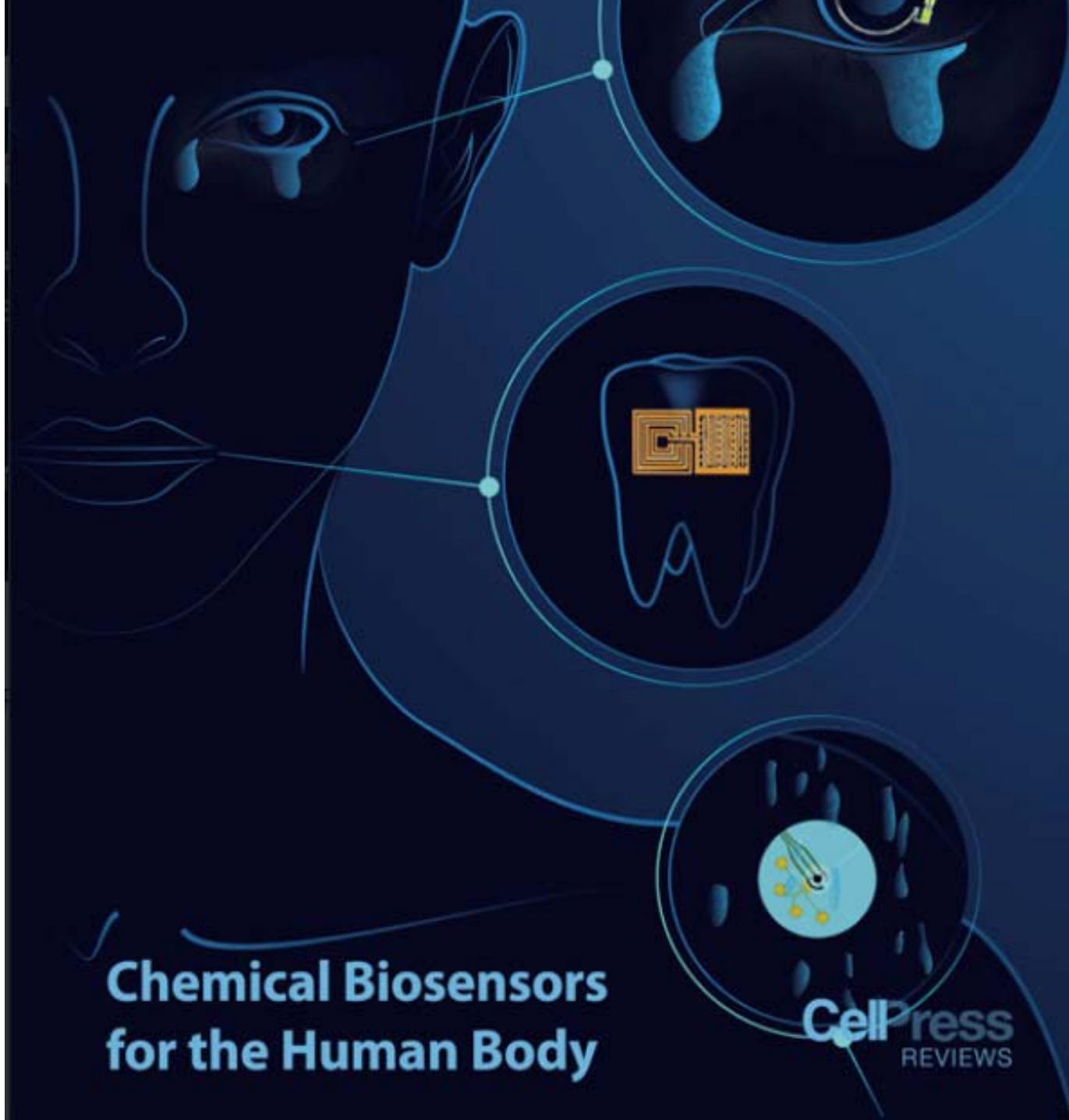


# Trends in Chemistry

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**Chemical Biosensors  
for the Human Body**

**CellPress**  
REVIEWS

## Review

## Body-Interfaced Chemical Sensors for Noninvasive Monitoring and Analysis of Biofluids

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**Body-interfaced sensors have received tremendous attention due to a broad and diverse collection of potential applications, from monitoring of health status to managing and preventing disease conditions. In this review article, we highlight the emerging sensing modalities and advanced materials/device designs in body-integrated flexible platforms capable of capturing and analyzing biofluids. In particular, we discuss different chemical sensors and biosensors for real-time noninvasive monitoring and analysis of metabolites and electrolytes in sweat, tears, saliva, and interstitial fluid. Finally, we show several techniques that could be integrated with body-interfaced sensors for data acquisition, signal processing, display, and communication.**

## Body-Interfaced Chemical Sensors

Interest in body-interfaced electronic biosensors is growing rapidly due to a diversity of potential applications, including monitoring of health status, tracking of athletic performance, and preventing disease conditions [1–4]. Although many research programs focus on physical and electrophysiological sensing [5–7], an important frontier is in systems capable of biochemical measurements, where chemistry plays a critically important role alongside device and systems engineering. Multimodal platforms that combine physical and chemical sensing together represent the ultimate goal, where the potential is for continuous, clinical-grade assessments of body processes outside of hospital or laboratory settings [1,8,9]. Emerging strategies in this area involve noninvasive measurements of chemical biomarkers using body-integrated electronic sensors configured to collect, capture, manipulate, and analyze underexplored classes of biofluids, including sweat [10–12], tears [13–15], saliva [16–18], and **interstitial fluid (ISF)** (see Glossary) [19–21].

## Sweat Sensors

Sweat is an interesting and relatively underexplored type of biofluid, with an abundance of chemical biomarkers, including electrolytes and metabolites [20,22,23] and xenobiotics, such as alcohol and drugs [24,25]. Compared with traditional biofluids such as blood, sweat is of interest due to its ability to be collected in a completely noninvasive fashion [26]. In certain cases, the concentrations of biomarkers in sweat can be correlated to those in the traditional biofluids and/or they, themselves, can provide established information relevant to health status. For example, some data suggest that sweat glucose is correlated, at least approximately and in certain cases, to blood glucose for diabetes screening and management [4,27–29]; sweat chloride is an indicator for cystic fibrosis [29–32]; and abnormally high sweat urea concentration suggests kidney failure [33–35]. Moreover, monitoring of alcohol concentrations in biofluids can be achieved without invasive blood sampling, as a step toward real-time measurements of alcohol intoxication [24]. In general, sweat rate and sweat loss are also important because the concentrations of certain

## Highlights

Body-interfaced chemical sensors allow for continuous, noninvasive collection, storage, and analysis of underexplored classes of biofluids, including sweat, tears, saliva, and interstitial fluid.

Different sensing modalities and advanced materials/device designs improve the stability, selectivity, and sensitivity of these body-interfaced sensors.

Multimodal platforms that combine physical and chemical sensing capabilities with wireless functionality are promising for precise, clinical-grade assessments of health status and disease conditions outside of hospital or laboratory settings.

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biomarkers can vary depending on these parameters. Loss itself is an important quantity that influences athletic performance and serves as a metric to maintain proper hydration levels [12,36]. Despite the potential as a vehicle to quantify biochemical signatures of health, reliable analysis requires schemes for collecting microliter volumes of sweat in ways that avoid contamination from dirt, debris, and oils from the skin and alterations in concentration that arise from partial evaporation. Real-time sampling in a time-sequential fashion that also simultaneously quantifies the rate of sweating and allows for *in situ* chemical analysis are additional desired features. Results of recent research form the foundations of various flexible and/or stretchable, skin-integrated platforms that address many of these challenges, to allow for effective sweat extraction [12,29,37], collection [38–40], and sensing [1,8–12,39,41–43]. These advances exploit fabrication techniques that combine traditional methods from the semiconductor industry, such as photolithography and selective etching, with those of soft lithography, such as molding and casting, to yield microfluidic systems that enable time-sequential biofluids analysis when integrated with colorimetric or fluorometric assays for optical sensing or functionalized electrodes for electrochemical sensing.

### Sweat Collection

Conventional methods for sweat generation involve physical activity [12,26], exposure to heat/humidity [12,44], or iontophoresis [12,29]. Additionally, recent work reports that warm showering or bathing can induce significant sweating [37]. Collection is the first step in analysis of sweat dynamics and chemical composition. Absorbent pads or sponges represent the most traditional vehicles for collection, where extraction allows for *ex situ* analysis using laboratory instrumentation [45,46]. More advanced approaches rely on hydrogel- [47], paper- [21,48], or textile-based materials [49] that route sweat toward an electrochemical sensing module for *in situ* measurement(s). Recent development of soft, skin compatible microfluidic technologies based on poly(dimethylsiloxane) (PDMS) [9,38] enables precise capture, routing, and chemical analysis in a way that can leverage some of the most sophisticated methods from the lab-on-a-chip community, including the ability for time-sequenced capture and storage of minute volumes of sweat in isolated chambers with negligible mixing or cross-contamination. Optimized versions of these systems use the elastomer poly (styrene-isoprene-styrene) (SIS) to allow sweat sampling under extreme conditions, including aquatic and arid environments, enabled by the small water permeability of SIS [26]. Nanofluidic films offer capabilities for sweat sampling in the nanoliter regime [50].

### Metabolites: Enzymatic Sensors

Most research on selective sensing of metabolites relies on enzymes (e.g., oxidoreductase [3,10,25,37,51]) on various substrates, such as fabrics [52], plastics [8], or elastomers [9]. The associated sensing modalities include **amperometry** [3], the piezoelectric effect [51], and colorimetry [37,53]. Figure 1A summarizes a route to metabolic detection in a skin-mounted device that leverages an enzyme functionalized amperometric sensor (range: glucose, 50  $\mu$ M–10 mM; lactate, 4–20 mM) [3]. In this example, glucose oxidase (GOx) and lactate oxidase (LOx) enzymes bond to a flexible, screen-printed electrode of Prussian blue (PB). The resulting enzymatic reaction produces  $H_2O_2$ , which is selectively reduced at the PB electrode to generate a current signal (Figure 1B,C). Piezoelectric detection of the  $H_2O_2$  byproduct without the need for complex electronics, as required by amperometric systems, represents an interesting, self-powered means for metabolic sensing (Figure 1D, range: glucose, 20–200  $\mu$ M; lactate, 2–20 mM) [51]. The enzymes, LOx, GOx, urease, and uricase, are coated on their corresponding sensing unit, which is composed of interdigital Ti electrodes with ZnO nanowires aligned in between. The LOx/ZnO nanowire generates a piezoelectric voltage in pure water under an applied force, but when in contact with lactate, the output voltage decreases

### Glossary

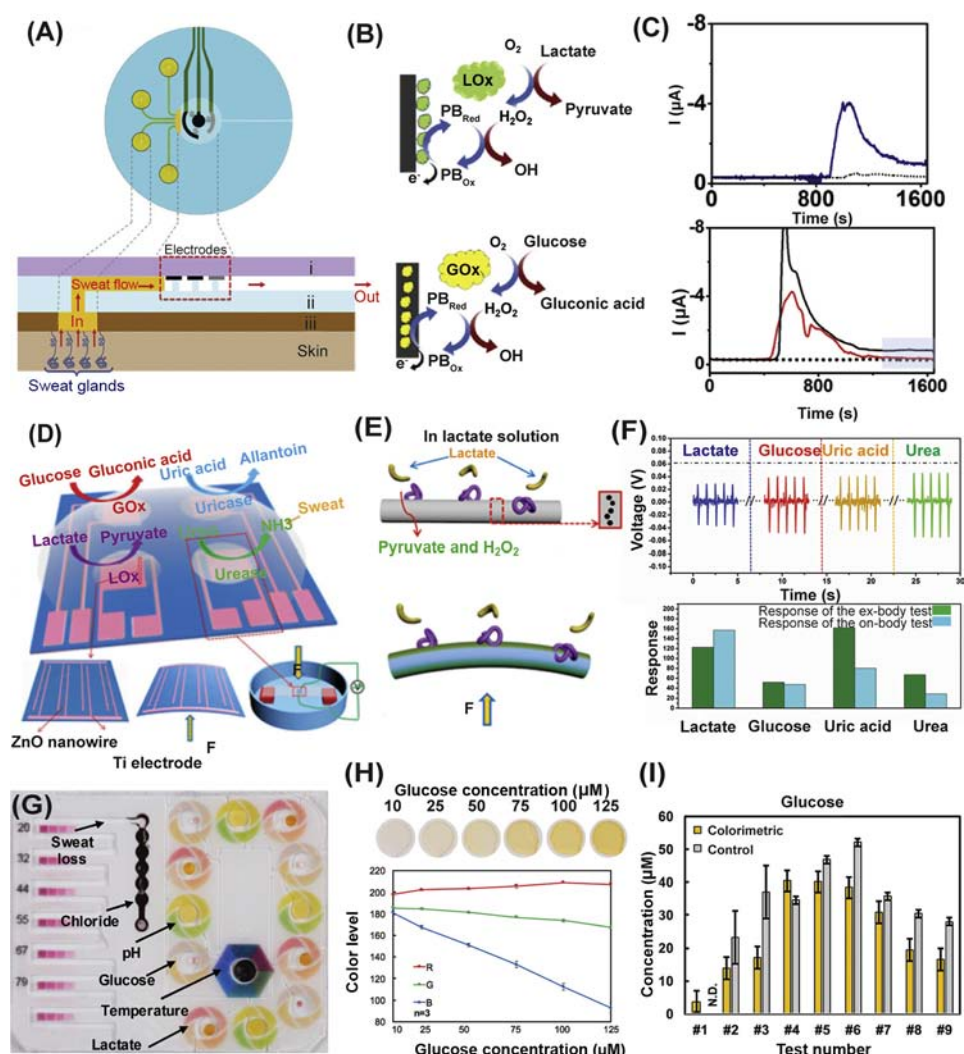
**Amperometry:** an electrochemical technique that measures current generated from the oxidation or reduction of an electroactive analyte in a chemical reaction.

**Interstitial fluid (ISF):** a solution that bathes and surrounds the cells.

**Potentiometry:** a zero-current technique that measures the potential appearing between the working electrode and the reference electrode in an electrochemical cell.

**Stripping voltammetric technique:** a voltammetric method that involves (i) preconcentration of the analyte of interest on the electrode surface and (ii) selective oxidation during the stripping step.

**Reverse iontophoresis:** a technique that involves passing a current across two electrodes applied to the skin. This causes electro-osmotic flow of the metabolites from the subcutaneous layer to surface of the skin.



**Figure 1. Enzymatic Sensors for Metabolites in Sweat.** (A) Schematic illustration of a skin-interfaced, microfluidic device for sweat collection in top-down and cross-sectional views [3]. (B) Schematic representation of enzyme-based amperometric sensing mechanisms for (top) lactate and (bottom) glucose. (C) Continuous (top) lactate and (bottom) glucose sensing. (D) Schematic illustration of a piezoelectric sensing unit for lactate, glucose, uric acid, and urea [51]. (E) Schematic illustration of a  $\text{LOx}/\text{ZnO}$  nanowire in lactate aqueous solution without applied deformation (top) and the piezoelectric output under applied deformation (bottom). (F) Piezoelectric output of four piezo-biosensing units while running at a speed of 12 km/h for 30 min (top) and comparison of response between *ex situ* (ex-body) and *in situ* (on-body) tests. (G) Top view illustration of soft, flexible microfluidic devices for colorimetric analysis of sweat [53]. (H) Optical images of color development as a function of concentration (top) and color level for each color (bottom) of glucose. (I) Sweat glucose concentration from colorimetric methods and control tests for *in situ* trials. Abbreviations:  $\text{GOx}$ , glucose oxidase;  $\text{LOx}$ , lactate oxidase; PB, Prussian blue.

due to the piezo-screening effect of  $\text{H}^+$  and  $\text{e}^-$ , where  $\text{H}^+$  and  $\text{e}^-$  are generated from  $\text{H}_2\text{O}_2$ , one product of the enzymatic reaction (Figure 1E,F). Colorimetry is attractive due to its ability to operate without an external supply of power and to enable readout via semiquantitative visual evaluation or quantitative color extraction from digital images. Recent work shows that colorimetric analysis of glucose and lactate is possible with the assay directly immobilized onto the microfluidic platform, instead of paper substrates, to improve the uniformity of color development and temporal



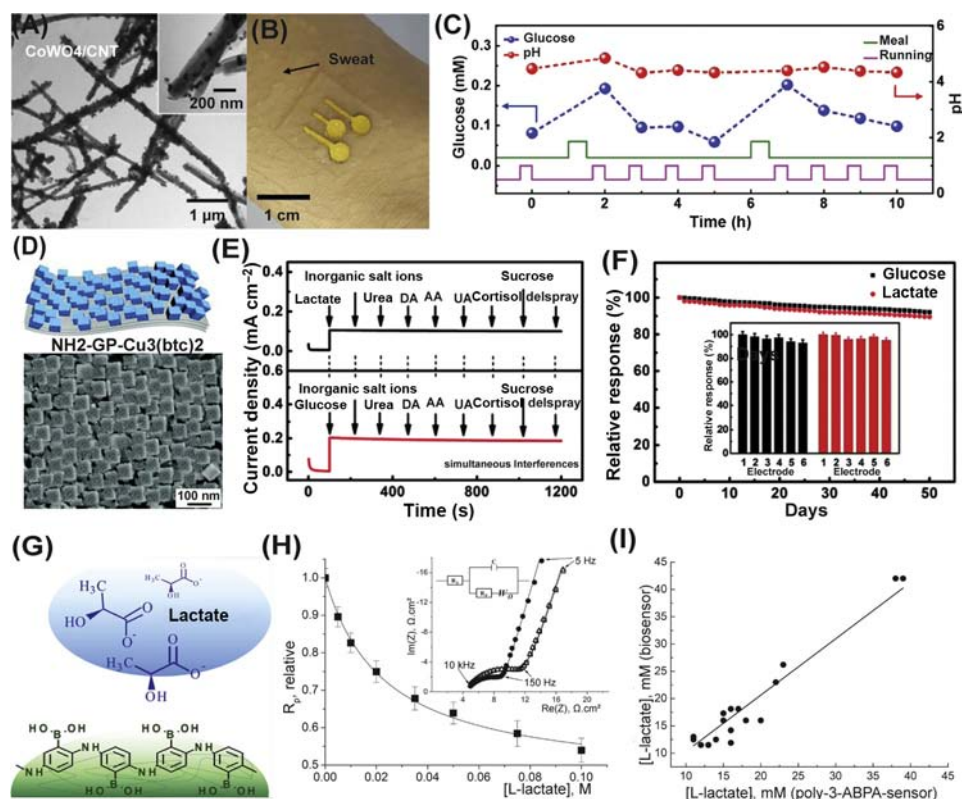
analysis (Figure 1G, range: glucose, 10–125  $\mu\text{M}$ ; lactate, 5–20 mM) [53]. Well-tuned reference color markers enable accurate analysis under various light conditions. The byproduct  $\text{H}_2\text{O}_2$  from the enzymatic reaction reacts with substrate dye, resulting in a change in color that corresponds to the concentration of analyte (Figure 1H). This type of colorimetric analysis, performed *in situ* in soft, skin-interfaced microfluidic platforms, yields results comparable with those obtained using conventional methods (Figure 1I).

#### Metabolites: Nonenzymatic Sensors

Disadvantages of enzymatic sensors include denaturation of enzymes during storage/long-term use [54], reduced activity due to immobilization [55], and high sensitivity toward variations in pH, ionic strength, and temperature. Effects of poisoning species represent additional possible complications. Recent demonstrations of nonenzymatic sweat sensors for glucose and lactate suggest that synthetic receptors can provide the basis of sensing characteristics that rival those of enzyme-based sensors in ways that bypass many of the disadvantages listed above. One widely employed route involves selective oxidation of glucose in alkaline media [56], adapted for use with sweat, which is a slightly acidic biofluid [57]. The approach uses a multistep detection process in which  $-2.0\text{ V}$  applied for 20 s to a flexible gold-based sensor generates an alkaline condition ( $\sim\text{pH } 11$ ) irrespective of the pH of sweat [58]. The following step involves the application of a bias of  $0.2\text{ V}$  for 5 s for glucose determination (range: 30–1100  $\mu\text{M}$ ; sensitivity:  $114\text{ }\mu\text{A mM}^{-1}\text{cm}^{-2}$ ), and finally a potential of  $1.0\text{ V}$  for 2 s to clean the electrode surface, thereby completing the process. This type of sensor exhibits excellent reproducibility and stability. Reliance on inorganic catalysts can circumvent the need for such multistep detection protocols to allow direct, selective detection of metabolites. For example, cobalt wolframate-carbon nanotube nano-composites applied to gold nanosheet electrodes allow for selective electrooxidation of glucose (Figure 2A) [59]. Integration of such nano-composite electrodes with silicone substrates allows irritation-free mounting on the skin for real-time monitoring of glucose in sweat (range: up to at least 300  $\mu\text{M}$ ; sensitivity:  $10.98\text{ }\mu\text{A mM}^{-1}\text{cm}^{-2}$ ) (Figure 2B,C). Another related approach leverages the excellent catalytic and modular features of metal-organic frameworks (MOFs) [60]. In one example, synthesis of HKUST-1 (MOF comprising of copper nodes with 1,3,5-benzenetricarboxylic acid struts) nanocubes followed by their 2D self-assembly onto an amino-functionalized graphene paper at water-oil interface yields a flexible glucose and lactate sensor (Figure 2D) [61]. The selective detection of lactate can be attributed to the host-guest interactions resulting in displacement of the coordinated water molecules and exposure of  $\text{Cu}^{2+}$  sites for selective oxidation of lactate at  $-0.1\text{ V}$ . The sensor achieves its selectivity toward glucose due to its catalytic oxidation at the copper nodes at  $+0.65\text{ V}$ . Studies of interference (Figure 2E) and stability (Figure 2F) along with human trials illustrate the potential of this method. Another nonenzymatic path leverages molecularly imprinted polymers (MIPs) comprising templates in poly(3-aminophenylboronic acid) for sensing sweat lactate (Figure 2G) [62]. Phenylboronic acid serves as the building block due to its binding selectivity to compounds that possess 1,2- or 1,3-diol functionalities. The electrostatic binding of the lactate to the template via host-guest chemistry results in variations in electrode-sample impedance that can be detected via impedance spectroscopy (Figure 2H). Systematic benchtop and human studies demonstrate the feasibility of such an approach for enzyme-free sweat analysis with high correlation to standard enzyme-based assays (Figure 2I).

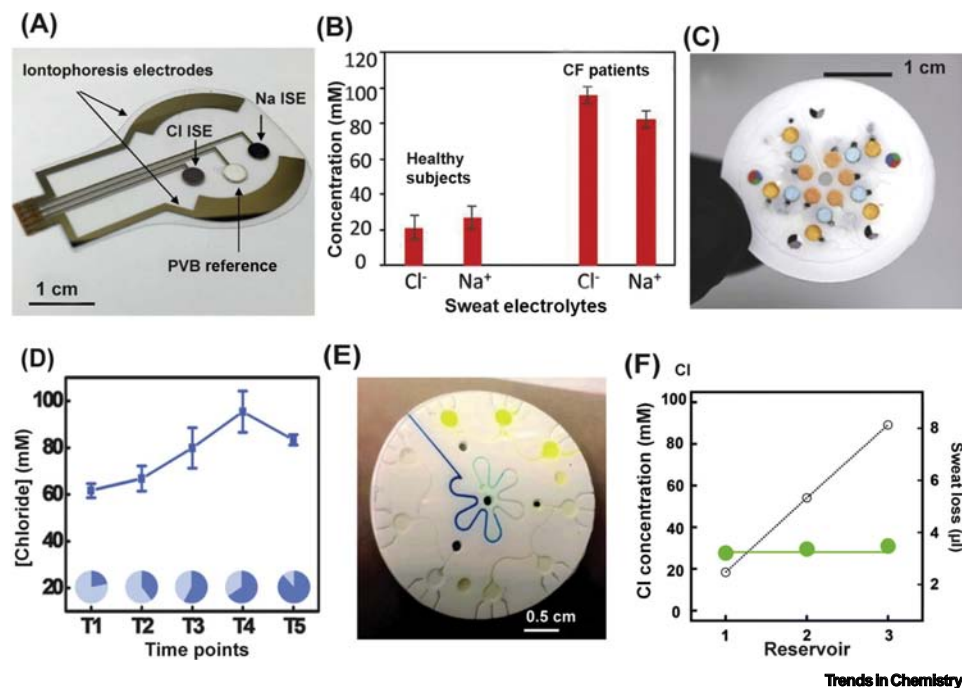
#### Electrolytes

**Potentiometry** is the most common method for electrolyte analysis due to the availability of well-developed ion-selective electrodes (ISEs) for  $\text{Na}^+$  [1,29],  $\text{K}^+$  [1],  $\text{Ca}^{2+}$  [41],  $\text{H}^+$  [41], ammonium [63], and  $\text{Cl}^-$  [29]. Optical approaches, such as colorimetry and fluorometry, can be used for



**Figure 2. Nonenzymatic Sensors of Sweat Biochemistry.** (A) Scanning-electron micrograph (SEM) of a CoW<sub>4</sub>/CNT nanocomposite [59]. (B) Image of a soft CoW<sub>4</sub>/CNT-based skin-integrated glucose sensor. (C) Correlation of sweat glucose and blood levels as reported by the skin-integrated device and blood meter, respectively. (D) Schematic illustration and SEM image of a 2D assembled metal-organic framework-based (MOF-based) flexible glucose sensor [61]. Effect of (E) interference and (F) stability on the MOF-based sensor. (G) Schematic illustration of a molecularly imprinted polymer-based (MIP-based) lactate sensor [62]. (H) Calibration plot and (I) correlation of MIP-based sensor as compared with conventional enzyme-based lactate sensor. Abbreviations: AA, ascorbic acid; DA, dopamine; UA, dopamine.

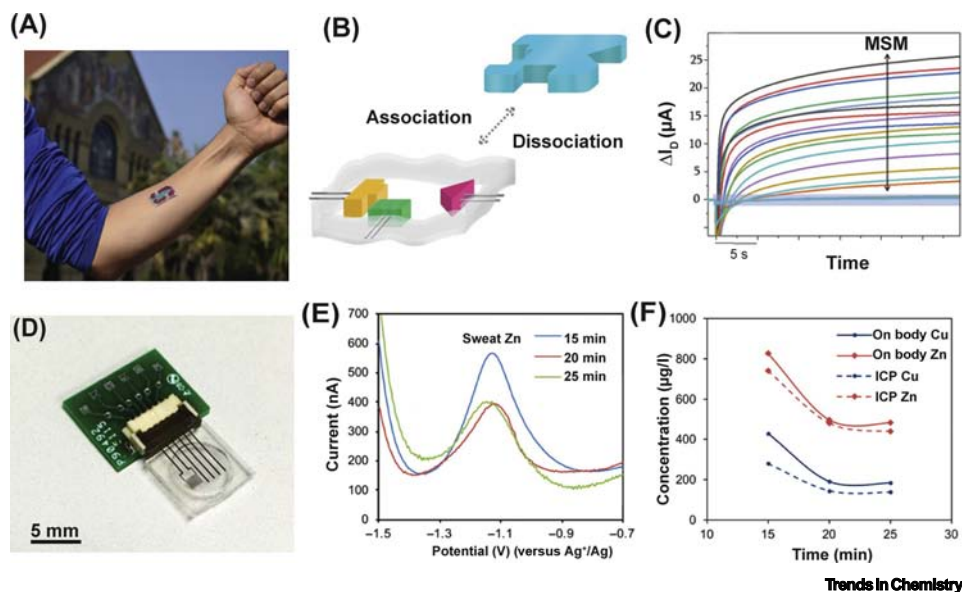
analysis of Na<sup>+</sup> [64], H<sup>+</sup> [9,10,53], and Cl<sup>-</sup> in sweat [9,10,53,64]. Figure 3A shows a representative potentiometric example for Na<sup>+</sup> and Cl<sup>-</sup> in a platform that also provides iontophoresis for localized sweat stimulation [29]. Here, the sodium ionophore X-based ISEs and Ag/AgCl electrodes serve as indicator electrodes for Na<sup>+</sup> and Cl<sup>-</sup> detection with polyvinyl butyral-coated electrodes using saturated Cl<sup>-</sup> as the common reference electrode. Real-time *in situ* measurements of healthy subjects and cystic fibrosis patients suggests the potential of this technology for cystic fibrosis screening (Figure 3B). A recent colorimetric approach provides an alternative that avoids the need for electronics for readout (Figure 3C) [39]. The principle of this assay involves the competitive chelation of Hg<sup>2+</sup> and Fe<sup>2+</sup> to the chelator, 2,4,6-tris(2-pyridyl)-s-triazine (TPTZ), where Hg(TPTZ)<sub>2</sub> is colorless and Fe(TPTZ)<sub>2</sub> has an absorption peak around 595 nm. Cl<sup>-</sup> reacts with Hg(TPTZ)<sub>2</sub> to form HgCl<sub>2</sub>, release TPTZ, and produce additional Fe(TPTZ)<sub>2</sub> such that the compensatory blue color level changes with concentration of Cl<sup>-</sup>. The chloride levels of healthy subjects evaluated in this manner fall into a reasonable range (Figure 3D). Na<sup>+</sup> and Cl<sup>-</sup> sensing can also be achieved by fluorometric assays (Figure 3E) [64], where CoroNa green and lucigenin serve as selective fluorescent probes and a smartphone-based optical setup offers means for readout. Field studies show that fluorometric results compare well with those from conventional assays, within the normal range (Figure 3F).



**Figure 3. Electrolyte Sensors for Sweat.** (A) Image of electrodes for sensing Na<sup>+</sup> and Cl<sup>-</sup> in sweat [29]. (B) Comparison of sweat electrolyte concentration between six healthy subjects and three CF patients. (C) Optical image of a chrono-sampling microfluidic device for sweat chloride monitoring [39]. (D) Sweat chloride concentrations of one subject at five time points. (E) Optical images of the fluorescent device for Na<sup>+</sup> and Cl<sup>-</sup> sensing after sweat collection [64]. (F) Calculated concentrations of sweat chloride from a fluorescent device. Abbreviations: CF, cystic fibrosis; ISE, ion-selective electrode; PVB, polyvinyl butyral.

### Other Analytes

Hormones (cortisol [65]), exogenous drugs (caffeine [24]), and minerals (zinc [42,48,64] and copper [42]) represent biochemicals of additional interest. A recent study leverages MIPs constructed using methacrylate building blocks as cortisol selective membranes coated over poly(3,4-ethylenedioxythiophene) polystyrene sulfonate (PEDOT:PSS)-based electrochemical transistors (Figure 4A,B) [65]. Host-guest interactions of cortisol with the templated MIP change the conductivity of the PEDOT:PSS-based channel and allow for easy detection with a detection limit of 10 nM (Figure 4C). Detection of caffeine [24], zinc [42,48], and copper [42] can be accomplished via **stripping voltammetric techniques** where the characteristic peaks represent oxidation of these analytes. For minerals, multistep detection protocols involve deposition of the targeted mineral on tetrafluoroethyleneperfluoro-3,6-dioxo-4-methyl-7-octenesulfonic acid copolymer (Nafion) coatings on bismuth modified carbon or gold electrodes followed by their stripping (oxidation). The results yield oxidation waveforms unique to a particular analyte (Figure 4D,F) [42]. Detection of caffeine follows a similar scheme, but without the need for the deposition step. Direct, on-skin fluorometry represents an attractive alternative to the electrochemical routes for heavy metal detection, as demonstrated recently with a zinc sensor [64]. The device includes a soft microfluidic construct with a series of interconnected microchambers that house commercially acquired zinc fluorometric solid-state assays. Strategically located reference chambers include a fixed quantity of a fluorophore (Rhodamine chloride 110) dispersed in 1-ethyl-3-methylimidazolium ethyl sulfate for robust, comparative intensity analysis using a smartphone-based fluorometric imaging module.



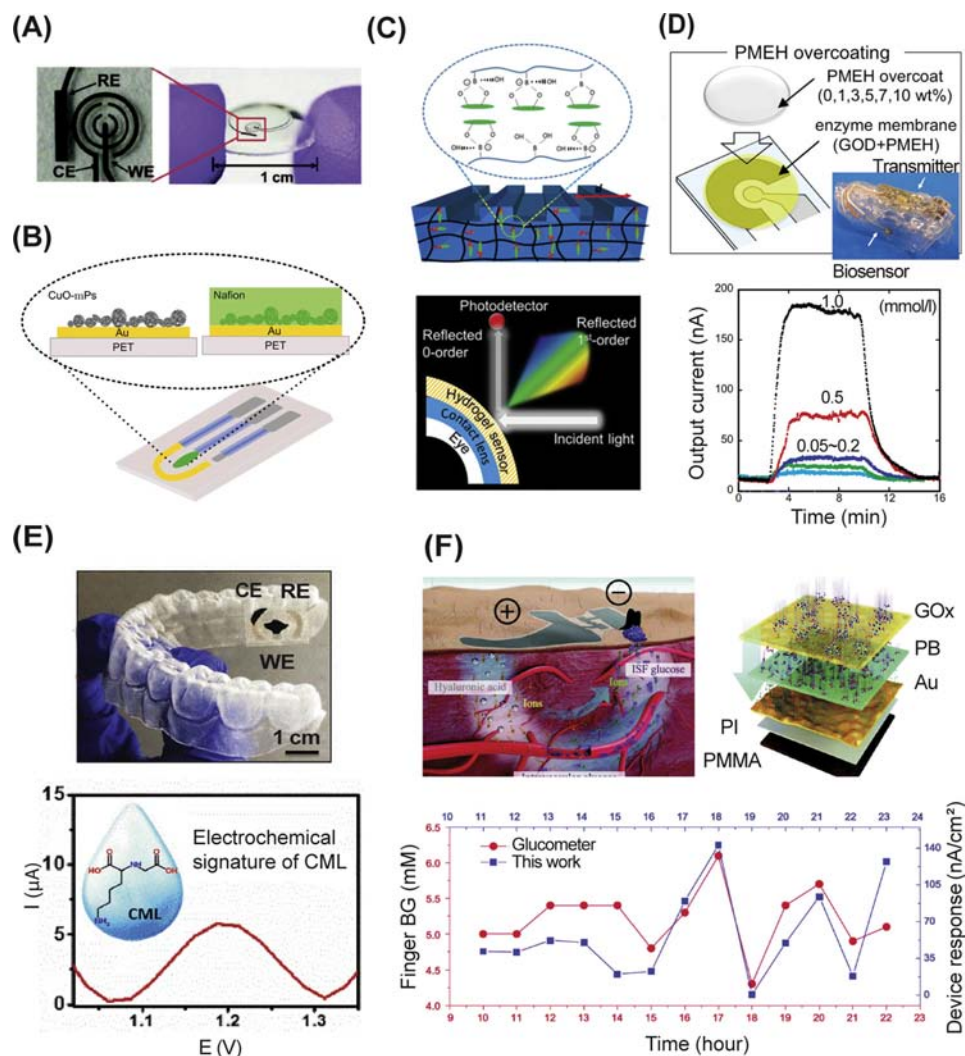
**Figure 4. Sensors for Other Chemicals in Sweat.** (A) Image of a skin-integrated cortisol sensor [65]. (B) Schematic illustration of the sensing mechanism of a molecularly imprinted polymer (MIP)-based cortisol sensor. (C) Linear variation in drain current as a function of cortisol concentration. (D) Image of a mineral-sensing platform [42]. (E) Voltammetric signals of Zn levels detected in sweat. (F) Correlation of zinc and copper concentrations as measured via a skin-integrated sweat sensor and inductively coupled plasma mass spectrometry (ICP-MS). Abbreviation: MSM, molecularly selective membrane.

## Chemical Sensors for Other Biofluids

### Tear Sensors

Tear, saliva, and ISF represent additional classes of biofluids that can be captured and analyzed using body-integrated devices [66,67]. Worn mainly for vision correction and cosmetic reasons, contact lenses continuously interface with tear fluids and thus provide a unique platform for real-time diagnostics. Tear contains salts, proteins, enzymes, and lipids that reveal information on ocular conditions and systemic disorders [68]. Certain studies suggest some correlations between the concentration of glucose in tears and that in blood (0.15 mM tear glucose corresponding to 5 mM blood glucose) [13,69]. Figure 5A shows an early-stage contact lens integrated with an amperometric glucose sensor, suggesting the possibility of *in situ* health monitoring [70]. A titania sol-gel film immobilizes GOx, and a Nafion permselective film reduces potential interference from ascorbic acid, lactate, and urea present in the tear. The sensor shows good linearity across a range of glucose concentrations in tear (0.1–0.6 mM) with minimal interference. The limited reproducibility and long-term stability of enzyme-based methods can be circumvented using enzyme-free methods [71]. Figure 5B shows a nonenzymatic sensor based on inkjet printed electrodes on a flexible polyethylene terephthalate substrate [71]. A CuO-modified electrode detects glucose (linear range of 0.003–0.7 mM) by electro-catalytic oxidation and Nafion serves as an immobilizing layer to firmly anchor the particles. The device exhibits a stable response for more than 1 week. Optical sensing represents another option for tear sensors. Recent work shows that fluorescence-based sensors and photonic-based sensing structures can serve as the basis for reusable glucose monitoring systems [15,72,73]. Figure 5C highlights a one-dimensional photonic structure with a periodicity of 1.6 μm imprinted on a phenylboronic acid-functionalized hydrogel film [74]. The volumetric change due to the binding of glucose alters the periodicity of the structure, thereby changing the diffraction properties. The periodicity depends in a systematic way on glucose concentration across concentrations from 0 to 50 mM. The sensor has a response time of 3 s and a saturation period of 4 min, with a sensitivity of 12 nm mM<sup>-1</sup>.





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**Figure 5. Chemical Sensors for Tear, Saliva, and Interstitial Fluid (ISF).** (A) Soft contact lens integrated with an amperometric glucose sensor [70]. (B) Schematic illustration of a nonenzymatic glucose sensor [71]. (C) Sensing principle of a photonic structure sensor (top). Schematic illustration of the measurement setup (bottom) [74]. (D) Mouthguard platform integrated with a saliva glucose sensor that uses GOx entrapped in Poly(MPC-co-EHMA) (PMEH) to enhance the sensitivity [18]. (E) Photograph of a customized mouthguard with an *N*-carboxymethyl-lysine (CML) sensor (top) [77]. Electrochemical signature of the CML as recorded by the differential pulse voltammetry (DPV) technique (bottom). (F) Schematic illustration of an ISF glucose monitoring system with a paper battery-powered electrochemical sensor with twin channels (ETC) (top) [21]. The sensor consists of multilayered structures. Comparison of hourly glucose monitoring during a 1-day period with a glucometer (red) and ETC (blue) (bottom). Abbreviations: BG, blood glucose; CE, counter electrode; GOx, glucose oxidase; PB, Prussian blue; PET, polyethylene terephthalate; PMMA, poly(methyl methacrylate); PI, polyimide; RE, reference electrode; WE, working electrode.

## Saliva Sensors

As many biomarkers in saliva pass directly from the bloodstream via paracellular or transcellular pathways, this class of biofluid represents another attractive option [66]. Antibodies in saliva are useful tools for diagnosing diseases such as HIV and intestinal infection, and certain proteins and mRNA biomarkers can be used to identify cancers [17]. With growing interest in saliva sensing, different oral platforms have been developed, with examples in sensing pH, fluoride, bacteria, uric acid, and glucose [16,18,75]. Figure 5D shows a detachable ‘cavitas sensor’ on a

mouthguard platform [18]. The salivary sensor consists of Pt and Ag/AgCl electrodes as a support with an enzyme membrane. An overcoat of poly(MPC-co-EHMA) (PMEH) on the sensing area enhances enzyme entrapment. The sensor operates over a range of 0.05–1 mmol/l for glucose, to encompass the range of concentrations in human saliva. The device further demonstrates stable and real-time monitoring (>5 h) using a phantom jaw and an open-loop artificial saliva injection system. Advanced glycation end-products, such as *N*-carboxymethyl-lysine (CML), are correlated with oxidative stress and long-term damage to proteins in ageing, atherosclerotic plaques, and diabetes [76,77]. Recent work reports printed, disposable sensors [working electrode (WE) and counter electrode (CE): carbon; reference electrode (RE): printed Ag/AgCl ink] that can be easily attached to the mouthguard (Figure 5E) for detection of CML via differential pulse voltammetry screening [77]. The CML concentration correlates well with current output (over the range of 2.45  $\mu$ M–12.24 mM), even in the presence of intrinsic and extrinsic interferences. This technology can apply to the detection of various salivary biomarkers, potentially providing comprehensive, useful real-time information regarding health condition and chronic disease states.

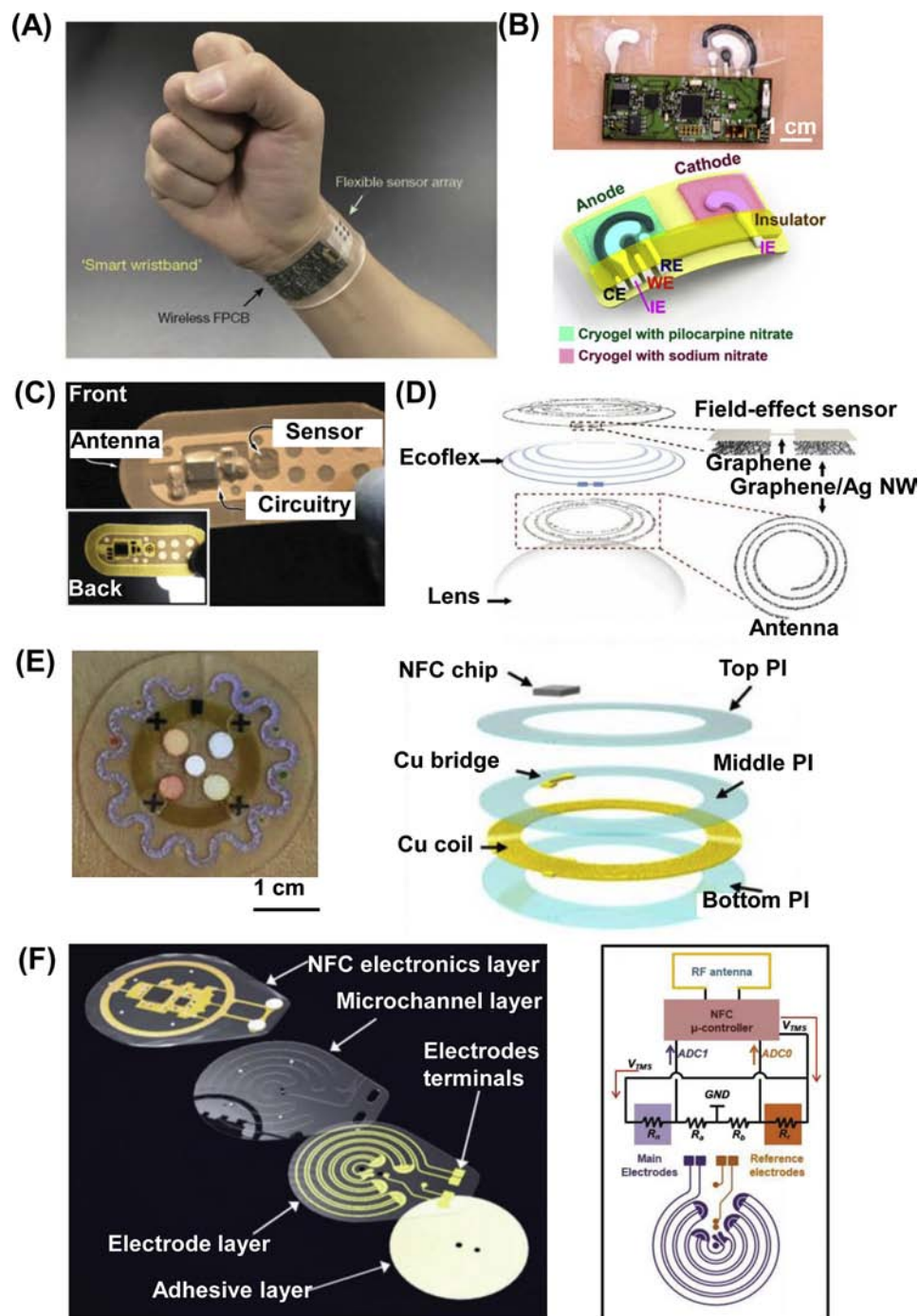
### ISF Sensors

The composition of the ISF is similar to blood in terms of small molecules such as salts, ethanol, glucose, and proteins [78]. Noninvasive ISF monitoring systems rely mainly on **reverse iontophoresis**, a process that involves application of a potential between two electrodes on the skin [19,78,79]. Figure 5F shows an advanced ISF glucose monitoring system with paper, battery-powered electrochemical twin channels [21]. When activated, high-density positively charged hyaluronic acid (HA) repels into the ISF under the anode; the extra HA (positively charged) raises the ISF osmotic pressure and thus promotes intravascular blood glucose transport into the ISF. The increased glucose concentration in the ISF greatly improves the blood–ISF glucose correlation. The biosensor consists of multilayered structures including poly(methyl methacrylate), polyimide, a deposited Au thin film, an electrochemically deposited transducer layer, and a GOx immobilization layer. As validated in human subjects, this noninvasive measurement shows a high degree of correlation coefficient (>0.9) to clinically measured blood glucose levels, typically with a 1-hour time lag in the ISF glucose concentration.

For tear sensors, difficulties in accurate noninvasive monitoring can arise in subjects with dry eye syndrome and in instances of reflex tearing due to emotional or environmental reasons [15]. Modulation in saliva composition due to food, bacterial growth, and/or dry mouth can lead to artifacts [20]. In general, the time lag for blood analytes to reflect in these biofluids represents an overarching issue that must be addressed explicitly. More generally, establishing correlations between the chemistry of blood and that of other body fluids will require extensive human subject studies.

### Wireless Systems for Chemical Sensors

For practical use, sensors of the type described in previous sections must be paired with electronic components for data acquisition, signal processing, display, and communication. Various strategies and designs are available for capabilities wireless operation, where a key consideration is on low power consumption and soft, skin-compatible mechanics. Figure 6A shows a fully integrated sweat sensor system for multiplexed *in situ* analysis [1]. The platform contains an analog front-end to condition the signal, an analog-to-digital converter, and a preprogrammed microcontroller for calibration. A Bluetooth transmission component relays the measurement results to a cell phone for information display and storage. This work merges skin-interfaced sensors with silicon integrated circuits consolidated on a flexible platform for complex signal processing. The wireless transmission requires several tens of mW and represents the dominating consideration in overall power consumption. Bluetooth Low Energy (BLE) technologies can further



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**Figure 6. Wireless Systems for Body-Integrated Chemical Sensors.** (A) Photograph of a wristband integrated with a multiplexed sweat sensor array and a flexible printed circuit board with Bluetooth capabilities [1]. (B) Photograph of an alcohol iontophoretic-sensing tattoo with integrated flexible electronics on human skin [43]. (C) A device with the form factor of a band-aid for continuous detection of ions in sweat with a radio frequency identification (RFID) antenna for wireless signal transmission [80]. (D) Schematic illustration of a contact lens integrated with a glucose sensor and an intraocular pressure sensor [47].

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improve performance in this sense by alternating between data transmission and low-power states for semi-continuous readings. Figure 6B shows an alcohol monitor that incorporates a tattoo-based iontophoretic biosensing system [43]. A platform of flexible electronics controls the operation and wireless data transmission. As an alternative to battery powered BLE systems, near-field communication (NFC) protocols can enable not only wireless signal transmission but also wireless power harvesting from the receiving device for measurement and data collection. Figure 6C shows a device in the form factor of a band-aid for continuous detection of ions in sweat with a radio frequency identification antenna that transmits readings to a smartphone [80]. Wireless power and radio frequency (RF) communication occurs with a receiver/transmitter powered at ~0.5 W. The reduction of the reflection value ( $S_{11}$ ) by -2.9 dB at 13.56 MHz confirms successful tuning of the antenna. Simultaneous collection of different biosignals through multiplexing is also possible. Figure 6D shows a glucose sensor integrated into a resistance (R), inductance (L), and capacitance (C) oscillating circuit that operates in the RF. In this design, a reader antenna inductively couples and powers the remote sensor, and a magnetic field connects the circuits. As a result, the wireless sensing antenna can analyze the correlation between the reflection condition and the change in resistivity of a piece of graphene due to exposure to glucose. The simple pyrene-chemistry immobilizes GOx onto graphene via  $\pi$ - $\pi$  stacking for selective detection of glucose, which can also be tuned for the sensing of a wide range of biomarkers. Because of the enzyme immobilization, the sensor responds specifically to glucose in the presence of interferents such as ascorbic acid, lactate, and uric acid in the tear [47]. A recent advanced system is a stretchable, transparent, and wireless contact lens-based device for glucose sensing along with display pixels to visualize sensor readouts [81]. The device consists of the GOx functionalized p-type graphene-based glucose sensor and micro-LED with stretchable interconnects and antennas formed from silver nanofibers. The device activates the LED when the glucose concentration exceeds the limit, eliminating the need for additional, bulky measurement equipment. Figure 6E shows a sweat sensor with a magnetic loop antenna and associated NFC for wireless colorimetric detection to reveal the concentration of multiple chemical components, such as metabolites, pH, and sweat rate [9]. The wireless interface with integrated electronics allows communication to external computing and digital analysis systems such as the smartphone. This technology capitalizes on NFC schemes to launch image capture and analysis software. As a complementary option, Figure 6F shows a microfluidic system that uses integrated electrodes as an electrical interface to a thin, battery-free, wireless electronic module for real-time measurement of sweat with capabilities in NFC [12]. This approach enables direct digital readout via an interface to smartphone or other NFC-enabled devices. One disadvantage for NFC is the relatively short-range operation, typically no more than 1 meter. Both BLE and NFC technologies support data transmission at rates of several hundred to several thousand samples per second, but not significantly more.

### Concluding Remarks

This review summarizes a broad set of emerging sensing modalities and advanced materials/device designs in body-integrated flexible platforms capable of analyzing biofluids that can be captured in a noninvasive or minimally invasive fashion. The focus is on sweat, tears, saliva, and ISF, with wireless technologies that have the potential to yield a comprehensive set of insights into physiological health and chronic diseases, beyond possibilities afforded by biophysical sensing.

### Outstanding Questions

Will advances in chemistry further improve the stability, selectivity, and sensitivity of body-interfaced chemical sensors?

Will new sensing modalities and materials allow for measurements of biomarkers with extremely low concentrations or those, such as proteins, DNA, and RNA, that currently require multistep preparation protocols with long waiting times?

What classes of energy harvesting strategies and low power system designs will allow for long-term, continuous operation of body-interfaced devices?

(E) Photograph and schematic illustration of an epidermal microfluidic sweat monitoring device integrated with near-field communication (NFC) capabilities [9]. (F) Schematic illustrations of device and electrode layouts of a battery-free, wireless microfluidics system with NFC electronics for digital measurements of sweat loss, sweat rate, and electrolyte concentration [12]. Abbreviations: CE, counter electrode; IE, iontophoretic electrode; RE, reference electrode; WE, working electrode.



Despite substantial recent advances, challenges remain in the stability, selectivity, sensitivity, and susceptibility to biofouling of the active components and in the mechanical properties of the overall platforms (see Outstanding Questions) [20,82]. External parameters (temperature, humidity, and sunlight) can influence the response of the sensors in ways that can be difficult to predict. Internal parameters (biofluid pH, ionic strength, and interfering chemicals) and disease/physiological states have additional effects. Long-term reliable performance without repeated calibration or continuous comparisons against reference standards can be difficult in many cases. In addition, body motions can lead to time variances in the interfaces between different components and the targeted tissues, thereby creating the potential for mechanical failure and/or measurement artifacts. In these and broader contexts, specialized interfacial materials and packaging strategies for the active and passive components of the systems are essential. The chemical sensor component must be in direct contact with the biofluids while the supporting electronics must be completely isolated from them. A frontier of research in body-interfaced sensors includes development of measurement modalities and materials that can target biomarkers with extremely low concentrations or those with challenging chemistries such as proteins, DNA, and RNA that require multistep preparation protocols and incubation times. Wireless capabilities and power supply are additional areas for future work in the engineering science of these systems.

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