Bioelectrochemical platforms to study and detect emerging pathogens

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The ongoing SARS-CoV-2 pandemic has emphasized the importance of technologies to rapidly detect emerging pathogens and understand their interactions with hosts. Platforms based on the combination of biological recognition and electrochemical signal transduction, generally termed bioelectrochemical platforms, offer unique opportunities to both sense and study pathogens. Improved bio-based materials have enabled enhanced control over the biotic–abiotic interface in these systems. These improvements have generated platforms with the capability to elucidate biological function rather than simply detect targets. This advantage is a key feature of recent bioelectrochemical platforms applied to infectious disease. Here, we describe developments in materials for bioelectrochemical platforms to study and detect emerging pathogens. The incorporation of host membrane material into electrochemical devices has provided unparalleled insights into the interaction between viruses and host cells, and new capture methods have enabled the specific detection of bacterial pathogens, such as those that cause secondary infections with SARS-CoV-2. As these devices continue to improve through the merging of hi-tech materials and biomaterials, the scalability and commercial viability of these devices will similarly improve.

Introduction

The COVID-19 pandemic, caused by Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2), has completely upended daily life since its initial identification in 2019. As of May 2021, the pathogen has infected more than 150 million people and has caused nearly 3.5 million deaths globally.¹ In addition to the acute symptoms of this deadly virus, long-term effects that remain for months following the initial infection impact a large percentage of those with COVID-19 (estimated 50–80% of patients).² In addition, dangerous bacterial secondary infections are prevalent, especially among those hospitalized because of SARS-CoV-2.³ These dangers have reinvigorated interest in the development of universal diagnostics platforms that can be rapidly adapted for the detection and identification of emergent and novel pathogens.

More specifically, the ongoing COVID-19 pandemic has spurred significant interest in the development of portable, rapid, and inexpensive technologies for pathogen detection. Reverse transcription polymerase chain reaction (RT-PCR) and rapid antigen testing have both been widely used during the pandemic for rapid diagnosis of an active infection or the identification of antibodies against the virus due to either a

prior infection or vaccination.⁴ However, both of these methods require costly materials, major pieces of scientific equipment, and highly skilled personnel, in addition to laboratory facilities.⁵ Moreover, pharyngeal tests for COVID-19 require high-quality specimens with sufficient concentrations of intact viral RNA for detection.⁶ Thus, the variation in viral titers in infected patients can lead to false-negative results. Similarly, the complexity of biological matrices from which pathogens are detected and lack of specificity and sensitivity in assays to detect pathogens can lead to both false-positive and false-negative results. It is, therefore, important to incorporate functional biological components, as assays based on biological activity are the most effective and fastest methods to diagnose an infection. Yet most diagnostics are not based on function or activity, making new methods for rapid, sensitive testing incorporating these features urgently needed.

Biosensors have arisen as valuable alternatives to conventional diagnostics, as they offer rapid turnaround, compatibility with point-of-care deployment, and low cost.⁴ In a typical biosensor, a biorecognition element (i.e., an antibody, protein, nucleic acid aptamer, or DNA or RNA sequence)⁵ is integrated with a transducer that produces a signal upon the binding of

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the target to the recognition element (**Figure 1**). Although optical and mechanical biosensors have been developed, electrochemical biosensors are favorable due to their low-cost assembly^{6,7} and rapid, label-free, quantitative readout.⁸ These platforms are also compatible with multiplexing, miniaturization, and automation.⁹ Thus, electrochemical biosensors are highly useful for the study of host–pathogen interactions, as well as point-of-care detection of these pathogens. A number of recent reviews, most notably that of Ruiz de Egulaz et al. and Bukkittgar et al., have cataloged the advances in the methods and materials used for these sensors.^{7,8}

Functional assays at the abiotic–biotic interface

One key aspect of electrochemical biosensors, and bioelectrochemical systems more generally, is the interface between the biotic components, such as the recognition elements, and the abiotic components, most often the electrode surface. As the components of these systems become ever more complex and advanced, this interface becomes increasingly important.⁹ Moreover, one consistent challenge in biosensing is the

development of systems that can monitor biological signals without perturbing or changing the system of interest, which can cause artifacts in sensor signals and prevent the production of a biologically relevant readout. Thus, there is increasing interest in bioelectrochemical platforms based on functional assays and the behavior and activity of biomolecules rather than simply monitoring passive binding interactions. For this reason, focus has shifted from electronic biosensors made from inorganic materials functionalized with biorecognition elements to the development of more effective biomimetic interfaces.

Studying emerging pathogens electrochemically

Conventionally, host–pathogen interactions are either studied *in cellulo* (which can make elucidating specific mechanisms challenging)^{10,11} or on platforms in which a specific receptor is bound to a sensor surface and simple binding events to the pathogen are monitored.¹² This second class of platform can oversimplify interactions, which can lead to the loss of key biological insights. Unlike the aforementioned platforms, those that incorporate biomimetic or bioderived cell membranes more closely imitate the natural lipid environment of a cell membrane. These technologies therefore provide biological information and native host features unavailable with conventional biosensing platforms.

Because lipid bilayers supported on electrodes facilitate the movement of proteins embedded in the membranes, these systems are well suited to the analysis of multivalent interactions between such mobile components and targets.¹³ Recent studies have focused on the development of biomimetic interfaces using synthetic cell membranes. The first of these early studies employed traditional supported lipid bilayers (SLBs).¹⁴ These bilayers, often generated through solvent-assisted lipid bilayer formation, enabled multiplexing and fluorescence-based studies of protein diffusion in those membranes.^{15,16} However, these models are highly simplified as compared to native membranes. The utility of these lipid layers has been limited due to the lack of native protein incorporation, as these proteins are often the key targets or receptors involved in the binding event being observed. To overcome this limitation, some studies have used peripheral or self-inserting membrane proteins introduced after a bilayer has been formed. This improves the compositional complexity of the membrane but continues to limit analysis to a small group of proteins.

To generate bilayers with higher biological relevance by increasing the complexity of incorporated membrane proteins, the Daniel lab has developed a novel method for the delivery of proteins to an SLB platform using cell blebs, or spherical membrane protrusions that are

Figure 1. Overview of key components of electrochemical biosensors. Biorecognition elements (i.e., antibodies, nucleotide aptamers, DNA probes, or enzymes) immobilized on an electrode bind a target molecule. This binding event is measured electrochemically using a potentiostat, which produces an electrochemical signal. In the example display shown, the binding of the analyte results in an increase in impedance.

formed when sections of the cell membrane detach from the actin cytoskeleton (**Figure 2a**).^{17,18} Investigations of this material have shown that membrane proteins delivered to an SLB using this mechanism not only retain crucial lipid interactions with the native membrane but remain functional as viral receptors.¹⁸ Delivering membrane proteins to the SLB in this manner allows for the retention of the fluidity, orientation, and function of these proteins that were originally derived from cells.

Figure 2. Formation and sensing of a bleb-supported lipid bilayer (SLB). (a) Illustration of the formation of an SLB derived from cell blebs. The SLBs shown contain an assortment of transmembrane proteins. (b) Optical detection of lipids in a bleb SLB using fluorescent lipids, proteins, or antibodies. (c) Electrochemical detection of a bleb SLB on an organic electrochemical transistor device.

Despite advances in bleb-derived bilayers, challenges remain with this technology from the abiotic side. One significant challenge lies in the generation of a biotic–abiotic interface that allows for the transduction of signals across the membrane to characterize the lipid bilayer within the SLB. Further, if this signal transduction is not optimized, it can be challenging to elucidate biologically relevant information from the SLB. Early work with SLBs was often performed on surfaces such as mica,¹⁹ silicon dioxide,²⁰ and glass²¹ due to the ease of their formation on these surfaces. However, these surfaces have significant drawbacks for sensing modalities beyond fluorescence imaging, including electrochemistry. These surfaces are electrically insulating, and their rigidity hinders the mobility, insertion, and function of transmembrane proteins.²² Additionally, limited headspace between the lipid headgroups and the surface can sometimes denature the proteins that protrude beneath the bilayer.²³ These challenges limited the sensing methods that were compatible with SLBs.

To address this problem, a new abiotic substrate material was needed at the interface of the sensor. Because they are pliable and inert, polymers have offered a useful alternative to the aforementioned rigid substrates. Further, conducting polymers can act both as a cushion to preserve native protein function and also transmit signals across the membrane. Because of these advantages, in addition to the low cost and biocompatibility of these materials, interfacing conducting polymers with lipid structures is well represented in the literature. Not only have conducting polymers such as poly(3,4-ethylenedioxythiophene) poly(styrene sulfonate) PEDOT:PSS been used for a platform for vesicle fusion²⁴ and eukaryotic cell growth,²⁵ but they have also been incorporated into liposomes.²⁶ Work by Daniels and Owens has shown that the assembly of SLBs on organic conductive polymers such as PEDOT:PSS,²³ along with the coupling of this material to organic electrochemical transistors (OECTs), can generate a sensor to enable monitoring of pathogen–host interactions that maintains native-like protein mobility and excellent electronic properties (Figure 2).^{22,27} This recent progress in the development of biomimetic interfaces has led to fundamental insights into the structure and function of viruses such as Influenza²⁸ and the herpes simplex virus (HSV),²⁹ and could be used to provide similar insights in viruses such as SARS-CoV-2. These platforms have also been used as bioanalytical tools for antiviral drug studies.^{30,31} The different methods used to form SLBs are summarized in **Table I**.

Biomembrane-functionalized sensors serve as an optimal platform to embed host membrane proteins (such as ACE2) within the membrane of the device, allowing for viral binding and fusion at the surface. For COVID-19, specific recognition of the receptor by the spike protein of the virus can induce changes in the electrical properties of the OECT channel, allowing for rapid, real time detection of binding and fusion events. To further capture functional features and biological complexity in these platforms, introducing other critical interaction partners is key. One example is TMPRSS2,

Table I. Summary of supported lipid bilayer (SLB) formulations for the study of Transmembrane proteins and host–pathogen interactions.

SLB type	Detection	Shortcoming	Ref.
Solvent-assisted lipid bilayer formation	Protein diffusion in membrane	Overly simplified model due to lack of native protein incorporation in the membrane	15, 16
Protein delivery into SLB via cell bleb on mica, silicon dioxide, glass	Allows for study of proteins whose fluidity, orientation, and function has been better preserved	Substrates are electrically insulating and rigid, thereby hindering the insertion, function, and mobility of transmembrane proteins	17–23
Protein delivery into SLB via cell bleb on conductive polymers such as PEDOT:PSS	Allows for detection of pathogen–host interactions that preserve native-like protein mobility and superior electronic properties	n/a	22, 23, 27–31

a membrane-bound protease that must cleave the spike for fusion to occur, which can be expressed in the membranes on the sensor surface. With this increased compositional complexity, it is clear why the ability of proteins to diffuse within the membrane is essential to faithfully replicate the biological milieu. Finally, we note that SLBs have been used to evaluate the role of calcium in the fusion of COVID-19 and to investigate the use of calcium channel blockers as a therapeutic.³¹ The expansion of these investigations to bioelectronic platforms would allow for rapid, accurate evaluation of potential drug candidates that are needed during a pandemic.

Detecting emerging pathogens electrochemically

Developing rapid, low-cost, point-of-care diagnostic technologies is urgently needed for the effective monitoring and control over COVID-19 spread. Although significant advances have been made in diagnostics for COVID-19, the majority of approved platforms continue to fall short because they require multi-step preparations, costly equipment, and trained personnel for both sample preparation and evaluation. Ideal point-of-care tests must both identify and quantify the pathogen in a single, easy-to-use device. Such sensors are especially important for COVID-19 to prevent community spread caused by asymptomatic patients.

Conventional diagnostics for infections are most often either genotypic methods that detect pathogen genes or immunological assays such as ELISA (enzyme-linked immunosorbent assay) and are based on antigen–antibody interactions. The major drawback of such techniques is that they are time-consuming and require centralized laboratory facilities.^{32,33} Further, genotypic methods are not capable of detecting unknown or emerging pathogens for which a target gene is not yet known or defined.³² In contrast, electrochemical biosensors can perform quantitative measurements at the point-of-care using low-cost, simple hardware.^{12,33} Additionally, functional electrochemical assays enable detection even in the absence of knowledge of the pathogen, as they are phenotypic.

Electrochemical biosensors are important technologies for point-of-care diagnostics, especially for self-testing and self-managed medicine, due to their speed and sensitivity, low cost, and inexpensive, user-friendly operation.³⁴ These technologies

have the potential to make significant commercial and clinical impacts. By innovating both the biological components of these sensors and the abiotic interface, electrochemical biosensors are emerging as ideal devices for direct pathogen detection. Advantages of conventional electrochemical biosensors to monitor infectious diseases has been thoroughly reviewed.^{35,36} However, conventional biorecognition elements increase the cost and complexity of sensors while decreasing their stability and necessitating highly controlled conditions for storage and transport. Further, these biorecognition elements cannot covalently capture pathogens, which can lead to variability in measurements and the inability to distinguish between living and dead cells for infectious microbes. Here, we focus on one key area where improving the abiotic–biotic interface has significantly improved the efficacy of detection: functional biosensors. These are sensors that rely on some inherent biological activity to detect a target pathogen, which often increases the sensitivity and specificity of the platform.

Recently, a functional electrochemical sensor was reported to detect pathogenic strains of *Escherichia coli*,³⁷ which are the most common cause of urinary tract infections (UTIs)^{38,39} and are a major contributor to foodborne illnesses.^{40–43} Antibody- and aptamer-based electrochemical sensors have been reported to monitor these pathogens, with detection limits as low as 10 colony-forming units (CFU)/mL.^{44–50} However, limitations with these platforms often hindered their ability to reproducibly detect pathogens in complex matrices. Furst and co-workers developed an electrochemical sensor to detect *E. coli* from food samples and bodily fluids based on bacterial incorporation of a synthetic amino acid and subsequent covalent cell capture at an electrode (**Figure 3a**). Because the *E. coli* had to incorporate a synthetic amino acid via peptidoglycan remodeling for electrode capture, only live cells were captured on the electrode, thereby increasing assay sensitivity and allowing for a detection limit of 12 CFU/mL (an order of magnitude below the infective dose),⁵¹ and the assay was specific for live cells. Thus, the need for microbial activity to incorporate the synthetic amino acid provides a functional assay that enables the detection of only live pathogens.

Similarly, diagnostics that take advantage of the inherent activity of endonucleases, namely those based on CRISPR, are finding increasing success.⁵² Companies such as Sherlock

Biosciences are developing colorimetric assays based on this activity for SARS-CoV-2 based on sequence-specific interactions between guide RNA and target genetic material. Despite the promise of these technologies, though, viable technologies remain relatively limited. Combining the trans-cleavage activity of these endonucleases with electrochemical readout

Table II. Examples of functional assays used for pathogen detection.

Functional assay	Detection	References
Cell capture via synthetic amino acid via peptidoglycan remodeling	<i>E. coli</i> , 12 CFU/mL	52
Electrochemical CRISPR-Cas12a-based nucleic acid detection	HPV DNA, 1.2×10^4 copies	54, 55

has enabled the development of inexpensive platforms for the detection of pathogenic genetic material. CRISPR-Cas12a is commonly used to make electrochemical biosensors. CRISPR-Cas12a is a RNA-guided enzyme that exhibits indiscriminate endonuclease activity upon binding to a specific sequence of double-stranded DNA. By changing the gRNA sequence that guides the Cas12a enzyme, one can engineer the Cas12a enzyme to bind to DNA specific to a pathogen such as HPV. One can confirm the presence of HPV by electrochemically detecting HPV-activated endonuclease activity of Cas12a.⁵² One of the first reports of this is from Liu and co-workers.⁵³ They successfully detected human papillomavirus 16 (HPV-16) and parvovirus B19 (PB-19) genetic material with picomolar sensitivity. However, due to the sensitivity required for viral detection in clinical samples, additional innovation was still needed to reach clinically relevant levels using this strategy.

Recently, Klapperich and co-workers improved detection with this general strategy through the incorporation of innovative, inexpensive electrodes and dual biological activity for signal amplification.⁵⁴ They reported an electrochemical platform for the detection of viral pathogens, including HPV-16, HPV-18, and HIV using loop-mediated isothermal amplification (LAMP), which is a nucleic acid amplification technique that operates at a single temperature, the endonuclease activity of Cas12a, and inexpensive, hand-fabricated gold leaf electrodes (Figure 3b). Although this is not a functional assay for the detection of intact, active virus, the researchers achieved improved sensitivity and specificity from clinical samples by incorporating functional DNA polymerase and Cas12a endonuclease activities. By innovating both the biotic and abiotic assay components, they were able to detect clinically relevant loads of human papillomavirus (HPV) from cervical swabs. Such innovation in both the application of polymerase and endonuclease activity and novel materials for inexpensive, easily fabricated electrodes are essential for the continuing improvement of these technologies.

The functional assays described are summarized in Table II. Despite the potential impact of functional electrochemical biosensors to improve on SARS-CoV-2 diagnostics, the applications of electrochemistry for such diagnostics has remained mostly in the traditional detection realm. Several effective sensors for electrochemical detection of

SARS-CoV-2 biomarkers have been developed but are limited by stability and sensitivity in complex matrices.⁴ We anticipate that continued development of more stable biomaterials for stability following electrode modification combined with functional assays for viral detection to improve sensitivity and selectivity will enable the translation and broad application of these diagnostic technologies.

Conclusions and outlook

Bioelectronic platforms are critical for both understanding the fundamental biology of emerging pathogens and combatting their spread through rapid, point-of-care diagnostics. The success of these technologies, though, depends on the biological materials, the electrode materials, and (most importantly) the abiotic–biotic interface between these components. Key innovations in these areas have led to significant technical advances in the recapitulation of membrane proteins in SLB bioelectronic platforms. These improvements have enabled insights into key host–pathogen interactions. Similarly, by incorporating functional biological assays into electrochemical biosensors, along with advances in electrode fabrication and deployment, these technologies have matured to a stage where they can be deployed with samples from patients, including bodily fluids. More work needs to be done to enable mass production, long-term storage and commercialization of these devices. For example, PEDOT:PSS causes degradation of devices during long-term storage; more work is being done to explore alternative materials and barrier layers to prevent said degradation.⁵⁵ In the next few years, as innovation continues to occur through improvements in organic and polymeric conductive materials, which are often more biocompatible than solid metals, these technologies will advance to a point where they can be commercialized and broadly implemented before an emerging pathogen repeats the global transmission seen with COVID-19.

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Conflict of interest

On behalf of all authors, the corresponding author states that there is no conflict of interest.

References

1. Johns Hopkins University, *COVID-19 Map—Johns Hopkins Coronavirus Resource Center* (Johns Hopkins Coronavirus Resource Center, Baltimore, 2020)
2. T. Greenhalgh, M. Knight, C. A'Court, M. Buxton, L. Husain, *BMJ* **370**, m3026 (2020)
3. R. Mirzaei, P. Goodarzi, M. Asadi, A. Soltani, H. Aljanabi, A.S. Jeda, S. Dashtbin, S. Jalalifar, R. Mohammadzadeh, A. Teimoori, K. Tari, M. Salari, S. Ghisvand, S. Kazemi, R. Yousefimasouf, H. Keyvani, S. Karampoor, *IUBMB Life* **72**, 2097 (2020)
4. A. Aziz, M. Asif, G. Ashraf, U. Farooq, Q. Yang, S. Wang, *Curr. Opin. Colloid Interface Sci.* **52**, 101418 (2021)
5. G. Balkourani, A. Brouzgou, M. Archonti, N. Papandrianos, S. Song, P. Tsiakaras, *Emerging Materials for the Electrochemical Detection of COVID-19* (Elsevier, Amsterdam, The Netherlands, 2021)
6. I.A. Mattioli, A. Hassan, O.N. Oliveira Jr., F.N. Crespilho, *ACS Sens.* **5**, 3655 (2020)
7. M.R. de Equilaz, L.R. Cumba, R.J. Forster, *Electrochem. Commun.* **116**, 106762 (2020)
8. S.D. Bukhtigar, N.P. Shetti, T.M. Aminabhavi, *Chem. Eng. J.* **420**, 127575 (2020)
9. C. Catania, A.A. Karbelkar, A.L. Furst, *Joule* (2021). <https://doi.org/10.1016/j.joule.2021.02.001>
10. A. De Logu, G. Loy, M.L. Pellerano, L. Bonsignore, M.L. Schivo, *Antiviral Res.* **48**, 177 (2000)
11. L.G. Barrientos, B.R. O'Keefe, M. Bray, A. Sanchez, A.M. Gronenborn, M.R. Boyd, *Antiviral Res.* **58**, 47 (2003)
12. A.L. Furst, M.B. Francis, *Chem. Rev.* **119**, 700 (2019)
13. V. Yamazaki, O. Sirenko, R.J. Schafer, J.T. Groves, *J. Am. Chem. Soc.* **127**, 2826 (2005)
14. A.R. Ferhan, B.K. Yoon, S. Park, T.N. Sut, H. Chin, J.H. Park, J.A. Jackman, N. Cho, *Nat. Protoc.* **14**, 2091 (2019)
15. A.P. Wong, J.T. Groves, *Proc. Natl. Acad. Sci. U.S.A.* **99**, 14147 (2002)
16. A.P. Wong, J.T. Groves, *J. Am. Chem. Soc.* **123**, 12414 (2001)
17. M. Dogterom, G. Koenderink, *Nat. Mater.* **10**, 561 (2011)
18. M.J. Richards, C.Y. Hsia, R.R. Singh, H. Haider, J. Kumpf, T. Kawate, S. Daniel, *Langmuir* **32**, 2963 (2016)
19. R.P. Richter, A.R. Brisson, *Biophys. J.* **88**, 3422 (2005)
20. V. Atanasov, N. Knorr, R.S. Duran, S. Ingebrandt, A. Offenhausser, W. Knoll, I. Koper, *Biophys. J.* **89**, 1780 (2005)
21. K.L. Weirich, J.N. Israelachvili, D.K. Fygenson, *Biophys. J.* **98**, 85 (2010)
22. H. Su, H.Y. Liu, A.M. Pappa, T.C. Hidalgo, P. Cavassin, S. Inal, R.M. Owens, S. Daniel, *ACS Appl. Mater. Interfaces* **11**, 43799 (2019)
23. Y. Zhang, S. Inal, C.Y. Hsia, M. Ferro, S. Daniel, R.M. Owens, *Adv. Funct. Mater.* **26**, 7304 (2016)
24. C. Lubrano, G.M. Matrone, G. Iaconis, F. Santoro, *ACS Nano* **14**, 12271 (2020)
25. S.A. Tria, M. Ramuz, M. Huerta, P. Leleux, J. Rivnay, L.H. Jimison, A. Hama, G.G. Malliaras, R.M. Owens, *Adv. Healthc. Mater.* **3**, 1053 (2014)
26. P.K. Johansson, D. Julleson, A. Elfving, S.I. Lin, C. Musumeci, E. Zeglio, F. Elinder, N. Solin, O. Inganas, *Sci. Rep.* **5**, 1 (2015)
27. H.-Y. Liu, A.M. Pappa, A. Pavia, C. Pitsalidis, Q. Thiburse, A. Salles, R.M. Owens, S. Daniel, *Langmuir* **36**, 7325 (2020)
28. H.-L. Hsu, J.K. Millet, D.A. Costello, G.R. Whittaker, S. Daniel, *Sci. Rep.* **6**, 1 (2016)
29. L. Nathan, S. Daniel, Single virion tracking microscopy for the study of virus entry processes in live cells and biomimetic platforms, in *Physical Virology: Virus Structure and Mechanics*, ed. by U.F. Greber (Springer, Berlin, Germany, 2019), pp. 13–43. https://doi.org/10.1007/978-3-030-14741-9_2
30. L. Nathan, A.L. Lai, J.K. Millet, M.R. Straus, J.H. Freed, G.R. Whittaker, S. Daniel, *ACS Infect. Dis.* **6**, 250 (2019)
31. M.R. Straus, M. Bidon, T. Tang, G. Whittaker, S. Daniel, FDA approved calcium channel blockers inhibit SARS-CoV-2 infectivity in epithelial lung cells. *bioRxiv* **382**, 2327 (2020)
32. C.A.D. Burnham, J. Leeds, P. Nordmann, J. O'Grady, J. Patel, *Nat. Rev. Microbiol.* **15**, 697 (2017)
33. A.A. Karbelkar, A.L. Furst, *ACS Infect. Dis.* **6**, 1567 (2020)
34. M. Labib, E.H. Sargent, S.O. Kelley, *Chem. Rev.* **116**, 9001 (2016)
35. F. Mollarasouli, S. Kurbanoglu, S.A. Ozkan, *Biosensors* **9**(3), 86 (2019)
36. J.L. Hammond, N. Formisano, P. Estrela, S. Carrara, *J. Tkac, Essays Biochem.* **60**, 69 (2016)
37. S.H. Klass, L.E. Sofen, Z.F. Hallberg, T.A. Fiala, A.V. Ramsey, N.S. Dolan, M.B. Francis, A.L. Furst, *Chem. Commun.* **57**, 2507 (2021)
38. W.E. Stamm, S.R. Norrby, *J. Infect. Dis.* **183**(Suppl.), S1–4 (2001)
39. A.L. Flores-Mireles, J.N. Walker, M. Caparon, S.J. Hultgren, *Nat. Rev. Microbiol.* **13**, 269 (2015)
40. J. Meng, J.T. LeJeune, T. Zhao, M.P. Doyle, *Food Microbiol.* **2**, 287 (2012). <https://doi.org/10.1128/9781555818463.ch12>
41. E. Scallan, R.M. Hoekstra, F.J. Angulo, R.V. Tauxe, M.A. Widdowson, *Emerg. Infect. Dis.* **17**, 7 (2011)
42. S. Viazis, F. Diez-Gonzalez, Chapter one—Enterohemorrhagic Escherichia coli: The Twentieth Century's Emerging Foodborne Pathogen: A Review. in *Advances in Agronomy*, D.L. Sparks, Ed. (Academic Press, 2011), vol. 11, pp. 1–50
43. S. Park, R.W. Worobo, R.A. Durst, *Crit. Rev. Food Sci. Nutr.* **39**, 481 (1999)

44. M. Amiri, A. Bezaatpour, H. Jafari, R. Boukherroub, S. Szunerits, *ACS Sens.* **3**, 1069 (2018)
45. Y. Wang, E.C. Alocilja, *J. Biol. Eng.* **9**, 16 (2015)
46. M. Barreiros dos Santos, S. Azevedo, J.P. Aguil, B. Prieto-Simon, C. Sporer, E. Torrents, A. Juarez, V. Teixeira, J. Samitier, *Bioelectrochemistry* **101**, 146 (2015)
47. V. Soheili, S.M. Taghdisi, K. Abnous, M. Ebrahimi, *Iran. J. Basic Med. Sci.* **23**, 901 (2020)
48. H. Jayamohan, B.K. Gale, B. Minson, C.J. Lambert, N. Gordon, H.J. Sant, *Sensors (Basel)* **15**, 12034 (2015)
49. H. Kaur, M. Shorie, M. Sharma, A.K. Ganguli, P. Sabherwal, *Biosens. Bioelectron.* **98**, 486 (2017)
50. S.L. Burrs, M. Bhargava, R. Sidhu, J. Kiernan-Lewis, C. Gomes, J.C. Claussen, E.S. McLamore, *Biosens. Bioelectron.* **85**, 479 (2016)
51. N.A. Cornick, A.F. Helgerson, *Appl. Environ. Microbiol.* **70**, 5331 (2004)
52. J.S. Chen, E. Ma, L.B. Harrington, M. Da Cost, X. Tian, J.M. Palefsky, J.A. Doudna, *Science* **360**, 436 (2018)
53. Y. Dai, R.A. Somoza, L. Wang, J.F. Welter, Yan Li, A.I. Caplan, C.C. Liu, *Angew. Chem. Int. Ed.* (2019). <https://doi.org/10.1002/anie.201910772>
54. M. Zamani, J.M. Robson, A. Fan, M.S. Bono, A.L. Furst, C.M. Klapperich, *ACS Cent. Sci.* (2021). <https://doi.org/10.1021/acscentsci.1c00186>
55. J. Cameron, P. Skabara, *Mater. Horizons* **7**, 1759 (2020) □

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