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Mini Review

Recent Development of Wearable Electrochemical Sensors for Sweat Analysis

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Sweating during training and competition leads to a large amount of water and electrolyte loss, along with the excretion of lactic acid, urea, creatine, creatinine and other metabolites into sweat. Through sweat analysis, we can quickly understand the physical condition, metabolism and adaptability of a human body. This is of great practical significance in athlete selection, daily sports training and physical fitness recovery. In recent years, with the rapid development of flexible printing of electronic technology and the Internet of Things, all kinds of miniaturized wearable devices that can obtain real-time data information have been increasingly developed. This short review summarizes the recent development of wearable devices in sweat analysis technology.

Keywords: Wearable device; Electrochemistry; Sweat analysis; Athlete; Sports training; Biosensor

1. INTRODUCTION

Sweating is a normal physiological activity of the human body that can be divided into active and passive sweating [1–3]. The liquid secreted by sweat glands is called sweat. Passive sweating is caused by hot or humid weather or an irritable mood. Body heat can be removed by water evaporation to maintain the body temperature within a normal range [4–6]. Active sweating is sweating caused by active exercise. It is not only conducive to heat dissipation but also removes metabolites produced by exercise [7–9]. Approximately 98 ~ 99% of human sweat is water [10]. The pH of sweat ranges from 4.2 to 7.5 [11]. The main chemical components of sweat are urea, lactic acid, glucose, uric acid, creatine, creatinine, amino acids and electrolytes (sodium ions, potassium ions, calcium ions, magnesium ions, chloride ions and inorganic phosphorus) [12,13]. During exercise, sweating can lead to a large amount of water and electrolyte loss and the excretion of human metabolic products [14,15]. Therefore, the

detection of the above-mentioned substances in sweat can help in the understanding of the physical condition, metabolism and adaptability of the studied body during exercise [16,17]. This information has very important practical significance in athlete selection, daily sports training and physical fitness recovery [18–22].Athletes' sweat is usually analysed by biosensors [23–27], ion chromatography [28,29], mass spectrometry [30–32], capillary electrophoresis [33,34], liquid chromatography and gas chromatography [35–39]. For biosensors, each sensor can only detect one indicator. The analysis time for mass spectrometry, capillary electrophoresis, liquid chromatography and gas chromatography is approximately tens of minutes. Because of their large volume, these instruments have difficulty meeting the needs of rapid analysis in the field of sports. Therefore, increasing attention has been paid to the development of portable analysis instruments and detection technology for athletes' sweat. In recent years, with the popularity and development of wearable devices and smartphones, the use of these devices in the field of athlete sweat analysis has been widely studied at home and abroad.

2. PRINTING TECHNOLOGY AND FLEXIBLE SUBSTRATES

At present, with the application of wearable devices in life and health, sensors, especially flexible sensors that can monitor physiological signals/biomarkers in real-time [40–46], have become an important research direction. Therefore, flexible printing technology has become an important basis for the fabrication of flexible sensors [47–50]. Commonly used flexible printing technology mainly includes lithography and printing technology [51,52]. Due to the need for ultra-clean laboratories and complex production processes, flat printing technology is often expensive, which is not suitable for the long-term development of flexible electronic products. In contrast, printing technology has the advantages of a low cost, good repeatability and easy operation, and this section will briefly introduce various printing methods [53–55]. Template free printing mainly includes inkjet and 3D printing. Due to the characteristics of template free printing, these two printing technologies highly rely on printing ink, which requires the viscosity of the ink to match the printing substrate so that it can be evenly distributed on the receiving substrate [56–59]. Therefore, template-free printing methods often involve a series of advanced technologies, such as piezoelectric, pneumatic and electrohydrodynamic drives. Template printing technologies include screen printing, embossing, aniline printing and gravure printing. Among them, screen printing is the most widely used printing method [60–62]. The wearable electrochemical sensor mainly includes a substrate and functional layer (including a conductive functional layer, electrochemical active layer, insulating packaging layer, biomarker enrichment layer, microfluidic channel and ion electroosmosis electrode). Screen printing technology is generally applied to the assembly of functional layers on the substrate, for instance, the preparation of a conductive functional layer and an electrochemically active layer.

Traditional electronic devices are bulky and cannot meet the requirements of wearable applications. Therefore, flexible and printed electronic products have great market competitiveness because of their flexibility, comfort and light weight. Although flexible printed electronic products have a variety of sensing mechanisms for active materials, along with various signal conversion methods, flexible substrates can indirectly determine the flexibility of electronic products.

2.1 Polyethylene

Polyethylene terephthalate (PET) has become one of the most commonly used flexible platforms for making wearable electrochemical sensors due to its advantages of good light transmission, low cost, moderate thermal expansion coefficient and chemical inertia [63–67]. In 2013, Kolliopoulos et al. [68] prepared a screen-printed graphite sensor for the determination of Sb(III) based on PET. Wang et al. [69–71] developed a sweat electrolyte and metabolite sensor with PET-based glasses. In 2016, the detection principle of glucose and lactic acid sensors was found to be based on the electrocatalytic oxidation reaction of glucose oxidase and lactate oxidase to a corresponding substrate for producing H_2O_2 with electrochemical activity (Figure 1) [72]. The sensing was realized by detecting the reduction current of H_2O_2, which was proportional to the substrate concentration. Additionally, a sodium-ion and potassium-ion sensor is based on the potential difference generated by the charge separation of the corresponding ion-selective electrode at the solution interface to measure the concentration.

Figure 1. Examples of minimally invasive CGM systems based on electrochemical sensing techniques (reprinted with permission from MDPI [72]).

2.2 Polydimethylsiloxane

Polydimethylsiloxane (PDMS) is a silicon-based polymer. Due to its adjustable Young's modulus, high extensibility, chemical resistance, easy processing, non-toxicity and high transparency (up to 95%), PDMS is widely used as the substrate of flexible printed electronic products. Cao et al. [73,74] successfully prepared patterned metal nanostructures using PDMS as a template for a sputtered Au film and a self-assembled monolayer of silicon as a substrate. This technology can be used to prepare nanostructures that are difficult to prepare by other technologies. Matsuhisa et al. [75] reported a printable elastic conductor with a high initial conductivity (738 S/cm) and high conductivity (182 S/cm) after a 215% tensile test. The elastic conductive ink is composed of a silver sheet, fluorine surfactant and organic solvent. To realize the early diagnosis of cystic fibrosis, Choi et al. [76] designed a wearable chloride sweat sensor based on PDMS. The detection mechanism established a balance between the reference solution and the test solution by using a salt bridge so that the battery potential was directly related to the ion concentration. This design can realize the long-term and accurate determination of chloride-ion content and has been successfully applied in human detection devices. Su et al. [77] designed a strain sensor based on PDMS by means of column pattern template-induced printing, nanoparticle self-assembly and vacuum thermal evaporation. The PDMS-based sensor could detect

facial expression changes by monitoring resistance changes in four different directions. Li et al. [78] prepared flexible wires with good mechanical stability and high conductivity by screen printing Ag ink on a pre-treated PDMS surface. Additionally, PDMS can be used for the preparation of microfluidic devices (Figure 2). Wang et al. [79] designed an epidermis detection system that could realize the real-time collection and monitoring of glucose or lactic acid in sweat. Table 1 shows the characteristics of different materials for the microfluidic chip.

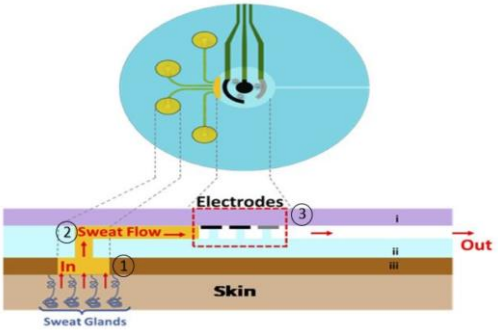

Figure 2. Schematic of wearable microfluidics: (1) sampling, (2) transfer to the site of detection, and (3) detection by electrochemical sensors (reprinted with permission from MDPI [78]).

Table 1. Characteristics of different materials for microfluidic chips.

Material	Silicon	Glass	Quartz	Polymethyl methacrylate	Polyphenyl ether	Polyethylene	Polydimethylsiloxane
Dielectric constant	11.7	3.7~16.5	-	3.5~4.5	2.2-~2.6	2.25	3.0~3.5
Field intensity	-	2500	-	>400	-	-	1000
Thermal conductivity	1.57	0.7~1.1	1.4	0.2	0.2	0.4	0.2
Dissipation of energy	-	2.8	-	-	-	-	1.0
Softening temperature	-	500~821	500~821	>1000	105	85~125	-
Transmittance	-	89~92	>76	>92	40~70	50~70	>70
CTE	0.26	0.05~1.5	0.04	7~9	6~10	12~18	3.5

2.3 Polyimide

As a thermally stable polymer, polyimide (PI) has a high glass transition temperature and good flexibility. It is a flexible electronic substrate material that can be used at a high working temperature [80]. In 2008, researchers established a miniaturized norepinephrine and glucose bioelectrochemical sensor using PI film, which provided an example for developing a micro-biosensor eye for monitoring biomarkers in tears [81]. Kuribara et al. [82] designed a heat-resistant and flexible organic thin film transistor using a PI substrate. The PI-based organic thin film transistors also exhibit flexibility and high thermal stability. Selvam et al. [83] constructed wearable biochemical Au and ZnO sensors on a PI substrate, which could monitor alcohol consumption by detecting ethyl glucuronide (ETG) (Figure 3).

Based on a chemical resistance sensing mechanism, the Au or ZnO sensor could test the impedance signal directly related to the ETG concentration when an AC voltage was applied. It has been proven that both Au and ZnO sensors have good stability, a wide detection range and good specificity. In 2017, Nakata et al. [84] reported a wearable, flexible sweat chemical sensor based on PI. The sensor could successfully measure the sweat pH and the skin temperature in real time through skin contact. This flexible integrated device was expected to be used for monitoring sweat in regard to health care and sports. However, due to the colour limitations and poor stretchability, PI is difficult to use as a flexible platform for highly stretchable and transparent electronic products.

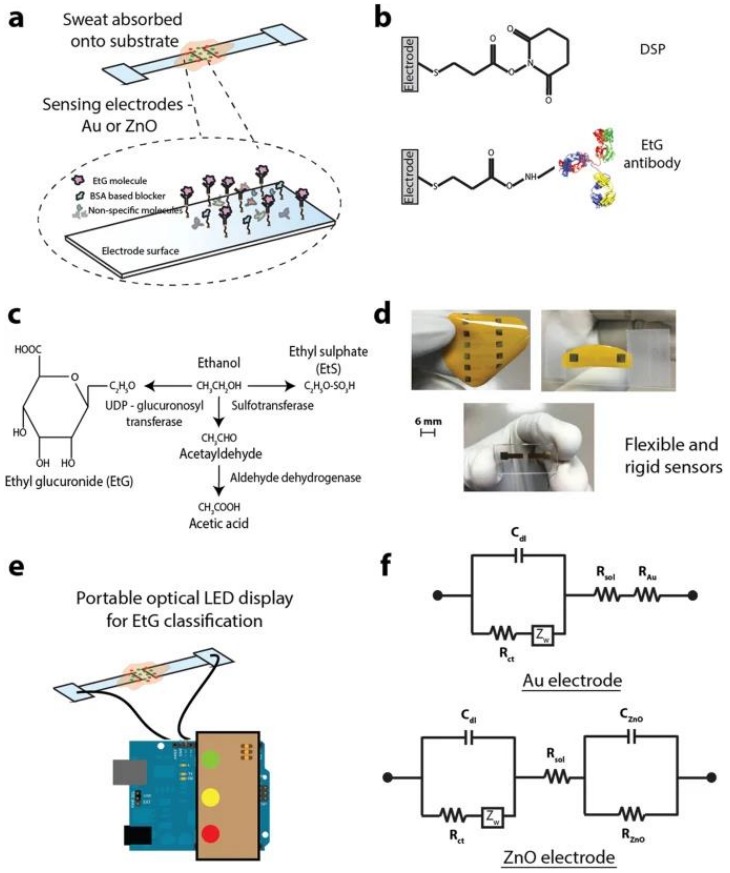

Figure 3. Schematic of integrated glass and flexible substrate-based sensing platforms for point and continuous detection (reprinted with permission from Scientific Report [83]).

2.4 Polyurethane

Polyurethane (PU) macromolecules contain many strong polar groups. They have high mechanical strength and oxidation stability, excellent flexibility and resilience, and good oil, solvent and water resistance. Based on the above properties [85], PU has become the best choice for a wearable electronic device platform. In 2013, Li et al. [86] developed a new type of PU-based conductive adhesive with polyethylene glycol (PEG) as a curing agent and a flake silver sheet as filler, which could be further combined with printing technology to design equipment with high conductivity and extensibility. Due

to its porous nature, commercial PU sponges can also be used as a platform for pressure-sensitive sensors. For example, Yao et al. [87] reported a graphene-coated PU sponge that could be used as a pressure-sensitive material; this sensor was prepared by dipping in graphene oxide to modify the PU sponge. Furthermore, Wang et al. [88] designed a fully printed electrochemical ion-selective NH_4^+ sensor, glucose sensor and glucose biofuel cell.

2.5 Tattoo paper

In 2012, scientists first reported printed tattoo electrochemical sensors [89]. Since then, this technology has brought new vitality to the development of wearable human electronic products. As a flexible substrate, tattoo paper is gradually used in various electrochemical sensors, biosensors, and energy collection and storage devices. Electrode arrays for monitoring NH_4^+, skin pH and Na^+ in sweat can be obtained by screen printing technology and transfer printing technology [90–92]. The sensing mechanism of these three sensors is to measure the potential generated between the corresponding ion-selective electrode and reference electrode. Tattoo sensors with bismuth as the working electrode can realize the non-invasive real-time detection of Zn^{2+} in human sweat by using stripping voltammetry [93]. In addition, tattoo biosensors for the dynamic monitoring of lactic acid and the non-invasive monitoring of glucose and alcohol have also been developed, which provides inspiration for other biosensor systems [94–96]. Notably, the glucose and alcohol tattoo biosensors combine iontophoresis with the biosensor process to accelerate the collection of sweat to significantly improve sensing performance. In the field of microneedle drug delivery, tattoo-based devices have also made progress. Wang et al. [97] prepared a tattoo-based epidermal patch with thousands of microbubbles, which could complete the delivery of a micro-dose of drugs. Due to their excellent mechanical properties, tattoo stickers have also been used in energy collection and storage devices, such as epidermal biofuel cells and alkaline batteries, which can meet the requirements of high flexibility and tunable discharge capacity.

3. CONCLUSION

Through a non-invasive analysis of sweat, we can determine the physical condition, metabolism and adaptability of a body during sports. This information has very important practical significance in athlete selection, daily sports training and physical fitness recovery. Various conventional laboratory techniques based on spectrophotometry, atomic emission spectrometry, flame emission spectrometry, mass spectrometry, liquid chromatography, capillary electrophoresis, gas chromatography, potentiometry and other conventional laboratory technologies have become increasingly developed, but the required equipment is bulky; thus, these techniques and technologies are not suitable for the desired detection tests of athletes at sports scenes. In recent years, wearable and portable analysis techniques for real-time on-site analysis of athletes' sweat have received extensive attention. Wearable biochemical index detection systems have broad application prospects in competitive and national fitness sports. This review summarizes the application of printing technology and flexible substrates in the field of wearable

printed devices, such as electrochemical sensors and biosensors. The rapid development of information technology, printing technology and new material science represented by the Internet of Things has brought new opportunities for flexibility and printed electronics. However, there are still many key challenges to be solved before the practical application of wearable sensors. First, wearable sensors need to enhance their detection limit of biomarkers in body fluids to achieve the goal of precision medicine. Second, most of the existing wearable biosensors are enzyme-based sensors, which have some problems, such as enzyme activity being easily disturbed by environmental pH, temperature, ionic strength, etc. In addition, enzyme immobilization will reduce the efficiency of electron transfer. Therefore, the development of highly selective non-enzymatic biosensors is still a major hurdle. Third, reports about wearable devices with a multifunctional integrated sensing system are still relatively few. The development of intelligent wearable sensing systems, such as the combination of physical and chemical sensing and the detection of multiple analytes in the same body fluid, is a major development direction. Fourth, we should use various printing technologies, lithography methods, and chemical deposition and vacuum filtration techniques to obtain printed high-performance electronic products that are flexible. At the same time, the realization of wearable technology modules that are flexible, such as test circuits, and those used for data collection, data processing and data reading, should be considered.

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