol is used (blood, saliva, urine). Recalling our previous discussion in which it is clear the sensors should be wearable, and taking cost into consideration, electrochemical sensing [1] is an attractive platform for sweat analytes. However, the electrochemical sensing should ideally be: (1) selective, as the sweat sample is not pre-treated or filtered; (2) label and reagent-free; (3) shelf-stable. For mM to μM analyte concentrations, there are many ion-selective electrodes and enzymatic/amperometric sensors, which readily satisfy all three of these ideal requirements for sweat [1,3–6]. Requirements (2) and (3) generally prevent use of antibody-based electrochemical sensors. Thus, a major challenge is revealed in the sensing of nM to pM analytes. Such analytes include, for example, small molecule hormones or peptides/proteins[2]. For nM sensing, arguably the most convincing data has been shown for electrochemical aptamer-based (EAB) sensors [17], which can work continuously even for 24 hours in whole blood [18]. Importantly, EAB sensors that have an attached redox couple [17], ion-selective electrodes, and enzymatic sensors, are fairly insensitive to surface fouling because the charge-transduction mechanisms are local to the electrode surface. However, the EAB sensors for nM analytes are also the least commercially developed. EAB sensors work well for large molecules and proteins because of greater sites for unique hydrophilic/hydrophobic interactions between the analyte and aptamer. Conversely, using this same rationale, aptamers often lack the affinity needed to detect low concentrations, particulary when the target is a small molecule (fewer sites for hydrophilic/hydrophobic interactions). This is unfortunate, as hormones are an important class of analytes, and some hormones are so small and lipophilic that they readily diffuse into sweat with near 1:1 correlations with their unbound concentrations in blood (see next section and [2]). Therefore, for low-concentration small molecules, the emerging field of molecularly-imprinted polymer sensors might find a particularly strong niche in sweat biosensing [19].

Some additional comments regarding EAB sensors are merited, and may have context that relates to other sweat sensor modalities as well. For small molecules, EAB sensors can rapidly release (seconds or faster) the analyte as concentration decreases, and hence in many cases no sensor regeneration is needed. For larger analytes such as proteins, if rapid measurements are needed then some sort of sensor regeneration scheme is required. However, most protein concentrations change very slowly in the body, and do not need to be measured faster than periods of hours. Therefore, for some proteins a very slow self-regeneration of the EAB sensor may be acceptable. Another potential issue, is that both pH and salinity change significantly with sweat generation rate. The extent to which EAB signaling might be altered by physiologically-driven changes in the pH or ionic strength of sweat, has not yet been characterized. This said, the absence of ionizable groups that protonate over the 4.5 to 7 pH range of sweat suggests that the former is unlikely to be a hurdle. The latter issue (salinity) may be more problematic as changes in ionic strength can alter the thermodynamics of the binding-induced conformational shift that drive EAB signaling. Typical levels, though, only range from 20 to 80 mM and can be easily measured with ion-selective electrodes to help correct for salinity-induced errors in the EAB signal.

- *Most large analytes (e.g. proteins) are filtered*

Large analytes such as proteins are filtered in sweat, which means they are highly diluted compared to blood concentrations. Exceptions include defensive proteins like dermcidin, but only because they are actively secreted into sweat [20]. To predict just how dilute proteins are in sweat, a speculative calculation is now provided. Dermci-

 © 2016 Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim

These are not the final page numbers!

din constitutes about 50% of the total protein content in sweat [20]. Albumin, which is ~60% of protein content in blood, is only 5% of the protein in sweat [20]. If the total protein content of sweat is only a few tenths of a mg/mL [21], Albumin only 5% of that, and albumin in plasma is ~40 mg/mL, then for at least the case of high sweat rates [20] sweat albumin is at least 1000X more dilute than in blood. It is therefore reasonable to expect, that a protein analyte at nM to pM concentrations in blood becomes a very difficult to measure pM to fM analyte in sweat. Fortunately, there are several bits of encouragement for sweat analysis of proteins:

(1) peer-reviewed papers show that *ratios* of proteins correlate between blood and sweat [7].
(2) many proteins change in concentration slowly (over periods of hours) and would therefore require less frequent sampling, opening up the possibility for microfluidic sample pre-concentration [22]. Pre-concentration for proteins can arguably be achieved using simple size-selective membrane techniques.
(3) unlike tears and saliva, sweat can be generated and collected at very low fluid generation rates, which could allow more time for proteins to enter into sweat, and therefore improve both total concentration and correlation with blood.

● *Further discussion on expected filtration trends in sweat, based on data available for saliva*

The comparison to saliva (point #3 above), is worthy of further discussion. At some point, a greater understanding of what analytes readily exist in sweat will be thoroughly reported in the literature. In the short term, for some hints, researchers can turn to the greater volume of published work that exists for saliva. A particularly exceptional review of salivary analytes has been published by Vining and colleagues [23]. The plots adapted in Fig. 5 from Vining et al. [23] show clear effects of analyte size and lipophilicity on analyte concentrations in saliva. For example, the analytes in Fig. 5a are all hydrophilic and show a clear size dependence on dilution. As another example, in Fig. 5b, DHEAS is hydrophilic and diluted whereas cortisol is equally as large but is lipophilic (membrane permeable) and therefore not diluted. Such evidence is further interesting to consider for monitoring of lipophilic analytes (DHEA) vs. their metabolites (DHEAS) which are most often hydrophilic. This could effect applications in pharmaceutical monitoring (e.g. monitor the drug vs. it's metabolite).

However, several cautionary statements must be made on comparison of sweat to saliva: (1) as stated previously, saliva generation rates can be much greater than sweat generation rates, so their analyte dilutions due to filtration cannot be directly compared; (2) many salivary analyte papers for large analytes should be ignored, as they detect analytes that partition into saliva from the gum crevices, which is unpredictable and which is obviously absent in the case of sweat; (3) enzymes and other routes

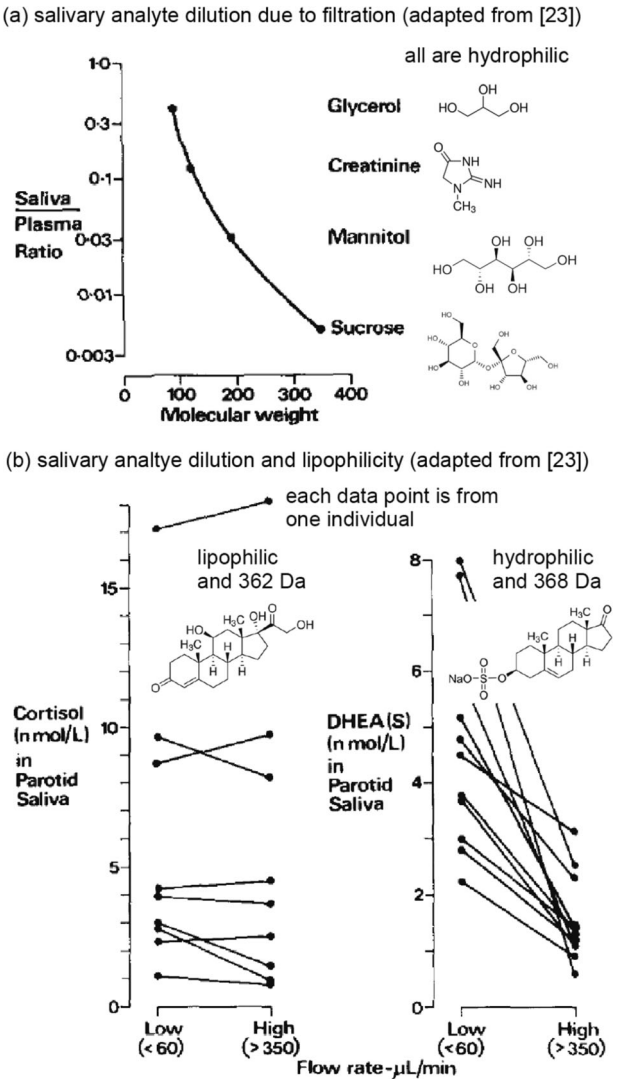


Fig. 5. Analyte dilution in saliva due to [a] hydrophilic analyte size and/or [b] lipophilicity vs. hydrophilicity (adapted from [23]). All of the above analytes are those that passively diffuse into saliva, and similar trends are possible for sweat as well.

for analyte degradation are often different between saliva (a digestive fluid) and sweat.

● *Some analyte concentrations only represent skin and/or the eccrine sweat gland itself, not the body*

Some analyte concentrations in sweat such as NaCl represent only active channels for secretion and recapture of analytes in the eccrine gland itself [2] (Table 1). As another example, Lactate content in sweat is a by-product of the local metabolism required to support sweat generation [2] (Table 1). This is not to say that metabolite correlation between sweat and blood is not possible, but it does suggest that proxy analytes or very low sweat generation rates may be needed to gain meaningful information for some metabolite analytes. Fortunately, concentrations of some 'Holy grail' analytes such as glucose seem to be fairly independent of sweat generation rate [6] and have been shown to trend with blood glucose [12]. How-

© 2016 Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim

Table 1. Brief summary of the present understanding of major analyte classes and comments on their partitioning into sweat. For some limited concentration data, please see reference [2].

Analyte class	Partitioning into sweat?
Electrolytes	Some are active (Na^+, Cl^-) and have no serum correlation, and some are passive (K^+) and are similar to blood levels.
Metabolites	Key metabolites such as lactate represent local sweat gland activity and metabolism (not blood levels). Some other metabolites may provide sweat-serum correlations.
Amino Acids	Certainly appear in sweat [24]. Molecular weights could be in range for filtration at some sweat rates (Fig. 5).
Small molecules	Lipophilic molecules such as cortisol can provide 1:1 correlations between sweat and serum (unbound fractions) whereas hydrophilic molecules may be filtered depending on size and sweat rate (Fig. 5).
Proteins	Undoubtedly filtered and dilute, but evidence shows ratios in sweat are equivalent to ratios in plasma [7]. Rarely need to be measured quickly, opening the possibility for pre-concentration [22] or other techniques to compensate for dilution.

ever glucose appears to be ~100X dilute in sweat [6,12] and its precise partitioning pathway into sweat [2] has not yet been confirmed. Other examples of analytes that are confounded by local activity include the previously-discussed defensive proteins such as dermcidin. Further examples may include analytes associated with inflammatory responses that are localized to the skin. For example, you would not want a sweat sensor to indicate a whole body inflammatory response when instead the only issue is that you are recovering from a mild sun-burn.

5 Two Key Conclusions circa 2016

Circa 2016, there has been a clear and rapid increase in both interest and technological advances for non-invasive analyte sensing in eccrine sweat. A greater understanding is also unfolding on individual analyte information available in sweat (Table 1). Furthermore, beyond measuring single analytes, it should be expected that over time more multi-analyte studies will publish which provide meaningful physiological correlations. A very recent example even links lung cancer to amino acid ratios in sweat [24]. *It is therefore the position of this author, that the most important conclusions that can be made at this time are two-fold:*

(1) the opportunity provided in eccrine sweat is clearly compelling compared to other non-invasive biofluids, and has the potential to enable a true performance leap for wearable technology.
(2) the many challenges for eccrine sweat are likely resolvable directly or indirectly through technological innovation. Importantly, circa 2016, the roadmap of requirements for addressing these challenges is now much more clear.

Acknowledgements

The author acknowledges financial support at the Univ. of Cincinnati provided by the National Science Foundation (Eager Grant # 1347725). The author highlights and acknowledges foundational and ongoing support from Air Force Research Labs 711th Human Performance Wing (Dr. Joshua Hagen, Dr. Rajesh Naik), including USAF contract # FA8650-15-C-6625. A large portion of this review, builds upon on a microfluidic model of the eccrine sweat gland [2], which was previously supported by the industrial members of the Center for Advanced Design and Manufacturing of Integrated Microfluidics (NSF I/UCRC award number IIP-1362165). The author would also like to acknowledge input and guidance by collaborators Prof. Gerald Kasting and Prof. Kevin Li in the Univ. of Cincinnati College of Pharmacy. In terms of disclosure of potential conflicts of interest, the author also acknowledges that he is a Chief Science Officer and co-founder of Eccrine Systems, Inc.

References

[1] J. Bandodkar, J. Wang, *Trends Biotechnol.* **2014**, *32*, 363–371.
[2] Z. Sonner, E. Wilder, J. Heikenfeld, G. Kasting, F. Beyette, D. Swaile, F. Sherman, J. Joyce, J. Hagen, N. Kelley-Loughnane, R. Naik, *Biomicrofluidics* **2015**, *9*, 031301.
[3] X. Huang, Y. Liu, K. Chen, W.-J. Shin, C.-J. Lu, G.-W. Kong, D. Patnaik, S.-H. Lee, J. Fajardo Cortes, J. A. Rogers, *Small*, **2014**, *10(15)*, 3083–3090.
[4] D. P. Rose et al., *IEEE Trans. Biomed. Eng.* **2015**, *62(6)*, 1457–1465.
[5] B. Schazmann, D. Morris, C. Slater, S. Beirne, C. Fay, R. Reuveny, N. Moyna, D. Diamond, *Anal. Methods*, **2010**, *2*, 342–348.
[6] W. Gao, S. Emaminejad, H. Y. Y. Nyein, S. Challa, K. Chen, A. Peck, H. M. Fahad, H. Ota, H. Shiraki, D. Kiriya, D.-H. Lien, G. A. Brooks, R. W. Davis, A. Javey, *Nature*, **2016**, *529*, 509–514.
[7] G. Cizza, A. Marques, F. Eskandari, I. Christie, S. Torvik, M. Silverman, T. Phillips E. Sternberg, *Biopsych.* **2008**, *64*, 907–911.
[8] B. R. Brunet, A. Barnes, R. Choo, P. Mura, H. Jones, M. Huestis, *Therapeutic Drug Monitoring* **2010**, *32(1)*, 40–49.
[9] A. Luchini, C. Fredolini, B. H. Espina, F. Meani, A. Reeder, S. Rucker, E. F. Petricoin, L. A. Liotta, *Curr. Mol. Med. Mar* **2010**, *10(2)*, 133–141.
[10] A. Mena-Bravo, L. de Castro, *J. Pharm. Biomed. Anal.* **2014**, *90*, 139–47.
[11] M. C. Desax et al., *Eur. J. Pediatrics* **2007**, *167(3)*, 299–304.
[12] T. C. Boysen, S. Yanagawa, F. Sato, K. Sato, *J. Appl. Physiol. Respir. Environ. Exerc. Physiol.* **1984**, *56(5)*, 1302–7.
[13] http://www.coresyte.com
[14] S. Derler, L.-C. Gerhardt, *Tribol. Lett.* **2012**, *45*, 1–27.
[15] L. Hou et al., *Lab. Chip* **2013**, *13*, 1868–1875.

 © 2016 Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim

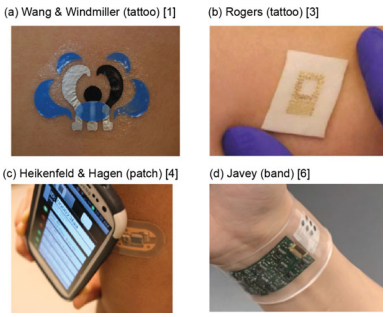
These are not the final page numbers! ↗↗

[16] L. Brunton, B. Chabner, B. Knollman , *Goodman and Gilman's The Pharmacological Basis of Therapeutics, 12th Ed.*, McGraw-Hill (2011).
[17] A. Lubin, K. Plaxco, *Acc. Chem. Res.* **2010**, *43 (4)*, 496–505.
[18] Private communication from Prof. Kevin Plaxco, Univ. California at Santa Barbara, 2015.
[19] A. Ethirajan, R. Thoelen, T. J. Cleij, P. Wagner, M. Peeters, F. J. Troost, B. van Grinsven, F. Horemans, J. Alenus, M. S. Murib, D. Keszthely, *Sensors and Actuators B* **2012**, *171–172*, 602–610.
[20] É. Csősz, G. Emri, G. Kalló, G. Tsaprailis, J. Tőzsér, *Journal of the European Academy of Dermatology and Venereology*, **2015**, *29(10)*, 2024–2031.
[21] M. Raiszadeh et al., *J. Proteome Res.* **2012**, *11(4)*, 2127–2139.
[22] B. Giordano, D. Burgi, S. Hart, A. Terray, *Analytica Chimica Acta* **2012**, *718*, 11–24.
[23] R. Vining, R. McGinley, *J. Steroid Biochem.* **1987**, *27*, 81–94.
[24] M. Calderón-Santiago, F. Priego-Capote, N. Turck, X. Robin, B. Jurado-Gámez, J. Sanchez, M. Castro, *Anal. Biochem.* **2015**, *407*, 5381–5392.

Received: January 11, 2016
Accepted: February 16, 2016
Published online: ■■ ▮▮, 0000

SPECIAL ISSUE

© 2016 Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim



*J. Heikenfeld**

■■–■■

Non-invasive Analyte Access and Sensing through Eccrine Sweat: Challenges and Outlook circa 2016

© 2016 Wiley-VCH Verlag GmbH & Co. KGaA, Weinheim