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Research Article

Electrochemical Biosensors: Mechanism and Applications

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ABSTRACT

Presently, the extensive variety of biomaterials and their broad range of applications have broadened the scope of these materials. Biomaterials are integrated into biosensing platforms to create devices characterized by high sensitivity and specificity, rapid detection capabilities, portability, cost-effectiveness, and user-friendly operation. The limelight of this review focus on potential role and applications of bio based electrochemical sensors. The eMIP/PB/SPCE-based lactate sensors demonstrate significant promise as point-of-care (POC) devices for detecting sweat lactate. This technology could be adapted for reagent-free detection of a wide range of molecules. We discussed a dual-trigger and reusable electrochemical biosensor for microRNA. It achieved sequential signal amplification by integrating CHA and tripedal walker mechanisms. The tripedal DNA walker was activated through APE1 enzymatic cleavage reactions. Strand displacement was engineered for electrode regeneration and optimizing signal strength. It effectively discriminates the target from other interfering RNAs and enables detection in serum. This study introduces an innovative non-invasive electrochemical biosensor designed for detecting glucose concentration in human saliva. The biosensor utilizes a nickel foam substrate decorated with needle-like CoO nanowires, providing an optimal surface area for nanowire growth. These nanowires form a self-assembled flower-like nanostructure with a highly porous configuration, featuring an impressive BET surface area of 154.3 m2 g−1 and demonstrating excellent catalytic capabilities.

KEYWORDS: lactate sensors, eMIP/PB/SPCE, CHA and tripedal walker, CoO nanowires

Introduction

Biosensors are analytical devices that combine bio cognitive components and signal transduction devices. Taking advantage of the precision and resolution inherent in biological sensors, they have an enormous ability to study a wide range of target molecules, including small entities, proteins, nucleic acids and cells [1].Among biological sensors, electrochemical biosensors, using electrochemical agents for signalling have clear advantages such as easy integration, higher sensitivity, fast detection kinetics and cost-effectiveness.
Electrochemical biosensors are analytical devices that combine a biological recognition element (such as enzymes, antibodies, or DNA) with an electrochemical transducer to detect and quantify specific analytes in a sample [2-4] They are widely used in various fields including medical diagnostics, environmental monitoring, food safety, and bioprocess control.

The key components of an electrochemical biosensor typically include:

1. **Biological Recognition Element**: This could be enzymes, antibodies, nucleic acids, or whole cells that selectively interact with the target analyte, leading to a measurable signal.

2. **Transducer**: Converts the biochemical reaction into a quantifiable electrical signal. Common types include electrodes (working, reference, and counter electrodes), which can measure changes in current, potential, impedance or conductance.

3. **Electronics and Signal Processing**: These components amplify and process the electrical signal generated by the transducer to provide quantitative information about the analyte concentration.
Currently, a variety of biologically active agents, including enzymes, antibodies, antigens, microorganisms, cells, tissues, and nucleic acids, can directly function as both bio recognition and transduction components. Nano enzymes, which exploit their strong physicochemical properties and greatly improve electro catalytic performance, have become an important driving force in the development of electrochemical sensors [5-6]. In essence, the amalgamation of nanozymes and electrochemical sensing has permeated diverse domains, spanning food science, healthcare, environmental monitoring, disease diagnostics, and immune analysis**.**

Reagentless Lactate Sensing with Prussian Blue and Electro polymerized Molecularly Imprinted Polymers in Electrochemical Bio sensing: The rapid progress in bio sensing, microfluidics, flexible electronics, and wireless communications technologies has spurred the creation of wearable biosensors for sweat analysis, with a focus on lactate monitoring. An increase in lactate production can result in cellular acidosis and impair muscle function [7-9]. Elevated levels may also indicate ischemic pressure and serve as a biomarker for tissue perfusion progression.

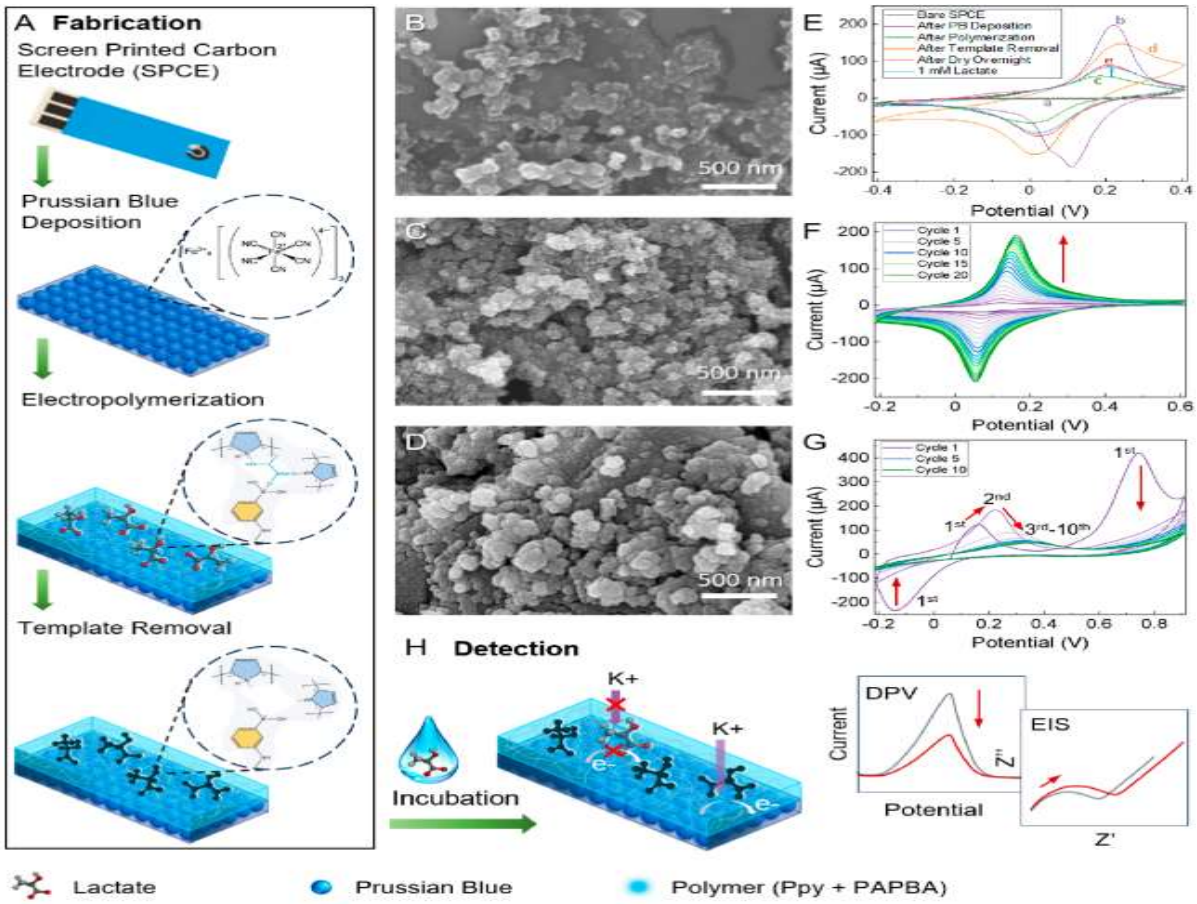

Fig 1: (A) Diagram depicting the fabrication process of the eMIP/PB/SPCE-based sensor; Field Emission Scanning Electron Microscopy (FE-SEM) images showing (B) the bare SPCE, (C) PB/SPCE, and (D) eMIP/PB/SPCE configurations; (E) Characteristic cyclic voltammetry (CV) curves measured in 0.1 M KCl for (a) bare SPCE, (b) PB/SPCE after PB deposition, (c) after eMIP polymerization, (d) post-lactate template removal, (e) eMIP/PB/SPCE after overnight drying, and (f) following 1 mM lactate incubation; (F) Cyclic voltammograms detailing PB electrodeposition, (G) electro polymerization of 50 mM 3-APBA, 100 mM pyrrole, and 10 mM lactate on PB/SPCE, and (H) an illustration depicting the lactate detection mechanism, showing the eMIP partially obstructing electron transfer, with corresponding Differential Pulse Voltammetry (DPV) and Electrochemical Impedance Spectroscopy (EIS) responses

Therefore, detecting lactate in sweat provides a non invasive and convenient method for assessing exercise intensity [10], optimizing athletic training, enhancing performance, and supporting medical diagnosis and monitoring. Electro polymerization provides a rapid and straightforward method for constructing MIP-based electrochemical sensors, enabling direct deposition of the MIP layer onto the electrode surface. However, many MIP-based electrochemical sensors rely on external redox couples (e.g., ferricyanide/ferrocyanide) to detect binding events between electrochemically inactive analytes and the MIP. This dependence may interfere with the interaction between MIP and analyte molecules and complicate on-body applications [11-14]. Therefore,

there is an urgent need to develop reagent-free MIP-based biosensors for straightforward and convenient lactate detection, advancing wearable MIP-based biosensor technology.
Prussian Blue (PB) stands as a prominent electron-transfer mediator in enzymatic biosensors. The sensor incorporates an electrodeposited layer of Prussian Blue (PB) as the internal redox probe, along with an electro-polymerized Molecularly Imprinted Polymer (eMIP). The eMIP utilizes 3-APBA and pyrrole as functional monomers for the specific recognition of lactate. The growth of eMIP [15-18], removal of the template, and lactate rebinding processes were monitored and confirmed in real-time using Electrochemical-Surface Plasmon Resonance (EC-SPR). The sensor's efficacy was assessed in spiked 0.1 M KCl solution and artificial sweat (AS) using Differential Pulse Voltammetry (DPV) and Electrochemical Impedance Spectroscopy (EIS).

Creation of a reusable tripedal DNA walker electrochemical biosensor for highly sensitive detection of microRNA-155: Given that microRNAs (miRNAs) are pivotal biomarkers for disease diagnosis and prognosis, there is a growing emphasis on enhancing detection sensitivity through signal amplification strategies in research on detecting low-abundance miRNAs. MicroRNAs (miRNAs) are short endogenous RNA molecules, typically 20–24 nucleotides long, that play critical regulatory roles within cells [20–24]. Extensive research has demonstrated that miRNAs can either inhibit or promote the expression of genes associated with various diseases [25], establishing them as valuable biomarkers in disease classification, diagnosis, and prognosis [26,27]. For instance, miRNA-21 shows increased expression in breast cancer patients, which significantly decreases post-surgery [28]. Meanwhile, miRNA-155 is notably upregulated in several cancers, including breast cancer [29], leukemia [30], thyroid cancer [31], gastric cancer [32], colorectal cancer [33], bladder cancer [34], and renal cell carcinoma [35]. This intimate connection between miRNAs and cancer progression underscores their importance in cancer screening and therapeutic strategies. Various techniques, such as electrochemistry [36,37], fluorescence [38,39], colorimetry [40,41], and electro chemiluminescence (ECL) [42], have been developed for miRNA detection. However, these methods may lack sufficient sensitivity to detect ultra-low levels of miRNA without signal amplification. To bolster detection capabilities, various signal amplification strategies have been integrated into electrochemical analysis. These include catalytic hairpin reaction (CHA) [44–46], hybridization chain reaction (HCR) [47–49], entropy-driven catalytic reaction (EDC) [50–52], as well as enzyme-free and nuclease-mediated signal amplification methods [53,54]. Notably, DNA walkers have emerged as dynamic nano devices responsive to targets, capable of converting chemical energy into mechanical kinetic energy. This enables incremental movement along a track and amplification of signals throughout the walking process.

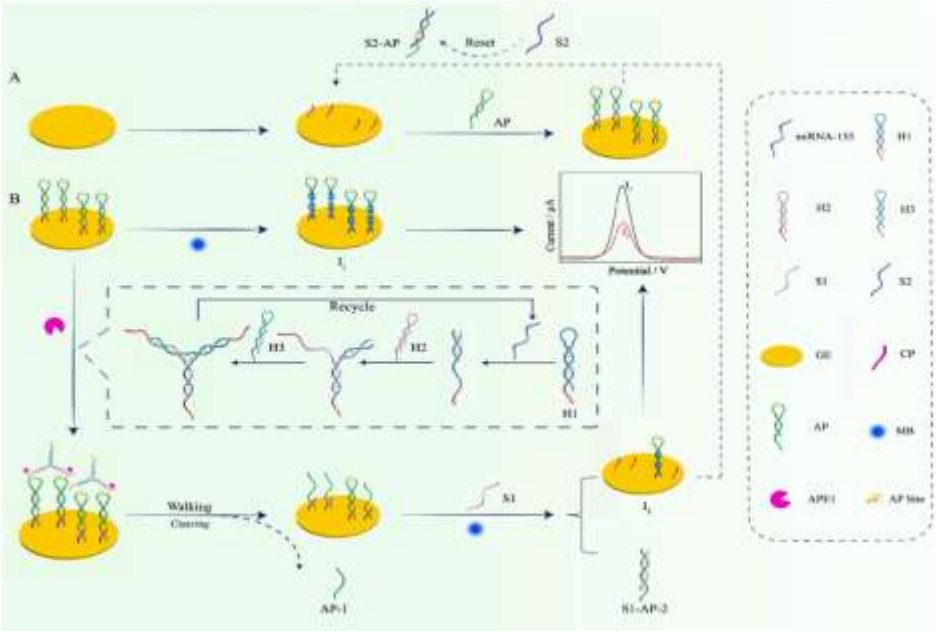

Fig 2: Diagram illustrating a regenerative electrochemical biosensor featuring a robust cascade amplification method. This includes CHA activation for generating a tripedal DNA walker (TDW) triggered by the target, along with APE1 enzyme digestion-driven amplification. A: Electrode regeneration via S2 strand displacement reaction; B: Bio sensing process for miRNA 155, utilizing S1 strand displacement to maximize current signal changes induced by the target.

This technology for electrode regeneration, based on a strand displacement strategy, allows for the repetitive use of DNA-modified gold electrodes. This presents an innovative method toward achieving an efficient and economical detection system [55]. The novel electrode regeneration technique exhibits excellent scalability, making it suitable for detecting various disease-related RNA targets. This development shows potential for

implementing signal amplification through DNA nanostructures and introduces further analytical strategies for clinical diagnostics.

Electrochemical biosensor for glucose detection in human saliva: Diabetes, a metabolic disorder, poses significant health risks [61,62]. Hence, monitoring glucose levels is crucial for diabetes management. However, many current methods for detecting glucose, such as blood collection from fingertips or veins, are invasive, increasing the risk of infection and causing psychological distress among patients [63,64]. Therefore, there is a strong need to develop safe and painless non-invasive methods for diagnosing glucose levels in human biofluids. Several electrochemical biosensors utilizing glucose oxidase (GOx) have been created for saliva glucose detection [67]. However, research indicates that GOx enzymes possess inherent drawbacks such as instability and susceptibility to environmental factors like pH, moisture, and temperature [68]. Consequently, researchers have explored enzyme-free glucose sensors employing noble-metal and transition-metal catalysts to overcome these limitations. In contrast, Ni foam (NF) is recognized as an ideal substrate for manufacturing glucose biosensors due to its three-dimensional interconnected microstructure. It is cost-effective owing to its abundant availability, large surface area, and excellent conductivity [66–68]. Therefore, growing transition metal oxides directly on NF presents a promising approach to developing glucose sensors that are both cost-effective and high-performing. Cobalt oxide (CoO), a readily available transition metal oxide material, has garnered significant interest in non-enzymatic glucose sensors due to its easy preparation and theoretically significant electrocatalytic activity [69,70].

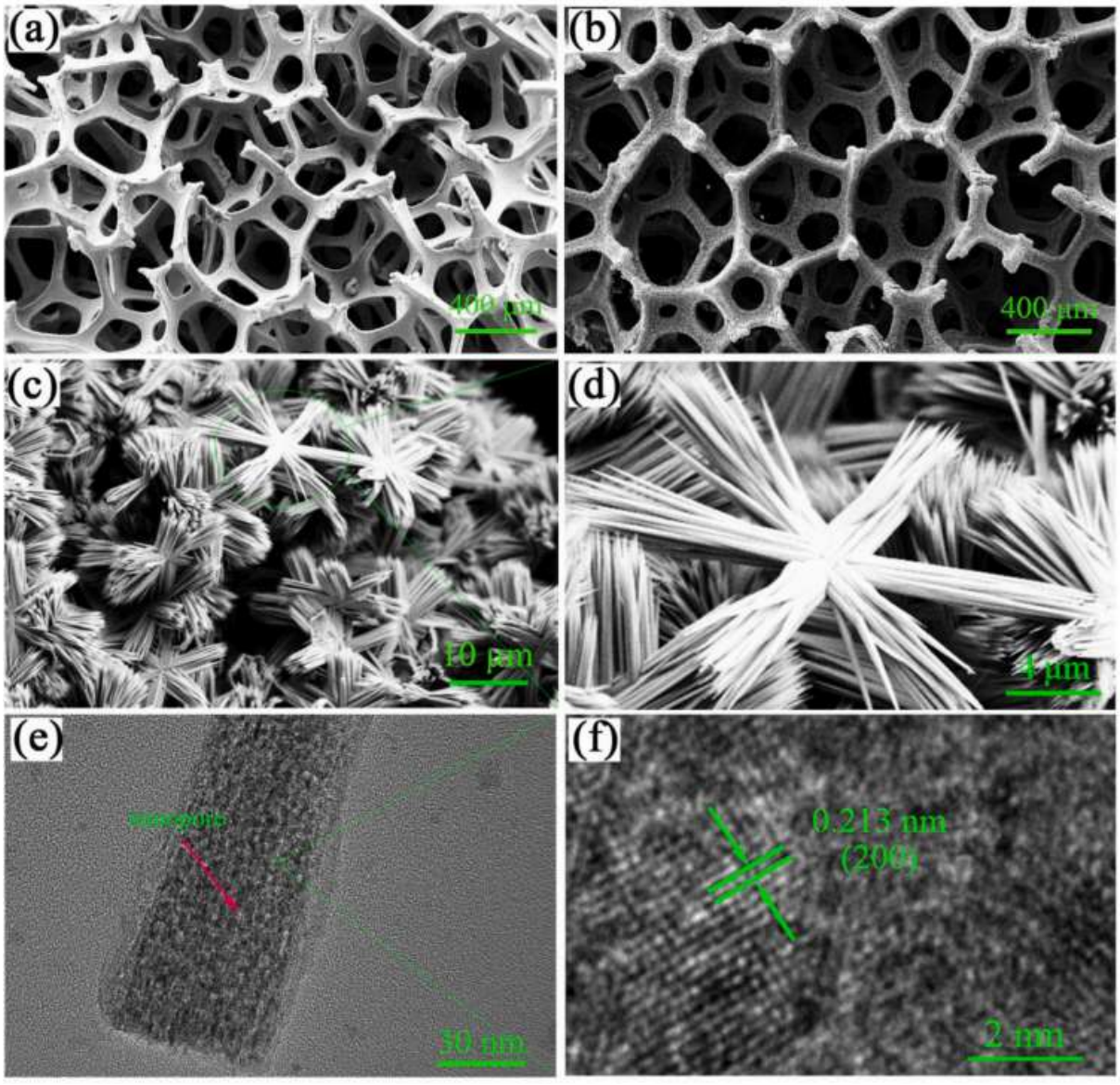

Fig 3: SEM images of (a) NF and (b, c, and d) CoO NWs/NF with different magnifications.

Despite these advantages, bulk CoO suffers from limited active sites and poor conductivity, which hinders achieving optimal catalytic performance. As a result, efforts have focused on nano structuring bulk CoO to enhance electrocatalytic activity by increasing active surface areas and accelerating reaction kinetics [81,82].

Among nanostructured CoO materials, cobalt oxide nanowires (CoO NWs) offer short radial pathways and efficient electron transport in the axial direction, effectively reducing resistance and enhancing reaction kinetics. In this investigation, we successfully achieved the uniform and extensive growth of mesoporous nanoneedle-assembled CoO microflowers on a nickel foam (NF) substrate. Utilizing this distinctive nanostructure, we developed a non-invasive electrochemical biosensor for detecting glucose levels in human saliva. We systematically evaluated the sensing capabilities of the biosensor.

Conclusion:

The diverse array of biomaterials and their wide-ranging applications have expanded the possibilities for these materials. Biomaterials are now integrated into biosensing platforms to develop devices known for their high sensitivity, specificity, rapid detection, portability, cost-effectiveness, and user-friendly operation. The tripedal DNA walker is activated via APE1 enzymatic cleavage reactions. Strand displacement is engineered for electrode regeneration, optimizing signal strength and effectively distinguishing the target from other interfering RNAs, allowing for detection in serum. Furthermore, we introduce an innovative non-invasive electrochemical biosensor designed for detecting glucose concentration in human saliva. This biosensor utilizes a nickel foam substrate decorated with needle-like CoO nanowires, providing an optimal surface area for nanowire growth. This technology can be adapted for reagent-free detection across various molecules.

References:

1. Baker, L. B.; Wolfe, A. S. Physiological Mechanisms Determining Eccrine Sweat Composition. Eur. J. Appl. Physiol 2020, 120 (4), 719– 752.
2. Chien, M.-N.; Fan, S.-H.; Huang, C.-H.; Wu, C.-C.; Huang, J.-T. Continuous Lactate Monitoring System Based on Percutaneous Microneedle Array. Sensors 2022, 22 (4), 1468.
3. Jia, W.; Bandodkar, A. J.; Valdés-Ramírez, G.; Windmiller, J. R.; Yang, Z.; Ramírez, J.; Chan, G.; Wang, J. Electrochemical Tattoo Biosensors for Real-Time Noninvasive Lactate Monitoring in Human Perspiration. Anal. Chem. 2013, 85 (14), 6553–6560.
4. Green, J. M.; Pritchett, R. C.; Crews, T. R.; McLester, J. R.; Tucker, D. C. Sweat Lactate Response between Males with High and Low Aerobic Fitness. Eur. J. Appl. Physiol 2004, 91 (1), 1–6.
5. Derbyshire, P. J.; Barr, H.; Davis, F.; Higson, S. P. J. Lactate in Human Sweat: A Critical Review of Research to the Present Day. Journal of Physiological Sciences 2012, 62 (6), 429–440. ACS Applied Materials & Interfaces www.acsami.org Forum Article https://doi.org/10.1021/acsami.3c19448 ACS Appl. Mater. Interfaces XXXX, XXX, XXX−XXX I
6. Garland, N. T.; Schmieder, J.; Johnson, Z. T.; Hjort, R. G.; Chen, B.; Andersen, C.; Sanborn, D.; Kjeldgaard, G.; Pola, C. C.; Li, J.; Gomes, C.; Smith, E. A.; Angus, H.; Meyer, J.; Claussen, J. C. Wearable Flexible Perspiration Biosensors Using Laser-Induced Graphene and Polymeric Tape Microfluidics. ACS Appl. Mater. Interfaces 2023, 15 (32), 38201–38213.
7. [7] Bandodkar, A. J.; Gutruf, P.; Choi, J.; Lee, K.; Sekine, Y.; Reeder, J. T.; Jeang, W. J.; Aranyosi, A. J.; Lee, S. P.; Model, J. B.; Ghaffari, R.; Su, C.-J.; Leshock, J. P.; Ray, T.; Verrillo, A.; Thomas, K.; Krishnamurthi, V.; Han, S.; Kim, J.; Krishnan, S.; Hang, T.; Rogers, J. A. Battery-Free, Skin-Interfaced Microfluidic/Electronic Systems for Simultaneous Electrochemical, Colorimetric, and Volumetric Analysis of Sweat. Sci. Adv. 2019, 5 (1), No. eaav3294.
8. Gao, W.; Emaminejad, S.; Nyein, H. Y. Y.; Challa, S.; Chen, K.; Peck, A.; Fahad, H. M.; Ota, H.; Shiraki, H.; Kiriya, D.; Lien, D.-H.; Brooks, G. A.; Davis, R. W.; Javey, A. Fully Integrated Wearable Sensor Arrays for Multiplexed in Situ Perspiration Analysis. Nature 2016, 529 (7587), 509–514.
9. Li, M.; Wang, L.; Liu, R.; Li, J.; Zhang, Q.; Shi, G.; Li, Y.; Hou, C.; Wang, H. A Highly Integrated Sensing Paper for Wearable Electrochemical Sweat Analysis. Biosens Bioelectron 2021, 174, No. 112828.
10. [10] Hojaiji, H.; Zhao, Y.; Gong, M. C.; Mallajosyula, M.; Tan, J.; Lin, H.; Hojaiji, A. M.; Lin, S.; Milla, C.; Madni, A. M.; Emaminejad, S. An Autonomous Wearable System for Diurnal Sweat Biomarker Data Acquisition. Lab Chip 2020, 20 (24), 4582–4591.
11. Xuan, X.; Chen, C.; Molinero-Fernandez, A.; Ekelund, E.; Cardinale, D.; Swarén, M.; Wedholm, L.; Cuartero, M.; Crespo, G. A. Fully Integrated Wearable Device for Continuous Sweat Lactate Monitoring in Sports. ACS Sens 2023, 8 (6), 2401–2409.
12. [12] Zhang, Q.; Jiang, D.; Xu, C.; Ge, Y.; Liu, X.; Wei, Q.; Huang, L.; Ren, X.; Wang, C.; Wang, Y. Wearable Electrochemical Biosensor Based on Molecularly Imprinted Ag Nanowires for Noninvasive Monitoring Lactate in Human Sweat. Sens Actuators B Chem. 2020, 320, No. 128325.
13. Kumar, N.; Lin, Y.-J.; Huang, Y.-C.; Liao, Y.-T.; Lin, S.-P. Detection of Lactate in Human Sweat via Surface-Modified, ScreenPrinted Carbon Electrodes. Talanta 2023, 265, No. 124888.
14. Shapiro, F.; Silanikove, N. Rapid and Accurate Determination of D- and l-Lactate, Lactose and Galactose by Enzymatic Reactions Coupled to Formation of a Fluorochromophore: Applications in Food Quality Control. Food Chem. 2010, 119 (2), 829–833.
15. Biswas, A.; Bornhoeft, L. R.; Banerjee, S.; You, Y.-H.; McShane, M. J. Composite Hydrogels Containing Bioactive Microreactors for Optical Enzymatic Lactate Sensing. ACS Sens 2017, 2 (11), 1584– 1588.

16. Baretta, R.; Raucci, A.; Cinti, S.; Frasconi, M. Porous Hydrogel Scaffolds Integrating Prussian Blue Nanoparticles: A Versatile Strategy for Electrochemical (Bio)Sensing. Sens Actuators B Chem. 2023, 376, No. 132985.
17. Crapnell, R.; Hudson, A.; Foster, C.; Eersels, K.; Grinsven, B.; Cleij, T.; Banks, C.; Peeters, M. Recent Advances in Electrosynthesized Molecularly Imprinted Polymer Sensing Platforms for Bioanalyte Detection. Sensors 2019, 19 (5), 1204.
18. Pirot, S. M.; Omer, K. M. Surface Imprinted Polymer on Dual Emitting MOF Functionalized with Blue Copper Nanoclusters and Yellow Carbon Dots as a Highly Specific Ratiometric Fluorescence Probe for Ascorbic Acid. Microchemical Journal 2022, 182, No. 107921.
19. Yang, W.; Ma, Y.; Sun, H.; Huang, C.; Shen, X. Molecularly Imprinted Polymers Based Optical Fiber Sensors: A Review. TrAC Trends in Analytical Chemistry 2022, 152, No. 116608.
20. Pirot, S. M.; Omer, K. M.; Alshatteri, A. H.; Ali, G. K.; Shatery, O. B. A. Dual-Template Molecularly Surface Imprinted Polymer on Fluorescent Metal-Organic Frameworks Functionalized with Carbon Dots for Ascorbic Acid and Uric Acid Detection. Spectrochim Acta A Mol. Biomol Spectrosc 2023, 291, No. 122340.
21. Ali, G. K.; Omer, K. M. Molecular Imprinted Polymer Combined with Aptamer (MIP-Aptamer) as a Hybrid Dual Recognition Element for Bio(Chemical) Sensing Applications. Review. Talanta 2022, 236, No. 122878.
22. Liu, Y.; Dykstra, G. Recent Progress on Electrochemical (Bio)Sensors Based on Aptamer-Molecularly Imprinted Polymer Dual Recognition. Sensors and Actuators Reports 2022, 4, No. 100112. [23] Moreira Gonçalves, L. Electropolymerized Molecularly Imprinted Polymers: Perceptions Based on Recent Literature for Soon-to-Be World-Class Scientists. Curr. Opin Electrochem 2021, 25, 100640. [24] Zidaric, ˇ T.; Majer, D.; Maver, T.; Finsgar, ˇ M.; Maver, U. The Development of an Electropolymerized, Molecularly Imprinted Polymer (MIP) Sensor for Insulin Determination Using SingleDrop Analysis. Analyst 2023, 148 (5), 1102–1115.
23. Dykstra, G.; Reynolds, B.; Smith, R.; Zhou, K.; Liu, Y. Electropolymerized Molecularly Imprinted Polymer Synthesis Guided by an Integrated Data-Driven Framework for Cortisol Detection. ACS Appl. Mater. Interfaces 2022, 14 (22), 25972–25983.
24. Xuan, X.; Pérez-Rafols, ` C.; Chen, C.; Cuartero, M.; Crespo, G. A. Lactate Biosensing for Reliable On-Body Sweat Analysis. ACS Sens 2021, 6 (7), 2763–2771.
25. Matos-Peralta, Y.; Antuch, M. Review‐Prussian Blue and Its Analogs as Appealing Materials for Electrochemical Sensing and Biosensing. J. Electrochem. Soc. 2020, 167 (3), No. 037510.
26. Payne, M. E.; Zamarayeva, A.; Pister, V. I.; Yamamoto, N. A. D.; Arias, A. C. Printed, Flexible Lactate Sensors: Design Considerations Before Performing On-Body Measurements. Sci. Rep 2019, 9 (1), 13720.
27. Karyakin, A. A. Prussian Blue and Its Analogues: Electrochemistry and Analytical Applications. Electroanalysis 2001, 13, 813.
28. Tang, W.; Yin, L.; Sempionatto, J. R.; Moon, J.-M.; Teymourian, H.; Wang, J. Touch-Based Stressless Cortisol Sensing. Advanced materials 2021, 33 (18), 2008465.
29. Rattu, G.; Krishna, P. M. TiO2 Nanoparticles Reagent Based Nonenzymatic Label-Free Optical Sensor for the Rapid Detection of L-Lactate in Apple Juice. Sensors and Actuators Reports 2021, 3, No. 100067.
30. Dhiraj, H. S.; Ishizuka, F.; Saeed, M.; Elshaer, A.; Zetterlund, P. B.; Aldabbagh, F. Lactate and Glucose Responsive Boronic AcidSubstituted Amphiphilic Block Copolymer Nanoparticles of High Aspect Ratio. Eur.
31. Tang, W.; Yin, L.; Sempionatto, J. R.; Moon, J.; Teymourian, H.; Wang, J. Touch-Based Stressless Cortisol Sensing. Adv. Mater. 2021, 33 (18), 2008465.
32. Karyakin, A. A. Prussian Blue and Its Analogues: Electrochemistry and Analytical Applications. Electroanalysis 2001, 13 (10), 813–819.
33. Dykstra, G.; Reynolds, B.; Smith, R.; Zhou, K.; Liu, Y. Electropolymerized Molecularly Imprinted Polymer Synthesis Guided by an Integrated Data-Driven Framework for Cortisol Detection. ACS Appl. Mater. Interfaces 2022, 14 (22), 25972–25983.
34. Tajik, S.; Beitollahi, H.; Nejad, F. G.; Shoaie, I. S.; Khalilzadeh, M. A.; Asl, M. S.; Van Le, Q.; Zhang, K.; Jang, H. W.; Shokouhimehr, M. Recent Developments in Conducting Polymers: Applications for Electrochemistry. RSC Adv. 2020, 10 (62), 37834–37856.
35. Gomes, J. C. M.; Souza, L. C.; Oliveira, L. C. SmartSPR Sensor: Machine Learning Approaches to Create Intelligent Surface Plasmon Based Sensors. Biosens Bioelectron 2021, 172, No. 112760.
36. Abu-Alsoud, G. F.; Hawboldt, K. A.; Bottaro, C. S. Comparison of Four Adsorption Isotherm Models for Characterizing Molecular Recognition of Individual Phenolic Compounds in Porous TailorMade Molecularly Imprinted Polymer Films. ACS Appl. Mater. Interfaces 2020, 12 (10), 11998–12009.
37. Tamayo, F. G.; Casillas, J. L.; Martin-Esteban, A. Evaluation of New Selective Molecularly Imprinted Polymers Prepared by Precipitation Polymerisation for the Extraction of Phenylurea Herbicides. J. Chromatogr A 2005, 1069 (2), 173–181.
38. T.P. Chendrimada, K.J. Finn, X. Ji, D. Baillat, R.I. Gregory, S.A. Liebhaber, et al., MicroRNA silencing through RISC recruitment of eIF6, Nature 447 (2007) 823–828.
39. [41] L. He, G.J. Hannon, MicroRNAs: small RNAs with a big role in gene regulation, Nat. Rev. Genet. 5 (2004) 522–531.

40. C.D. Johnson, A. Esquela-Kerscher, G. Stefani, M. Byrom, K. Kelnar, D. Ovcharenko, et al., The let-7 microRNA represses cell proliferation pathways in human cells, Cancer Res. 67 (2007) 7713–7722.
41. Z. Qing, J. Xu, J. Hu, J. Zheng, L. He, Z. Zou, et al., In situ amplification-based imaging of RNA in living cells, Angew. Chem., Int. Ed. Engl. 58 (2019) 11574–11585.
42. P. Gai, C. Gu, T. Hou, F. Li, Integration of biofuel cell-based self-powered biosensing and homogeneous electrochemical strategy for ultrasensitive and easyto-use bioassays of MicroRNA, ACS Appl. Mater. Interfaces 10 (2018) 9325–9331.
43. L.M. Li, Z.B. Hu, Z.X. Zhou, X. Chen, F.Y. Liu, J.F. Zhang, et al., Serum microRNA profiles serve as novel biomarkers for HBV infection and diagnosis of HBV-positive hepatocarcinoma, Cancer Res. 70 (2010) 9798–9807.
44. S. Song, N. Li, L. Bai, P. Gai, F. Li, Photo-assisted robust anti-interference selfpowered biosensing of MicroRNA based on Pt–S bonds and the inorganic–organic hybridization strategy, Anal. Chem. 94 (2022) 1654–1660.
45. J. Du, S. Yang, D. An, F. Hu, W. Yuan, C. Zhai, et al., BMP-6 inhibits microRNA-21 expression in breast cancer through repressing deltaEF1 and AP-1, Cell Res. 19 (2009) 487–496.
46. A.R. Cardoso, F.T.C. Moreira, R. Fernandes, M.G.F. Sales, Novel and simple electrochemical biosensor monitoring attomolar levels of miRNA-155 in breast cancer, Biosens, Bioelectron 80 (2016) 621–630.
47. K. Dhuri, R.R. Gaddam, A. Vikram, F.J. Slack, R. Bahal, Therapeutic potential of chemically modified, synthetic, triplex peptide nucleic acid-based oncomir inhibitors for cancer therapy, Cancer Res. 81 (2021) 5613–5624.
48. X. Zhang, M. Li, K. Zuo, D. Li, M. Ye, L. Ding, et al., Upregulated miR-155 in papillary thyroid carcinoma promotes tumor growth by targeting APC and activating Wnt/beta-catenin signaling, J. Clin. Endocrinol. Metab. 98 (2013) E1305–E1313.
49. S.M. Oana, B. Claudia, R.A. Lelia, M. Simona, C. Claudia, D.E. Daniela, Differential expression of tissular miRNA-155 in pediatric gastritis, J. Clin. Med. 11 (2022) 3351.
A. Asadirad, K. Baghaei, S.M. Hashemi, S. Dehnavi, H. Ghanbarian, E. Mortaz, et al., Dendritic cell immunotherapy with miR-155 enriched tumor-derived exosome suppressed cancer growth and induced antitumor immune responses in murine model of colorectal cancer induced by CT26 cell line, Int. Immunopharm. 104 (2022), 108493.
50. L.T. Pan, Y. Sheung, W.P. Guo, Z.B. Rong, Z.M. Cai, Hedyotis diffusa plus scutellaria barbata induce bladder cancer cell apoptosis by inhibiting akt signaling pathway through downregulating miR-155 expression, Evid Based Complement Alternat. Med. (2016), 9174903, 2016.
51. Y. Chen, L. Teng, W. Liu, Y. Cao, D. Ding, W. Wang, et al., Identification of biological targets of therapeutic intervention for clear cell renal cell carcinoma based on bioinformatics approach, Cancer Cell Int. 16 (2016) 16.
52. K.M. Koo, L.G. Carrascosa, M.J. Shiddiky, M. Trau, Poly(A) extensions of miRNAs for amplification-free electrochemical detection on screen-printed gold electrodes, Anal. Chem 88 (2016) 2000–2005. [56] H.A. Rafiee-Pour, M. Behpour, M. Keshavarz, A novel label-free electrochemical miRNA biosensor using methylene blue as redox indicator: application to breast cancer biomarker miRNA-21, Biosens. Bioelectron. 77 (2016) 202–207.
53. X. Deng, S. Wu, S. Zang, X. Liu, Y. Ma, PDA-PEI-Copolymerized nanodots with tailorable fluorescence emission and quenching properties for the sensitive ratiometric fluorescence sensing of miRNA in serum, Anal. Chem 94 (2022) 14546–14553.
54. M. Laurenti, M. Paez-Perez, M. Algarra, P. Alonso-Cristobal, E. Lopez-Cabarcos, D. Mendez-Gonzalez, et al., Enhancement of the upconversion emission by visibleto-near-infrared fluorescent graphene quantum dots for miRNA detection, ACS Appl. Mater. Interfaces 8 (2016) 12644–12651.
55. S. Gong, X. Wang, P. Zhou, W. Pan, N. Li, B. Tang, AND logic-gate-based CRISPR/ Cas12a biosensing platform for the sensitive colorimetric detection of dual miRNAs, Anal. Chem 94 (2022) 15839–15846.
56. H. Lee, J. Lee, S.G. Lee, P.S. Doyle, Hydrogel-based colorimetric assay for multiplexed MicroRNA detection in a microfluidic device, Anal. Chem 92 (2020) 5750–5755.
57. [61] M.Y. Wang, W.J. Jing, L.J. Wang, L.P. Jia, R.N. Ma, W. Zhang, et al., Electrochemiluminescence detection of miRNA-21 based on dual signal amplification strategies: duplex-specific nuclease -mediated target recycle and nicking endonuclease-driven 3D DNA nanomachine, Biosens. Bioelectron. 226 (2023), 115116.
58. J. Wu, W. Lv, Q. Yang, H. Li, F. Li, Label-free homogeneous electrochemical detection of MicroRNA based on target-induced anti-shielding against the catalytic activity of two-dimension nanozyme, Biosens. Bioelectron. 171 (2021), 112707.
59. L. Wang, P. Liu, Z. Liu, K. Zhao, S. Ye, G. Liang, et al., Simple tripedal DNA walker prepared by target-triggered catalytic hairpin assembly for ultrasensitive electrochemiluminescence detection of MicroRNA, ACS Sens. 5 (2020) 3584–3590.
60. P. Yin, H.M. Choi, C.R. Calvert, N.A. Pierce, Programming biomolecular selfassembly pathways, Nature 451 (2008) 318–322.

61. Y. Zhao, J. Xiang, H. Cheng, X. Liu, F. Li, Flexible photoelectrochemical biosensor for ultrasensitive microRNA detection based on concatenated multiplex signal amplification, Biosens. Bioelectron. 194 (2021), 113581.
62. H. Chai, W. Cheng, D. Jin, P. Miao, Recent progress in DNA hybridization chain reaction strategies for amplified biosensing, ACS Appl. Mater. Interfaces 13 (2021) 38931–38946.
63. F. Jin, D. Xu, A cascaded DNA circuit in bead arrays for quantitative single-cell MicroRNA analysis, Anal. Chem 93 (2021) 11617–11625. [29] Y. Peng, Y. Pan, Y. Han, Z. Sun, M. Jalalah, M.S. Al-Assiri, et al., Direct analysis of rare circulating tumor cells in whole blood based on their controlled capture and release on electrode surface, Anal. Chem 92 (2020) 13478–13484.
64. Z. Fan, B. Yao, Y. Ding, D. Xu, J. Zhao, K. Zhang, Rational engineering the DNA tetrahedrons of dual wavelength ratiometric electrochemiluminescence biosensor for high efficient detection of SARS-CoV-2 RdRp gene by using entropy-driven and bipedal DNA walker amplification strategy, Chem. Eng. J. 427 (2022), 131686.
65. Q.M. Feng, Y.H. Guo, J.J. Xu, H.Y. Chen, Self-assembled DNA tetrahedral scaffolds for the construction of electrochemiluminescence biosensor with programmable DNA cyclic amplification, ACS Appl. Mater. Interfaces 9 (2017) 17637–17644.
66. L. Yu, L. Zhu, M. Yan, S. Feng, J. Huang, X. Yang, Electrochemiluminescence biosensor based on entropy-driven amplification and a tetrahedral DNA nanostructure for miRNA-133a detection, Anal. Chem 93 (2021) 11809–11815.
67. H. Chai, Y. Tang, Z. Guo, P. Miao, Ratiometric electrochemical switch for circulating tumor DNA through recycling activation of blocked DNAzymes, Anal. Chem 94 (2022) 2779–2784.
68. Y. Chang, S. Xu, Y. Li, W. Hu, H. Li, R. Yuan, et al., DNA three-way junction with multiple recognition regions mediated an unconfined DNA walker for electrochemical ultrasensitive detection of miRNA-182-5p, Anal. Chem. 93 (2021) 12981–12986. 1285 (2024).
69. S. Gupta, S.V. Sandhu, H. Bansal, D. Sharma, Comparison of salivary and serum glucose levels in diabetic patients, J. Diabetes. Sci. Technol. 9 (2014) 91–96.
70. S. Zhang, C. Liu, X. Sun, W. Huang, Current development of materials science and engineering towards epidermal sensors, Prog. Mater. Sci. 128 (2022) 100962.
71. M.-H. Lin, S. Gupta, C. Chang, C.-Y. Lee, N.-H. Tai, Carbon nanotubes/ polyethylenimine/glucose oxidase as a non-invasive electrochemical biosensor performs high sensitivity for detecting glucose in saliva, Microchem. J. 180 (2022).
72. L.-H. Fu, C. Qi, J. Lin, P. Huang, Catalytic chemistry of glucose oxidase in cancer diagnosis and treatment, Chem. Soc. Rev. 47 (2018) 6454–6472.
73. N. Xuan Viet, Enzyme-free glucose sensor based on micro-nano dualporous goldmodified screen-printed carbon electrode, Int. J. Electrochem. Sci. (2018) 8633–8644.
74. L.F. de Lima, et al., Enzymeless glucose sensor based on disposable Ecoflex®/ graphite thermoplastic composite substrate modified with Au@GQDs, Sens. Actuators. Rep. 4 (2022) 100102. [79] Y. Jiang, et al., Facet-dependent Cu2O electrocatalysis for wearable enzyme-free smart sensing, ACS. Catal. 11 (2021) 2949–2955.
75. X. Lin, Y. Wang, M. Zou, T. Lan, Y. Ni, Electrochemical non-enzymatic glucose sensors based on nano-composite of Co3O4 and multiwalled carbon nanotube, Chin. Chem. Lett. 30 (2019) 1157–1160.
76. Q. Dong, H. Ryu, Y. Lei, Metal oxide based non-enzymatic electrochemical sensors for glucose detection, Electrochim. Acta 370 (2021).
77. H. Wei, et al., Dendritic core-shell copper-nickel alloy@metal oxide for efficient non-enzymatic glucose detection, Sensors Actuators B: Chem. 337 (2021).