Food Research International 143 (2021) 110246

Contents lists available at ScienceDirect

Food Research International

journal homepage: www.elsevier.com/locate/foodres

Microfluidic devices for multiplexed detection of foodborne pathogens

Xiaoying Han [a,b,1], Yuanhui Liu [a,b,1], Juxin Yin [a], Min Yue [c,d,e,*], Ying Mu [a,*]

[a] Research Centre for Analytical Instrumentation, Institute of Cyber-Systems and Control, State Key Laboratory of Industrial Control Technology, Zhejiang University, Hangzhou 310023, PR China
[b] College of Life Sciences, Zhejiang University, Hangzhou 310058, PR China
[c] Department of Veterinary Medicine & Institute of Preventive Veterinary Sciences, Zhejiang University College of Animal Sciences, Hangzhou 310058, PR China
[d] State Key Laboratory for Diagnosis and Treatment of Infectious Diseases, National Clinical Research Center for Infectious Diseases, National Medical Center for Infectious Diseases, The First Affiliated Hospital, College of Medicine, Zhejiang University, Hangzhou 310003, PR China
[e] Hainan Institute of Zhejiang University, Sanya 572025, PR China

ARTICLE INFO

Keywords:
Microfluidics
Multiplexed detection
Foodborne pathogens
Food safety

ABSTRACT

The global burden of foodborne diseases is substantial and foodborne pathogens are the major cause for human illnesses. In order to prevent the spread of foodborne pathogens, detection methods are constantly being updated towards rapid, portable, inexpensive, and multiplexed on-site detection. Due to the nature of the small size and low volume, microfluidics has been applied to rapid, time-saving, sensitive, and portable devices to meet the requirements of on-site detection. Simultaneous detection of multiple pathogens is another key parameter to ensure food safety. Multiplexed detection technology, including microfluidic chip design, offers a new opportunity to achieve this goal. In this review, we introduced several sample preparation and corresponding detection methods on microfluidic devices for multiplexed detection of foodborne pathogens. In the sample preparation section, methods of cell capture and enrichment, as well as nucleic acid sample preparation, were described in detail, and in the section of detection methods, amplification, immunoassay, surface plasmon resonance and impedance spectroscopy were exhaustively illustrated. The limitations and advantages of all available experimental options were also summarized and discussed in order to form a comprehensive understanding of cutting-edge technologies and provide a comparative assessment for future investigation and in-field application.

1. Introduction

Unsafe or contaminated food causes about 600 million cases of foodborne illnesses and 420,000 deaths, including a total of 30% foodborne deaths among children under 5 years of age each year around the world (Hald et al., 2016; Havelaar et al., 2010). Foodborne illnesses are strongly associated with poverty in low- and middle-income countries, and it has become a growing public health issue. Increasing international trade and diverse food supply chains increase the risk of food contamination, particularly for food products across national and continental borders. The action of monitoring food to ensure its safety, including fast, on-site, and equipment-free detection is in high demand, particularly for those intensified trading borders, and countries with limited resources.

Foodborne illnesses are usually infectious or toxic in nature and caused by bacteria, including *Salmonella*, *Listeria*, *Vibrio cholerae*, Pathogenic *Escherichia coli*, etc., viruses, including Norovirus and Hepatitis A virus, entering the body through contaminated food or water. Since foodborne pathogens pose a great threat to public health, the detection of these pathogens is of great significance. Traditional foodborne pathogen detection methods such as culture plating which is the gold standard method, polymerase chain reaction (PCR), and enzyme-linked immune-sorbent assay (ELISA), etc. All the methods mentioned above rely on laboratory operations, which have a strong dependence on equipment and manual operations with cumbersome protocols, lengthy reaction time, the waste of reagents, and generation of poisonous wastes. Besides, such methods could not facilitate the need for on-site detection. Based on the practical demand, the rapid, cheap, portable, sensitive and multiple detection techniques become a research focus in recent years.

Microfluidics referring to the precise control and manipulation of fluids that are geometrically constrained to a small scale in recent years.

* Corresponding authors.
E-mail addresses: myue@zju.edu.cn (M. Yue), muying@zju.edu.cn (Y. Mu).
[1] These authors contributed equally to this study.

https://doi.org/10.1016/j.foodres.2021.110246
Received 20 November 2020; Received in revised form 2 February 2021; Accepted 16 February 2021
Available online 26 February 2021

It has been well developed as a powerful technology applied in the analysis of cell behavior, molecular biology, point of care diagnosis, plant tissue culture (Hong, Chen, Anderson, & Quake, 2006; Ko, Ju, Lee, & Cha, 2006; Yetisen, Akram, & Lowe, 2013; Yu, Meyvantsson, Shkel, & Beebe, 2005). Several assays related to microbe detection have been performed with microfluidic devices, including cell capture and enrichment, nucleic acid purification and amplification, ELISA, single-cell analysis, cell culture as well as droplet generation (Zhao, Li, & Liu, 2019). The combination of these assays can help to achieve "sample-in-answer-out" test in real sense. Based on the features of low volume and small size, microfluidic chips have the potential to achieve sensitive, multiplexed, automated, and rapid detection. The main process of pathogen detection with the technique is demonstrated in Fig. 1.

In process of microbiological surveillance sampling, a large number of samples of targeted food should be collected and tested over a relatively short period. Food serves as a transmission medium for multiple foodborne pathogens (e.g. milk acting as a transmission medium for *Cronobacter sakazakii, Listeria, Salmonella,* and *Escherichia coli*) (El-Sharoud, Darwish, & Batt, 2013; Van Kessel, Karns, Lombard, & Kopral, 2011) which results in the need for detection of multiple pathogens in the evaluation of food safety. Since the food can be sold only after all the indicators are qualified, multiplexed detection has been a new research focus due to the practical demands. The methods of multiplexed detection are established to achieve testing of multiple samples at the same time in the same device, leading to the prevention of sample waste, the reduction in equipment costs, the simplification of operation procedure as well as the shorten of testing time.

In this review, we introduced several multiplexed detection methods for foodborne pathogens based on microfluidic chip technology, including sample preparation methods and detection methods. Several cell lysis methods and nucleic acid extraction methods were introduced in the sample preparation chapter, and detection methods basing on biology and chemistry principles were also described in detail. We hope these methods could offer inspirations for future scientific research and industrial application.

2. Microfluidic devices

Microfluidics relying to the manipulation of microfluids to achieve their functions. Microfluidic devices allow for a flexible combination of multiple operating units and overall controllability, so some steps including sample preparation and detection can be integrated into a single chip. Since the chip is micron-scale or even nanoscale, it has a high specific surface area, a high diffusion coefficient, and fast heat transfer (Zhao, Li, & Liu, 2019). Thus, microfluidics possesses the advantages of reducing laboratory time, avoiding cross-contamination, and cutting down reagent and equipment costs. The integration with microfluidics could confer numerous analytical advantages to existing detection techniques. For instance, by recruiting microfluidics, electrochemistry biosensors obtain improvements in throughput, portability and rapidity (Rackus, Shamsi, & Wheeler, 2015).

For detection of pathogens, several types of microfluidic devices have been developed. Microchannel and chamber structures are the basic form of microfluidic chips. Usually, the chip is divided into different areas for various functions through the flow path design. With the help of centrifugal force, capillary force, or atmospheric pressure, regents and samples were mixed, wastes were washed, signals were detected in sequence to realize different procedures of detection in one chip (Koczula & Gallotta, 2016; Shin, Kim, Kim, & Choi, 2019; Strohmeier et al., 2015). The shape, size and materials could be designed and selected to fit for almost all kinds of methodologies, including PCR, ELISA, SPR, etc. (Coarsey, Coleman, Kabir, Sher, & Asghar, 2019; Strohmeier et al., 2014). The generation of droplets is also universally applied in analysis with microfluidics. Samples and regents were premixed and forms droplets with the help of immiscible fluids (Teh, Lin, Hung, & Lee, 2008). The size, shape and monodispersity can be manipulated to fit for various analysis requirements. The application of droplets could offer functions like high-throughput detection and digital

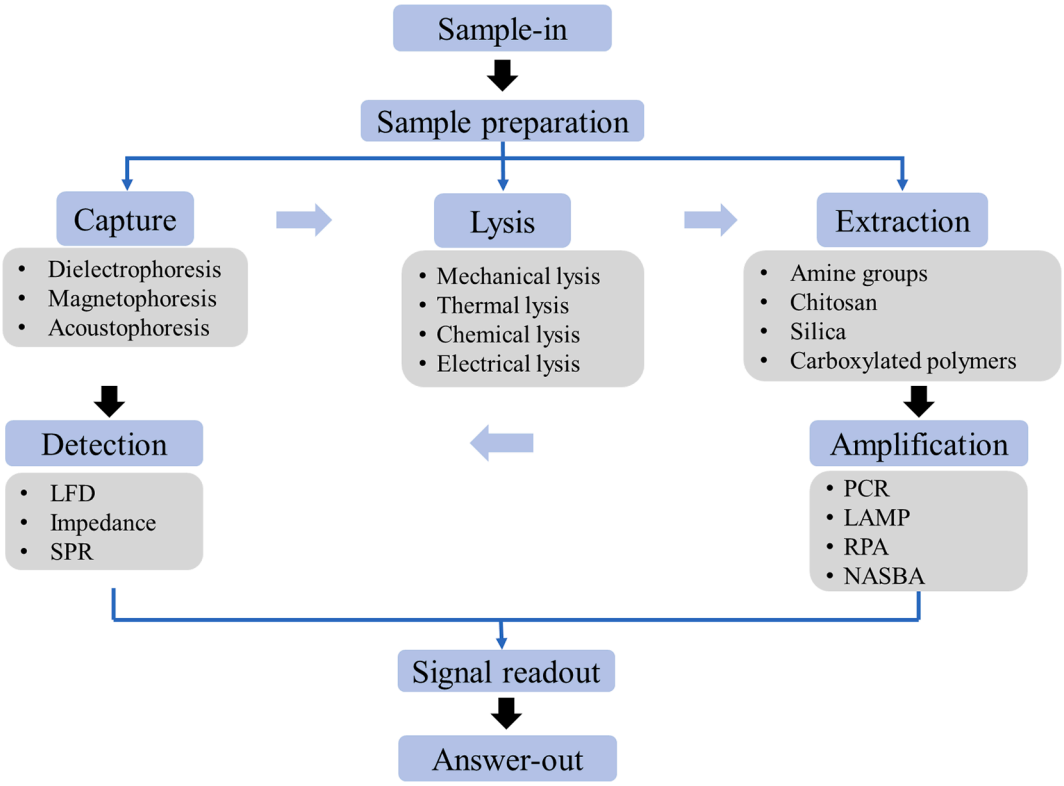

Fig. 1. An illustration for "sample in to answer out" detection of foodborne pathogens. SPR: Surface plasmon resonance; PCR: Polymerase chain reaction. LAMP: Loop-mediated isothermal amplification; RPA: Recombinase polymerase amplification; NASBA: Nucleic acid sequence-based amplification.

detection (Ahmadi, Samlali, Vo, & Shih, 2019; Kaushik, Hsieh, & Wang, 2018). Microarray structure is also common in microfluidic devices. Such two-dimension array on solid substrate possesses advantages of high-throughput, multiplexed and parallel processing and detection (Thissen et al., 2019).

Materials have an important impact on the realization of functions, costs of chip fabrication and transportation, and applications. Glass and silicon are a widely-used materials, which possesses properties like resistance to organic solvents, ease of metal deposition, high thermo conductivity, and stable electroosmotic mobility (Ren, Zhou, & Wu, 2013). Thus, they have been applied as the most commonly used materials, suitable for chemical synthesis and analysing, biomedical research and clinical diagnosis. (Gal-Or et al., 2019; Luitz et al., 2020; Muderrisoglu, Sargin, & Yesil-Celiktas, 2018; Regiart et al., 2017) However, high cost of fabrication and time-consuming labor limit their use, especially in point of care testing (POCT) (Hwang, Cho, Park, & Kim, 2019). Polymer-based materials including polydimethylsiloxane (PDMS), poly (methylmethacrylate) (PMMA), PC, polystyrene (PS), polyethylene terephthalate (PET), and polyvinylchloride (PVC) are also popular in recent years owing to their price advantage and better physical properties (McDonald & Whitesides, 2002). Take PDMS for example, high elasticity, low cost of microfabrication and most importantly, its air permeability makes cell culture and negative pressure injection in microchambers feasible (Ren et al., 2013). In order to achieve "in vitro" detection especially applying to single cell or molecule, hydrogels were also employed due to its cross-linked network structures and solidification at low temperature which enables target immobilized and observed continually and three-dimensionally (McDonald & Whitesides, 2002). It plays very important roles in drug delivery systems owing to its tunable properties makes better improvement in creating certain concentration gradient that needed (Qiu & Park, 2001). Paper-based analytical devices are recent focus owing to ease of fabrication and operation, low cost, equipment independence. The key characteristics including passive liquid transport and better biocompatibility and portability (Ren et al., 2013). Combing with methods like ELISA, electrochemistry detection, and Loop-mediated isothermal amplification (LAMP), they have a great potential of satisfying the needs of detection of various biomarkers and analytes especially in resource-limited settings (Dungchai, Chailapakul, & Henry, 2009; Hongwarittorrn, Chaichanawongsaroj, & Laiwattanapaisal, 2017; San Park, Li, McCracken, & Yoon, 2013; Verma et al., 2018).

According to the requirements based on detection accuracy, the microfluidic detection technology mainly includes: qualitative detection which determines whether there are target pathogens, quantitative detection which provides the concentration of pathogens, and digital detection that achieves detection of pathogens in low concentration and realization of absolute quantification (Li et al., 2020). Besides, requirements like multiplexed and high-throughput detection, on-site detection, real-time detection, continuous detection, etc. are also common in practical application. By selection and development of methodologies, materials and design of microfluidic chips, these requirements are gradually being fulfilled, but the technique still has much to improve.

3. Sample preparation

Sample preparation steps involving analyte purification and removal of inhibitors are of high significance in achieving high sensitivity and specificity. On-chip sample preparation including sorting, separation and patterning, characterization, purification of cells, viruses, nanoparticles, microparticles, and proteins is essential for subsequent detection procedure. According to the detection method, the preparation steps could be divided into two components: the enrichment of microbes and nucleic acid purification. For detections using complete microbes as analytes, only the capture of the target and the removal of irreverent impurities are required, while for detections based on nucleic acid analysis, nucleic acid purification is also an essential process.

3.1. The capture and enrichment of microbes

In view of the complicated composition of food matrixes and low concentration of pathogens, cell enrichment and removal of impurities is of great significance. Almost all detection methods required enrichment and capture of the target analyte, including ELISA that binding to ligands of pathogens with antibodies, impedance spectroscopy based on enrichment of pathogens on the surface of the medium, and PCR that requires nucleic acid extracted from target microbes. Among the methods of on-chip sample preparation, Dielectrophoresis (DEP)-based microbe enrichment, magnetophoresis and acoustophoresis have good compatibility with several detection methods to form integrated equipment. The methods are summarized in Table 1.

3.1.1. Dielectrophoresis

Dielectrophoresis (DEP) is an electrokinetic phenomenon referring to the motion of neutral but polarizable particles (Houssin & Senez, 2014). When subjected to an inhomogeneous electric field, the particle with a larger dielectric constant than that of medium moves towards the greatest electric field strength, while the one with a smaller dielectric constant than that of medium moves in the opposite direction (Hakoda & Shiragami, 2000). Since biological organisms have diverse dielectric properties, which represents their structural, morphological, and chemical characteristics, DEP possesses the ability to separate and sort different types of cells, virus, bacteria, subcellular components such as DNA and protein (Asbury, Diercks, & van den Engh, 2002; Green, Morgan, & Milner, 1997; Lapizco-Encinas, Simmons, Cummings, & Fintschenko, 2004; Moon et al., 2011; Nakano & Ros, 2013). The application of DEP could promote the enrichment of biomolecules and therefore improve the sensitivity of detection and speed up the capturing process (Yang, 2009). Besides, DEP has also been reported to possess the ability of cell lysis based on the principle of electroporation (Ramadan et al., 2006).

For application in the enrichment of foodborne pathogens, Cai *et al* applied positive DEP (pDEP) on a microfluidic device for direct enrichment of bacterial cells from milk (Cai et al., 2018). The device recruited a long winding channel for desalination of samples, and interdigitated microelectrodes with small spaces for enrichment of the bacterial cells (Fig. 2A). The capture efficiency of bacteria in the milk could achieve 90.0%. Antibodies or aptamers could be modified on the microfluidic devices for enrichment of particular bacteria. Shangguan *et al* employed pDEP coupled with a specific aptamer against *Staphylococcus aureus* on a microfluidic chip (Shangguan et al., 2015). The aptamers were immobilized onto functionalized fluorescent silica nanoparticles for capturing and labeling the bacteria, and then the complex moved to the electrodes and accumulated in the electrode gaps with pDEP.

DEP as a label-free technique with simple instrumentation, possesses the ability to achieve rapid and efficient enrichment of particles of a range of sizes (Cetin & Li, 2011). DEP combining with electrical lysis or with detection methods like impedance or Surface plasmon resonance (SPR) detection has been well developed (Galvan, Parekh, Liu, Liu, & Yu, 2018; Li & Anand, 2019; Nguyen & Jen, 2018). Thus, these reactions could be performed in the same chamber in order to save material costs, the size of the device as well as sample loss. However, DEP is restricted in use when the viability of the targeting microbes is required for the subsequential process, for that the voltage applied to generate DEP may induce serious Joule heating effect inside the channel. Besides, Joule heating effect could produce gas bubbles thus interfere the detection process. Non-biological particles may also accumulate at the electrodes leading to lower collection efficiency and shorter service life of the device.

Table 1
A list of cell capture and enrichment methods. IDEs: interdigitated electrodes; DI water: deionized water; PBS: phosphate-buffered saline.

Technology	Method	Target Pathogen	Capture Efficiency	Citation
Dielectrophoresis	Dielectrophoresis	*Escherichia coli*, *Staphylococcus aureus*, *Pseudomonas aeruginosa*	The recovery yield was 91.3%	(Abdullah et al., 2019)
	Electrokinetic velocity + Dielectrophoretic trapping assessments + Finite element multi-physics modeling	*Listeria monocytogenes*	Three serovars of *Listeria monocytogenes* (1/2a, 1/2b, and 4b) could be distinguished	(Crowther, Hilton, Kemp, & Hayes, 2019)
	Generate planar electrode patterns with enhanced volumetric electric fields	*Escherichia coli*	1.4 to 35.8 times more bacteria were captured than the IDEs ($p < 0.0016$)	(Han, Ha, & Jang, 2019)
	H-filter desalination + Dielectrophoresis	*Escherichia coli*	70.7% in 1 × PBS buffer, 90.0% in cow's milk and 80.2% in whole human blood	(Cai et al., 2018)
	Membraneless microfluidic dialysis + Dielectrophoresis	*Escherichia coli*, *Staphylococcus aureus*	79 ± 3% of *Escherichia coli* and 78 ± 2% of *Staphylococcus aureus* spiked into whole blood could be isolated at a processing rate of 0.6 mL/h. Collection efficiency was independent of the number of target bacteria up to 10^5 cells	(D'Amico, Ajami, Adachi, Gascoyne, & Petrosino, 2017)
	Dielectrophoresis	*Escherichia coli*	up to 97% capture rate could be obtained	(Bisceglia et al., 2015)
Magnetophoresis	Commercial polyethylene tube + magnet	*Staphylococcus aureus*, *Salmonella* Typhimurium, *Listeria monocytogenes*	higher than 92% and 110 fold in DI water, higher than 96% and 110 fold in food sample.	(Jung et al., 2020)
	Slanted ridge-arrays + Magnetic particles	*Escherichia coli*	91.68% ± 2.18% of *Escherichia coli* could be successfully separated from undiluted whole blood at a flow rate of 0.6 mL/h	(Jung et al., 2018)
	Antibody + Magnetic beads + Corresponding pathogen-specific biotinylated recombinant phages	*Salmonella* Typhimurium, *Escherichia coli* O157	72% of *Salmonella* Typhimurium-bound DNA beads and 67% of *Escherichia coli* O157-bound beads could be sorted from a 100 mL mixture within 1.2 min	(Ngamsom, Esfahani, & et al., 2016)
	The track-etched magnetic micropore (TEMPO) filter + Dielectrophoresis	*Escherichia coli*	enrichment of zeta > 500 could be achieved at a flow rate of Phi = 5 mL/h	(Muluneh et al., 2014)
Acoustophoresis	Ultrasound actuation	*Salmonella* Typhimurium	Pathogen recovery was 60%-90% in spiked samples of chicken and minced beef	(Ngamsom, Lopez-Martinez, & et al., 2016)
	Surface acoustic wave (SAW)	*Escherichia coli* DH5 alpha, *Escherichia coli* NIH/3T3	The recovery efficiencies of 81.0 ± 17.2% and 90.8 ± 5.0% were obtained, respectively.	(Mu, Zhang, Lin, Dai, & Cao, 2015)
	Thin-reflector multi-layered resonator + Acoustic radiation forces	*Escherichia coli* K12 *Staphylococcus epidermidis* ATCC 12228	A significant increase in bacterial concentration has been achieved, up to a maximum of ~60-fold.	(Carugo et al., 2014)
	GN6 aptamer + Aptamer affinity bead + Acoustophoresis	*Escherichia coli* DH5α, *Enterobacter cloacae*, *Sphingomonas insulae*, *Escherichia coli* KCTC 2571, *Pseudomonas pictorum*	pathogen recovery (up to 98%)	(Lee et al., 2019)

3.1.2. Magnetophoresis

Micro-sized iron particles (MPs) have been widely used in sample preparation for cell capture in microfluidic devices. The targeting microbes were covalently binding to the modified MPs surface and separated from samples by controlling the motion of MPs under controlled flow and magnetic field. The modification of beads could be either chemical compounds such as Concanavalin A and 4-polyamidoamine dendrimers for the capture of a variety of microbes, or specific antibodies or aptamers for immobilized binding with particular pathogens (Hao et al., 2019; Kwon, Gwak, Hyun, Kwak, & Jung, 2019). A magnetophoresis-based device using a commercial polyethylene tube wrapping around a permanent magnet was developed (Jung et al., 2020). Pathogens like *Staphylococcus aureus, Salmonella* Typhimurium, *and Listeria monocytogenes* could be labeled with various biologically active groups that are conjugated with magnetic particles and separated from the food matrix in the external magnetic field. The separation efficiency and concentration factor are higher than 92% and 110 times, respectively (Fig. 2B). Hao *et al* applied magnetic nanoparticles (MNPs) modified with monoclonal antibodies on a microfluidic chip for detection of *Salmonella* Typhimurium (Hao et al., 2020). The chip was designed into three parts, a mixing channel which is convergence-divergence shaped for mixing of MNPs and detection nanoparticles, an incubating channel for interaction of antibody and *Salmonella* Typhimurium, and a separation chamber for capturing of the target complexes with the external magnetic field.

Magnetophoresis manipulates biomolecules with magnetic field, thus the analytes could pass through chambers and channels in a flexible manner, making it adaptable to various device structures and having wide applications. While the method requires modifications for cell capture, high costs and complicated pretreatment process, which may

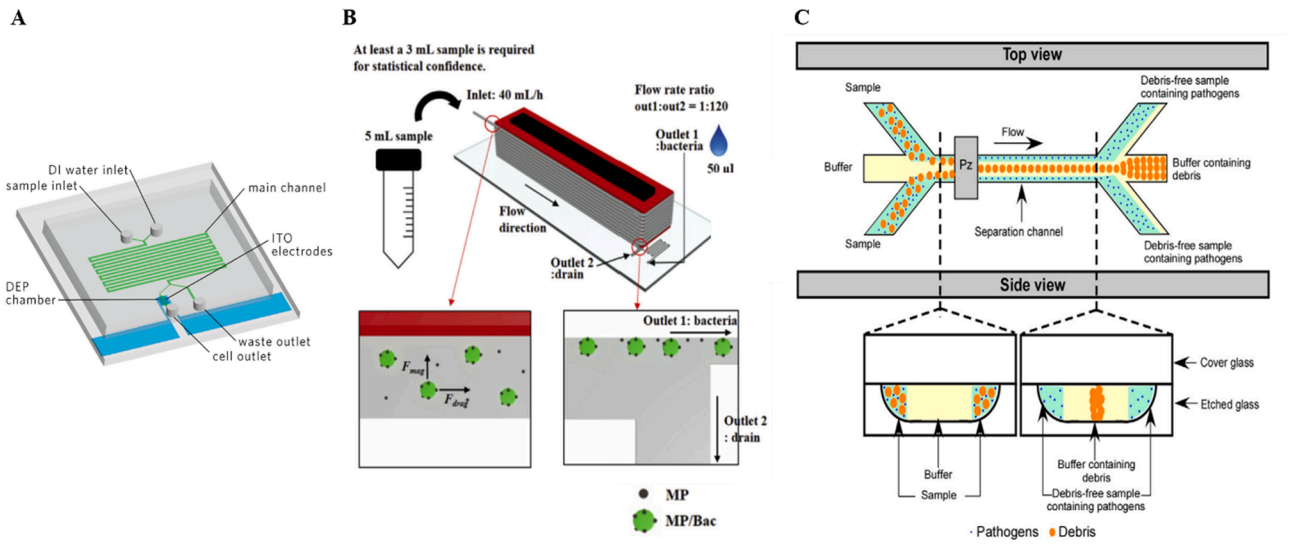

Fig. 2. Methods of cell capture and enrichment. (A) Illustration of the microfluidic chip integrated H-filter desalination and pDEP capture. Adapted with permission from AIP publishing (Cai et al., 2018). (B) Schematic flow of the continuous bacteria enrichment with the help of magnetic particles (MP) (left) and the detailed illustration of the microfluidic device (right). Adapted with permission from Elsevier (Jung, Jung, Ahn, & Yang, 2020). (C) Schematic flow of the on-chip acoustophoretic isolation of the pathogens from food debris under ultrasonic radiation force. The debris flows in the buffer stream in the central outlet, whilst the pathogens stay in the sample stream in the side outlets. Adapted with permission from Elsevier (Ngamsom et al., 2016).

restrict its application in some circumstances.

3.1.3. Acoustophoresis

Acoustophoresis is a non-contact and label-free method of manipulating particles as well as cells. Particles in an acoustic standing wave field can be affected by an acoustic radiation force, leading to the movement of particles if their acoustic properties differ from the surrounding medium. The size of the particle, the acoustic energy density, and the frequency of the sound wave are three main factors of the magnitude of the movement, and the direction of the particle movement relies on the density and speed of sound (Lenshof & Laurell, 2016). In the separation of foodborne pathogens, A acoustophoresis-based microfluidic protocol using aptamer-modified microbeads was developed to specifically capture gram-negative bacteria (Lee et al., 2019). The GN6 aptamer was chosen for its high specific binding affinity to various species of gram-negative bacteria therefore the aptamer-microbeads-bacteria complex could be successfully separated by size difference by means of acoustophoresis. 5 Gram-negative bacteria and 5 Gram-positive bacteria were taken as the targets and the control respectively, and it shows excellent separation performance, with high recovery (up to 98%), high purity (up to 99%), and a high-volume rate (500 μL/min). Ngamsom *et al* applied on-chip acoustophoresis as a pre-analytical technique for the detection of microbes from food and blood samples (Ngamsom et al., 2016). The microfluidic chip was constructed with three inlets for sample inputs and buffer solution, a central separation channel and three outlets. Upon ultrasound actuation, large debris particles (10–100 μm) from meat samples were continuously partitioned into the central buffer channel, while the pathogenic cells were collected over a 30 min operation cycle before further analysis (Fig. 2C). The system was successfully tested with *Salmonella* Typhimurium-spiked samples (ca. 10^3 CFU/mL) of chicken and minced beef with a high level of the pathogen recovery.

3.2. Nucleic acid sample preparation

For identification of a specific category of foodborne microbes, nucleic acid analysis possesses a greater sensitivity and specificity due to the amplification targeting the DNA barcode or RNA of a particular species. When using nucleic acid amplification methods for detection, cell lysis, nucleic acid extraction, and purification are essential in process of sample preparation in addition to cell enrichment. Since conventional off-chip nucleic acid extraction is highly labor-intensive, time-consuming, and requires a large amount of sample and reagents, on-chip nucleic acid extraction is developed to simplify the procedure and integrated with detection chip to form an enclosed system performing "sample-in to answer-out" analysis. The basic requirements of the preparation contain high quality and efficiency of nucleic acid extraction, elimination of inhibitors for subsequent reactions and high reaction rates at the microscale. Nucleic acid sample preparation on the microfluidic chip mainly includes cell lysis and nucleic acid extraction.

3.2.1. Cell lysis

The methods of on-chip cell lysis mainly include mechanical lysis, thermal lysis, chemical lysis, and electrical lysis (Kim, Johnson, Hill, & Gale, 2009).

Mechanical lysis refers to methods using a mechanical force to tear or puncture the cell membrane to achieve cell lysis. Mechanical vibration is a common method for cell disruption. Yan *et al* applied magnetic beads for bead-beating under a pair of rotating magnets to achieve cell lysis on a microfluidic chip (Yan et al., 2017). After that, the lysate was transferred to the clarification chamber via centrifugation. Stretching with nanowires has been also applied for cell disruptions (Yasui et al., 2019). This method could be successfully applied to Gram-positive bacteria with thick peptidoglycan layers and Gram-negative bacteria possessing strong resistivity to chemical damage due to their outer membrane. Compared with other methods, the mechanical method is not easy to be applied to a portable device due to its high dependence on assistant equipment and complex structure design, while it is suitable for lysis of almost all cell types, which offers a broader application.

Since proteins within the cell membranes are denatured leading to the release of the cytoplasmic contents in high temperature, thermal lysis is a reliable method for cell disruption. A multi-turn-serpentine-microchannel-platform was established with an attached resistive heater as a temperature controller (Packard, Wheeler, Alocilja, & Shusteff, 2013). The whole thermal lysis process can be easily demonstrated at temperatures greater than 65 °C and heating durations between 1 and 60 s without additional reagents. As the higher temperature is also required in the amplification procedure (e.g. LAMP), sample preparation and LAMP were combined on the paper-based microfluidic chip for detection of *Escherichia coli* and *Mycobacterium smegmatis* (Naik,

Jaitpal, Shetty, & Paul, 2019). Similarly, Lee *et al* applied thermal lysis at 95 °C for lysis of viral sample as well as the first step of RT-PCR (Lee, Lien, Lee, & Lei, 2008). Thermal lysis is convenient in combining detection methods thus simplifies detection procedure, while the high temperature could also lead to the degradation of RNA, which limits the use of this method.

Chemical methods basically depend on the presence of lysing agents, like organic solvents, detergents, or enzymes that help to degrade the surrounding layers of a cell. This process was originally described in 1979 saying with the help of sodium dodecyl sulfate (SDS) and NaOH can the cell wall weaken and lysed completely to release the intracellular contents (Birnboim & Doly, 1979). Other lytic agents like Triton X, chaotropic salts as well as several lysozymes and proteinase are also employed to break down the cell wall or membrane (Kim et al., 2009). Among these, SDS, an ionic detergent, can quickly denature proteins to achieve cell lysis. It is also used in sample preparations for nucleic acid extraction, as it denatures DNase and RNase enzymes (Pang, Al-Mahrouki, Berezovski, & Krylov, 2006). Cell lysis of *Escherichia coli* and *Salmonella* Typhimurium with SDS has been successfully performed on a microfluidic device (Geng, Bao, Sriranganathanw, Li, & Lu, 2012; Heo, Thomas, Seong, & Crooks, 2003). The use of a high concentration of chaotropic salts like guanidinium thiocyanate and guanidinium chloride is another common method in nucleic acid preparations. These chaotropic salts lyse cell membranes by disrupting protein intermolecular forces (Mason, Dempsey, Neilson, & Brady, 2005). Besides, they possess the ability of inactivation of RNases as well as deoxyribonucleases, which enzymatically digest DNA, and thus are valuable in nucleic acid extraction (Kim et al., 2009; Shaw et al., 2009). The high concentration of chaotropic salts also allows the salt bridge that forms to extract RNA with silica, which is widely used in microfluidic chips (Vandeventer, Mejia, Nadim, Johal, & Niemz, 2013). In an application for the detection of foodborne pathogens, the genome of *Escherichia coli* O157:H7 and *Listeria innocua* have been extracted successfully with chaotropic salts on microfluidic devices (Strohmeier et al., 2013; Zhang, Huang, Cai, Li, & Lin, 2018). Chemical lysis is less dependent on equipment compared to other methods, thus became the most popular method in cell lysis. However, the reagent costs can be considerable, and the chemistry applied for cell lysis needs to be modified for different cell types (Kim et al., 2009).

Electrical lysis is another reagent free method. Under a strong electric field, cell membranes are destabilized, permeable to macromolecules, and even dielectrically breakdown (Kim et al., 2009). An electrophoretic concentration and electrical lysis microfluidic device was employed for DNA extraction of *Escherichia coli* O157:H7*, Salmonella* and *Campylobacter* (Islam et al., 2017). The device contains two PDMS layers with two microchannels and a commercial nanoporous membrane in between. The microchannels with electrodes embedded in their reservoirs generate an electric field across the nanopores at the intersection, resulting in electrophoretically accumulation and lysis of the bacteria. The result shows an efficiency of 90% with a potential of 300 V for 3 min. Given that high voltage may damage the activity of cellular components and be dangerous to users, Wei *et al* established a new low-voltage cell lysis method (Wei, Li, Wang, & Yang, 2019). Two alternating current signals with a phase difference of 180°, voltage of 16 $V_{p\text{-}p}$, and a frequency of 10 kHz were applied to the two electrodes. Electrical lysis as a reagent-free, faster, and less expensive alternative to chemical treatment, has drawn comprehensive attention among researchers.

3.2.2. Nucleic acid extraction

The common extraction methods on the microfluidic chip mainly include modified beads and membranes. For general absorption of nucleic acid, solid-phase surface modified with chemical compounds like amine groups, chitosan, silica and carboxylated polymers possess the capability of capturing nucleic acid in a certain condition. In food sample preparation, silica modified microbeads were applied for the absorption of DNA of *Escherichia coli* O157:H7*, Salmonella* Typhimurium*, Vibrio parahaemolyticus* and *Listeria monocytogenes* in milk samples, according to the principle that nucleic acid bind with silica at a high chaotropic salt concentration (Oh et al., 2016). Sun *et al* conducted nucleic acid extraction with the help of carboxylated beads that could bind DNA on a high concentration of sodium chloride (Sun et al., 2014). As amine groups below neutral pH have a positive charge, inducing negatively charged DNA to bind, amino silane modified beads were applied for absorption of DNA of *Escherichia coli* O157:H7 (Jeong et al., 2019). Specific hybridization of targeting sequence is also a common form of nucleic acid extraction. A DNA probe conjugated with magnetic microbeads was employed for hybridization with target ribosomal RNA (rRNA) of *Salmonella* or *Listeria monocytogenes* bacteria (Weng, Jiang, & Li, 2012). When the analyte is RNA, reverse transcription may also be involved in the sample preparation process. Reverse transcription of rotavirus RNA was applied on a microfluidic device for further PCR process (Zhang, Li, & Wang, 2011). Noroviruses (NVs) and Rotaviruses (RVs) were reverse transcripted on a microfluidic chip for detection of with one-step RT-PCR (Li, Zhang, & Xing, 2010).

4. Biological detection methods

There are many detection methods for different types of foodborne microorganisms on microfluidic chips. At present, the detection process operates on the following principles: biological principle including amplification and immunoassay; Chemical methods including impedance spectroscopy and SPR. Table 2 lists various detection methods and their applications.

4.1. Amplification

16S rRNA and other specific nucleic acid sequences can be used for the detection and identification of a specific species (Huang et al., 2014). In this chapter, we introduced a variety of amplification methods for the detection of foodborne pathogens. As the concentration of microbes in the sample matrix is relatively low, amplification is essential in the nucleic acid-based detection method. The differences between samples could be magnified due to the amplification process, which is helpful for qualitative and quantitative analysis of target microbes in samples.

4.1.1. PCR

PCR technique is quite common in the use of identifying bacteria over the last 20 years. A spiral-channel microfluidic platform was developed for multiplexed detection of *Salmonella enterica, Listeria monocytogenes, Escherichia coli* O157:H7 and *Staphylococcus aureus* from banana, milk, and sausage (Shu et al., 2014). In this study, the PTFE capillary channel is coiled on 3 heating zones which are maintained at constant temperatures for denaturation, annealing and extension (Fig. 3A). The samples and PCR reagents were premixed forming a sample segment, and sequentially injected to the capillary as a regime. Several regimes could be injected in lines for multiplexed detection in different samples. Foodborne RNA virus also plays a significant role in inducing gastrointestinal diseases and food poisoning. Since these RNA viruses are hard to be cultivated or direct amplificated, reverse transcription-polymerase chain reaction was applied due to its sensitivity, rapidity and accuracy. A continuous-flow reverse transcription-PCR microfluidic chip was designed for the detection of two foodborne RNA viruses, Noroviruses (NVs) and Rotaviruses (RVs), at 6.4×10^4 copies/μL within 1 h (Li et al., 2010). The device is composed of two heated cylinders for reverse transcription and amplification reactions. A PTFE capillary is wound around these two cylinders in turn. To provide a more precise readout, digital PCR (dPCR) has also been applied to evaluate food safety. Bian *et al* applied droplet dPCR for detection of *Escherichia coli O157* and *Listeria monocytogenes* with a limit of detection (LOD) of 40 CFU/mL from milk in 18 h (Bian et al., 2015). Droplets were generated and trapped the individual bacterium in single droplet, and

Table 2
A list of various detection methods. LOD: limit of detection; LFD: lateral flow device; LAMP: loop-mediated isothermal amplification; AuNPs: gold nanoparticles; RPA: recombinase polymerase amplification; PCR: polymerase chain reaction.

Signal-readout	Amplification	Targeted Pathogen	Sample	LOD	Detection Time	Reference
Colorimetry	LAMP	*Salmonella*	Spiked Food samples	10 CFU/25 g	/	(Garrido-Maestu et al., 2017)
Fluorescence	LAMP	*Bacillus cereus, Escherichia coli, Salmonella enterica, Vibrio fluvialis, Vibrio parahaemolyticus*	/	7.2 copies/pi	60 min	(Xia et al., 2016)
	LAMP	*Escherichia coli, Proteus hauseri, Vibrio parahaemolyticus, Salmonella* subsp. *Enterica*	Serum samples	3 copies/μL	/	(Chen et al., 2017)
	LAMP	*Vibrio parahemolyticus*	/	1×10^3 CFU/mL	Less than 2 h	(Pang et al., 2017)
	PCR	*Enterobacter sakazakii*	Reconstituted skim milk (RSM)	10^2 CFU/mL	70 min	(El-Sharoud, Darwish, & Batt, 2013)
	PCR	*Escherichia coli O157:H7*	Spiked milk	12 CFU/mL	/	(Zhang, Huang, Cai, Li, & Lin, 2018)
Wavelength shift	RPA	*Salmonella*	/	50 CFU/10 mL	1 h	(Dao et al., 2018)
LFD	RPA	*Salmonella*	PBS/Milk	10 CFU/mL and 10^2 CFU/mL in PBS and milk, respectively	30 min	(Kim et al., 2014)
	LAMP	*Salmonella spp* , *Cronobacter spp* , *Staphylococcus aureus*	Powdered infant formula	4.2 CFU/g and 2.6 CFU/g and 3.4 CFU/g,	Less than 1 h	(Jiang et al., 2020)
	/	*Escherichia coli* O157:H7, *Salmonella* Typhimurium	Contaminated lettuces	1 CFU of pathogen/1 g of sample	7 h (with 6 h enrichment)	(Shin et al., 2018)
	/	*Escherichia coli O157:H7*		5×10^4 CFU/mL	22 min	(Shin & Park, 2016)
SPR	/	*Escherichia coli* O157:H7	/	1.87×10^3 CFU/mL	/	(Wang et al., 2016)
Impedance	/	*Escherichia coli*	Chicken sample	5×10^4 CFU/mL	6 min	(Liu et al., 2017)
	/	*Escherichia coli* O157:H7	/	12 CFU/ml	/	(Yao et al., 2018)
	/	*Salmonella*	Raw chicken products	10 cells/mL	less than 1 h	(Abdullah et al., 2019)
	/	*Listeria monocytogenes*	Milk	5.5 CFU/mL	/	(Chiriacò, Parlangeli, Sirsi, Poltronieri, & Primiceri, 2018)
	/	*Listeria monocytogenes*	/	1.6×10^2 CFU/mL	1 h	(Chen et al., 2016)
	/	*Pseudomonas aeruginosa, Streptococcus mutans*	/	10^5 CFU/mL	25 min	(Lillehoj, Kaplan, He, Shi, & Ho, 2014)
	/	*Escherichia coli* O157:H7	Synthetic chicken samples	5×10^4 CFU/mL	6 min	(Wang et al., 2017)

probes with fluorescent reporter dyes were used to label and discriminate these two bacteria.

Among these PCR techniques, digital PCR possesses advantages of absolute quantification and more tolerant to inhibitory substances, thus it has a wider prospect of use (Coudray-Meunier et al., 2015). Although it has not been fully applied in foodborne pathogen detection, dPCR was well developed in clinical application like detection of tumor, biomedical research, environmental monitoring of microbes, etc. (Gorgannezhad, Umer, Islam, Nguyen, & Shiddiky, 2018; Pinheiro et al., 2012; Tadmor, Ottesen, Leadbetter, & Phillips, 2011). PCR is very popular among various amplification techniques because of its high detection sensitivity, low reagent costs and wide range of applications. However, PCR also has some shortcomings. A thermal cycler is essential for amplification which promotes equipment costs and not suitable for portable device design, and lengthy cycling time making it quite time-consuming for the whole detection.

4.1.2. LAMP

LAMP is a well-developed isothermal nucleic acid amplification technique. In LAMP, the target sequence is amplified at a constant temperature of 60–65 °C with the help of either two or three sets of primers (Notomi, Mori, Tomita, & Kanda, 2015). Typically, 4 different primers are used to amplify 6 distinct regions on the target gene, which increases specificity. An additional pair of "loop primers" can further accelerate the reaction. In addition to a replication activity, a polymerase with high strand displacement activity is needed for amplification.

In the application of food safety, Jin *et al* designed a self-priming compartmentalization (SPC) microdevice with LAMP system in identification of *Salmonella* Typhimurium, *Bacillus cereus, Staphylococcus aureus, Yersinia enterocolitica, Vibrio parahaemolyticus and Listeria* monocytogenes within 1 h (Jin et al., 2020). The chip is composed of 24 independent microwells allowing multiplexed test simultaneously. During operation, primers are preloaded into microwells and dried, and then the excess air from channels and microwells is removed with a vacuum instrument. After that the sample mixed with reaction regents is loaded through inlet and flow to the microwells. Finally, the chip is put into a heater for reaction and the color change is observed with naked eyes. Pang *et al* developed a self-priming polydimethylsiloxane/paper hybrid microfluidic chip (SPH chip) capable of detection of *Staphylococcus aureus* and *Vibrio parahaemolyticus* with an LOD reached down to 10^3 CFU/mL (Pang et al., 2018). The SPH chip composed of four components, a PDMS top layer composed of an inlet and three microchannels, a PDMS reaction layers containing three reaction chambers, three chromatography paper which and a bottom glass coverslip for structural support. Before testing, the air in the empty chip was evacuated by vacuum machine. The LAMP regents were inserted with sample into the inlet of the SPH chip with the help of atmospheric pressure.

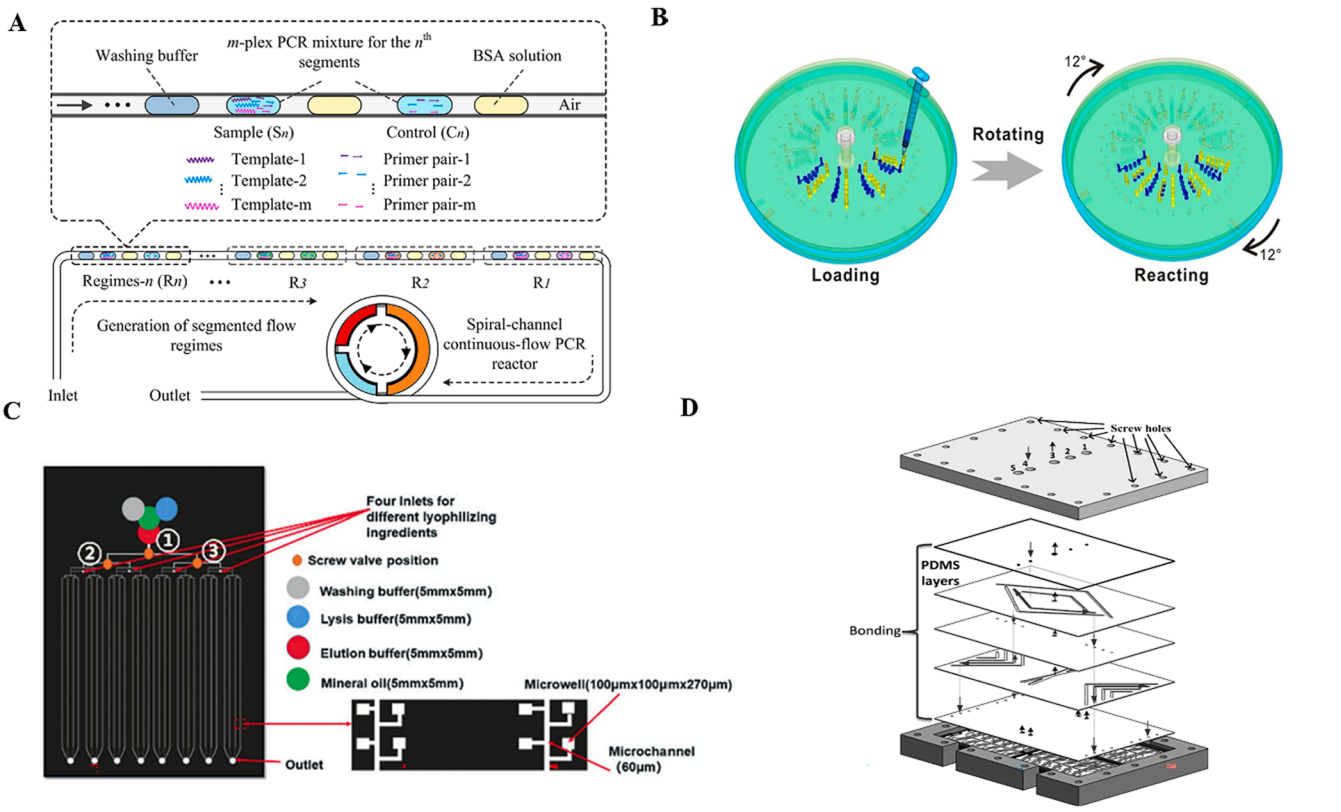

Fig. 3. Amplification methods for multiplexed pathogen detection. (A) Illustration of the design principle of spiral-channel segmented continuous-flow multiplex PCR. Adapted with permission from Elsevier (Shu, Zhang, & Xing, 2014). (B). Schematic illustration of the one-step rotational operation on the LAMP-based slipchip. Adapted with permission from Elsevier (Xia et al., 2016). (C) Illustration of the dRPA microfluidic chip for multiplexed detection. Adapted with permission from Royal Society of Chemistry (Yin et al., 2020) (D) Illustration of the 24-channel Q-NASBA chip. Adapted with permission from Royal Society of Chemistry (Zhao & Dong, 2012).

Finally, the chip was sealed for LAMP reaction and detected with Maestro in-vivo imaging system. Since primers were preloaded on the paper before fabrication of the chip, the three reaction chambers could work independently in order to achieve multiplexed detection. a sample-to-answer genetic analyzer was designed for multiplex detection of foodborne bacteria which integrating a 3D printed solution-loading cartridge and a centrifugal microfluidic disc (Van Nguyen, Nguyen, Lee, & Seo, 2019). The device contains two independent units available for multiplexed detection. The colorimetric data could be obtained within 1 h at an LOD of 10^2cells/mL when detecting *Escherichia coli O157:H7, Salmonella typhimurium, and Vibrio parahaemolyticus*.

For quantitative detection of food pathogens, a rotatory microfluidic disc was designed for quantitative detection of *Bacillus cereus, Escherichia coli, Salmonella enterica, Vibrio fluvialis* and *Vibrio parahaemolyticus* in 1 h (Xia et al., 2016). The disc was composed of 10 independent LAMP reaction units, and each reaction unit contained two individual sample injection paths as well as three reaction chambers (Fig. 3B). Dou *et al* integrated CD-like rotary microfluidic disc with aptamer-functionalized graphene oxide (GO) nano-biosensors for multiplexed detection of *Staphylococcus aureus and Salmonella enterica* of 6 copies and 12 copies respectively within 1 h (Dou, Sanjay, Dominguez, Zhan, & Li, 2017). The disc contains the porous paper inside the detection microzones and two PMMA plates outside. By rotation of the two plates, reagents are introduced from the inlet and then trapped and sealed between the two plates. When the LAMP reactions are completed, the chip is turned over and the LAMP products are distributed to the detection microzones to mix with probes for quantitative detection.

Compared with PCR, the amount of DNA produced in LAMP is considerably higher than PCR-based amplification, and lower LOD could be achieved with LAMP (Zhang et al., 2019). In Mass Besides, LAMP possesses ultrahigh specificity and sensitivity, and presents insensitivity to inhibitors, thus it is widely applied in clinical diagnosis, environmental monitoring, plant pathology and even genomics (Poon et al., 2005; Tao et al., 2011; Tomlinson, Boonham, & Dickinson, 2010; Xie, Yuan, Chai, & Yuan, 2015). Except for the mainstream detection methods like colorimetric indicators and fluorescent signals, electrochemical methods, immunochromatic technique, bioluminescence and SPR were also utilized with LAMP to fulfill requirements like real-time monitoring, label-free detection, etc. (Zhang et al., 2019). Besides, digital LAMP has also been developed to provide higher sensitivity (Zhu et al., 2012). As an isothermal amplification method, a thermal cycler is not needed for the reaction, making it possible for potable design. The demerit of the method mainly includes complicated primer design.

4.1.3. Recombinase polymerase amplification (RPA)

RPA is an isothermal alternative to PCR. In this method, The RPA process depends on three core enzymes – a recombinase capable of pairing primers with homologous sequence in duplex DNA, a single-stranded DNA-binding protein (SSB) which is responsible for binding to displaced strands of DNA and prevent the primers from being displaced, and strand-displacing polymerase starts DNA synthesis where the primer has bound to the target DNA (Euler et al., 2012; Piepenburg, Williams, Stemple, & Armes, 2006). the amplification happens at a constant temperature of 37–42 °C and still works at room temperature without thermal or chemical melting.

Yin *et al* developed a digital RPA chip for detection of *Escherichia coli* O157:H7, *Listeria monocytogenes* and *Salmonella enterica* within 45 min (Yin et al., 2020). The chip contains 4 detection areas with 12,800 chambers in total to achieve simultaneous multiplexed detection (Fig. 3C). the RPA reagents and primer mixture were introduced to

chambers with vacuum-based self-priming introduction method and freeze dried. When testing, the liquid sample is loaded and absorbed to reaction chambers for RPA reaction. The fluorescent signal could be observed with a CCD camera.

As an isothermal amplification method, RPA can react at a lower temperature and even room temperature, which can reduce energy and equipment costs. From the analytical sensitivity and specificity perspective, RPA is as sensitive as PCR, while due to the natural function of these enzymes for performing homology directed repair, it has difficulty in discrimination towards closely-related species (J. Li, Macdonald, & von Stetten, 2019). When testing field samples, RPA only present half as sensitive as the benchmark method, whereas the clinical specificity of RPA is most of time as specific as the benchmark method. The specificity and sensitivity of RPA needs to be improved in future research.

4.1.4. Nucleic acid sequence-based amplification (NASBA)

NASBA is a one-step isothermal amplification technique targeting viral and bacterial RNA. NASBA employs reverse transcriptase for transforming RNA to cDNA, RNase H for digestion of RNA in RNA–DNA hybrid, and T7 RNA polymerase for the synthesis of new RNA molecules (Sun, 2010). Since the choice of targeting RNA instead of DNA could reflect the viability of pathogens, NASBA has the potential to detect the infectivity of pathogens. This technique has been widely applied in clinical diagnosis for infections and the food industry to evaluate the microbiological safety of food products (Tillmann, Simon, Müller, & Schildgen, 2007; Wernecke & Mullen, 2014).

For on-chip detection of foodborne microbes, Zhao *et al* reported a NASBA microfluidic platform for quantitative detection of *Saccharomyces cerevisiae*, *Staphylococcus aureus* and *Escherichia coli* within 70 mins (Zhao & Dong, 2012). In this work, a NASBA chip unit was made of a silicon wafer, on which microchannels and gold-coated chambers were fabricated. Four of NASBA chip units were separately fixed onto the underplate. PDMS channels connected sample and control inlets and outlets were coated on the top. The whole device can simultaneously perform 148 NASBA assays or 24 quantification channels for different RNA targets, allowing multiplexed detection (Fig. 3D). The fluorescent signal of positive or negative results will be detected with common microplate readers.

As a method of RNA amplification, NASBA can detect and identify target microbes at the metabolic level without relying on reverse transcription, which is not available in other amplification methods. Therefore, NASBA has different amplifications from other amplification methods, for instance, the detection of infectious microbes.

4.2. Immunoassay

Immunoassay operates on the specific interaction of antibody-antigen or aptamer-antigen. In recent years, immunoassay method has become one of the most common foodborne microbe detection methods. ELISA as a powerful immunoassay method has been well developed and applied in the detection of foodborne pathogens. In recent years, ELISA has achieved more accurate and sensitive detection on microfluidic chips, but it is inferior in the design of multiplexed detection and dependent on equipment and professional operation (Zhao et al., 2016). Lateral flow assay is another immunoassay-based detection method established based on the similar principle of ELISA (Sajid, Kawde, & Daud, 2015). The assay usually performed on a paper-based flow device named as lateral flow devices (LFD), which has great progress on the application for qualitative and multiplexed detection. An LFD is generally composed of a sample pad containing a series of capillary beds allowing fluid to transport spontaneously, and two types of antibodies that bind specifically to the target analyte. When a liquid sample was loaded on the sample pad, target analytes in the sample flow into the conjugate area and are captured by the preloaded antibodies. The immunocomplexes continue flowing to the testing area to bind antibodies embedded in the pad to form a visual line. At least one control line should occur to prove the reliability of the test. The sample pad is generally made of nitrocellulose, and for detection, the antibodies could be labeled with latex, gold nanoparticles, fluorescent particles and magnetic particles (Borse & Srivastava, 2019; Lee, Mehta, & Erickson, 2016; Mansfield, 2005; Moyano et al., 2020; Shin et al., 2018).

In view of the rapidity and portability nature of LFD, simultaneous detection of multiple samples can be performed on different strips simultaneously. As a result, the most common way to achieve multiplexed detection is loading the samples on individual strips, while assembled device is still in development. Multiplexed detection methods have been applied in LFD to significantly improve the efficiency of the detection of foodborne pathogens. Song *et al* employed an LFD for the simultaneous detection of *Shigella boydii* and *Escherichia coli* O157:H7 at 4 CFU/mL from bread, milk and jelly samples (Song et al., 2016). Zhao *et al* established a 10-channel up-converting phosphor technology-based lateral flow (TC-UPT-LF) device that successfully identified 10 foodborne pathogens from 110 food samples, with a detection sensitivity of 10^4 CFU/mL or 10^5 CFU/mL for each pathogen (Zhao et al., 2016). In this study, a TC-UPT-LF disc is composed of 10 single-target UPT-LF strips allowing multiplexed detection of sample at the same time, and each strip contains a sample pad, a conjugate pad laid UCP-monoclonal antibody targeting specific bacteria, a nitrocellulose membrane where the corresponding antibodies and the goat anti-mouse IgG antibody are separately fixed, and an absorbent paper (Fig. 4A). Liquid sample loading on the sampling pad, a positive band would appear if there were target bacteria in the sample. Besides, LFD could be integrated with other methods to promote application scope. A multiplexed RPA chip integrated with LFD was designed for detection of *Salmonella Enteritidis* from milk samples within 30 mins (Kim, Park, Kim, & Cho, 2014). The round shaped chip employs 6 independent detection units, each of which contains a lysis chamber for sample preparation, an amplification chamber for RPA process, a heater, a metering chamber, a dilution chamber and a lateral flow strip for detection. After the amplification, the RPA product was diluted and further transformed into a lateral flow strip for the detection of positive/negative lane. Park *et al* also employed an integrated rotary microfluidic system combing LAMP and LFD detection (Park et al., 2017). Multiplex pathogens contaminated in water or milk could be detected using this device with a limit of detection of 50 CFU in 80 min.

Apart from LFD, Other immunoassays were also developed for multiplexed detection of foodborne pathogens. For instance, a microfluidic device based on the Carba NP test (CNPt) was designed which featured multiplex (Wasey, Yang, Sun, He, & Zhang, 2020). The microfluidic device contains 20 individually working areas and in total 1280 reaction chambers in which 16 bacterial concentration gradients could be automatically generated by gradual dilution when flowing series of chambers. 20 bacterial isolates could be detected simultaneously just by color observation.

Immunoassay methods are widely used in molecular diagnosis due to the high specificity and applicability. LFD, as one of the most common immunoassay detection methods, was widely used in diagnosis of human disease, bacteria and toxins due to its rapidity and convenience in use (Shin & Park, 2016; L. Wang et al., 2011; J. Zhang, Shen, Xiang, & Lu, 2016). Several applications combining with amplification methods like RPA and LAMP have also been developed (Kim et al., 2014; Park et al., 2017). The advantages of using LFD mainly include low material cost, small size, and the results can be observed with the naked eyes. While the disadvantages of LFD mainly include the difficulty of achieving quantitative detections, high cost of antibodies and low sensitivity. Other methods based on immunoassay is still under development in recent years.

5. Chemical detection methods

Apart from methods mentioned above, Chemical detection methods

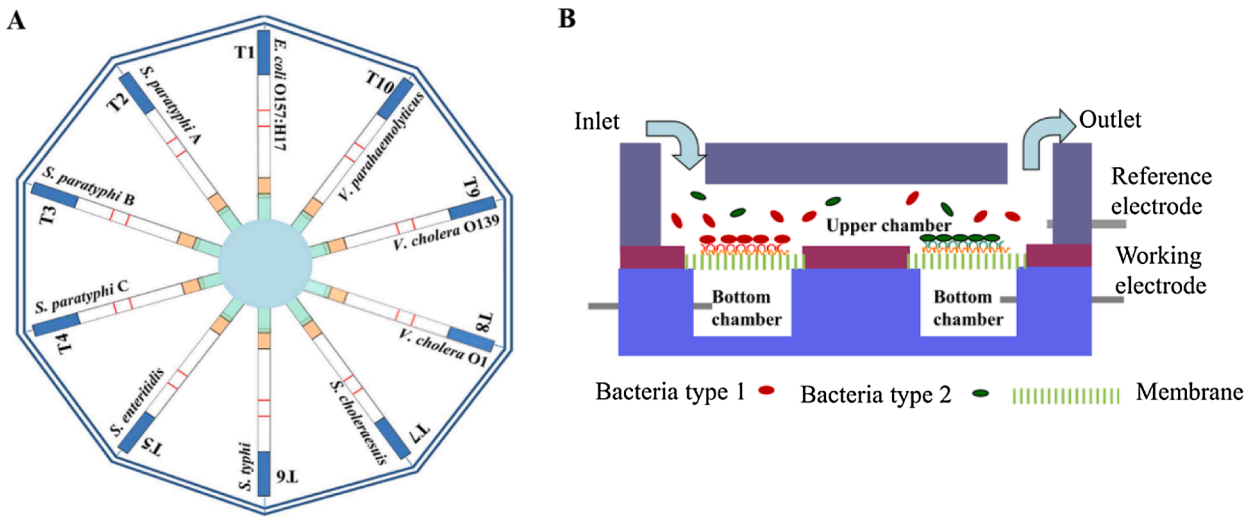

Fig. 4. Methods basing on immunoassay and impedance analysis. (A) Schematic illustration of the UPT-LF strip and TC-UPT-LF disc for detection of 10 foodborne pathogens. Adapted with permission from Nature Publishing Group (Zhao et al., 2016). (B) Schematic illustration of the simultaneous detection for two types of bacteria using the microfluidic device. Adapted with permission from Elsevier (Tian, Lyu, Shi, Tan, & Yang, 2016).

integrated with microfluidics are also common. These methods include SPR and impedance spectroscopy, which generate optical or electric signals by chemical changes caused by the binding with pathogens.

5.1. SPR

SPR becomes a popular optical biosensing technology due to its real-time, convenient and label-free nature (Tang, Zeng, & Liang, 2010). When the light is totally reflected on the surface of the metal film, it forms an evanescent wave. When a planar metal (typically gold or silver) is subject to electromagnetic interference, the electron density distribution of the metal would become uneven, inducing plasma oscillation on the surface. as the two waves resonate, which is called SPR, the detected reflected light intensity will be greatly reduced and forming a dark line (Amendola, Pilot, Frasconi, Maragò, & Iatì, 2017). The angle of the reflection required for the resonance is determined by the refractive index of the metal surface, thus can be used for detection of the adsorption–desorption or association-dissociation activities on the metal surface.

For detection of foodborne pathogens, a microfluidic platform integrated biosensor and a homemade microfluidic cell was developed for detection of *Escherichia coli* O157:H7 (Wang et al., 2016). A three-way solenoid valve was equipped and specific antibody was applied for capture of the bacteria. The theoretical detection limit was 1.87×10^3CFU/mL with a high sensitivity which was four orders of magnitude compare to ELISA kit. Tokel *et al* established a portable, multiplex, microfluidic-integrated SPR platform that quantifies *Escherichia coli* and *Staphylococcus aureus* with gold coated surfaces functionalized with specific antibodies within 20 min (Tokel et al., 2015).

In last two decades, SPR has been applied in clinical diagnosis, environmental monitoring, drug discovery, polymer engineering, etc. (Bhardwaj, Sumana, & Marquette, 2020; Puiu and Bala, 2016). Due to the nature of label-free capacity, SPR sensors present advantages over other conventional techniques. The binding of large biomolecules to the surface could be sensitively detected, as the plasmon resonance is extremely sensitive to surface dielectric properties. However, there are still problems like low sensitivity for small molecules and the fouling problem, and smart layers and various materials are being recruited to solve them in recent years (Qu, Dillen, Saeys, Lammertyn, & Spasic, 2020). SPR imaging (SPRi) is an advanced version of classical SPR, which combines the SPR of metallic surfaces with surface plasma-based optical techniques (Puiu and Bala, 2016). It enables real-time and multiplexed detection, and hopefully be recruited by foodborne pathogen detection.

5.2. Impedance spectroscopy

As cell membranes will block the current when cells are adherent to substrate electrodes, impedance measurements became a rising measurement of estimation of bacteria concentration (Xu et al., 2016). Most electrochemical and electrical sensors are label-free, which makes it easier and more promising to be widely used in the pathogen detection area. One of the most popular measurement techniques is impedance spectroscopy, which measures the impedance of a system over a range of frequencies (Rackus et al., 2015). A typical equivalent circuit model of the biosensors studied to analyze the impedance response is composed of two double layer capacitors (Cdl), a bulk medium resistor (Rs) in the middle that connects the two capacitors in series, and a dielectric capacitor (Cdi) connected in parallel with Rs and Cdl (Varshney, Li, Srinivasan, & Tung, 2007). In this model, Cdl represents the double layer capacitance of an electrode, Rs accounts for the electrical conductivity of the bulk medium and Cdi refers to dielectric capacitance of medium. The magnitude of impedance increases with the increase in the number of bacteria in the sample. When scanning from low frequency to high frequency, the dominant impedance signal is determined from Cdl, Rs to Cdi, and the change in impedance caused by the presence of bacteria is dominated by Rs as compared to Cdl and Cdi.

In terms of foodborne pathogen identification, Tian *et al* developed a microfluidic chip that combined impedance measurement and fluorescent detection of two types of bacteria, *Escherichia coli* O157:H7 and *Staphylococcus aureus* in range from 10^2CFU/mL to 10^5CFU/mL from mixed samples in 2 h (Tian et al., 2016). Two silanes functionalized nanoporous alumina membranes integrated with the PEG layer were placed on the microfluidic chip in parallel. The electrolytes were located on the upper chamber and bottom chamber of the device in order to generate vertical electrolyte current. Before detection, two specific antibodies were covalent bound to the membrane. When bacteria are captured by the corresponding antibodies, some nanopores are blocked and the electrolytes cannot pass through these blocked nanopores, leading to an increase of impedance. Quantitative data could be readout with an impedance analyzer. Impedance spectroscopy could also promote the efficiency of other methods. Sharif *et al* combined LAMP with an impedimetric sensor for sensing of various foodborne pathogens including *Escherichia coli* O157:H7, *Vibrio parahaemolyticus*,

Staphylococcus aureus, and *Listeria monocytogenes* at 10 copies (Sharif et al., 2019).

Except for application in foodborne pathogen detection, impedance spectroscopy was recruited in environmental monitoring, single-cell analysis, drug detection and clinical research (Cheng et al., 2020; Fan et al., 2019; McGrath et al., 2017; Petchakup, Li, & Hou, 2017). Impedance spectroscopy could offer a quantitative readout by measuring the magnitude of impedance in the frequency spectrum. Together with the rapid and sensitive detection features, it has a promising application in pathogen detection.

6. Conclusion and future perspectives

In this review, we introduced several sample preparation and detection methods, and various implementation methods for multiplex detection on microfluidic chips. Dividing the chip into several independent chambers for different reactions is the most common method to achieve multiplex detection. These chambers could share the same sample with the same sample preparation method (Jin et al., 2020; Pang et al., 2018; Yin et al., 2020; Zhao et al., 2016), or be totally independent, allowing for simultaneous detection of different samples (Kim et al., 2014; Shin et al., 2018; Song et al., 2016; Wasey et al., 2020; Xia et al., 2016; Zhao & Dong, 2012). Since all individual chambers share the same reaction conditions, this method has the ability of simultaneous detection of several samples. Marking the analytes with various labels and antibodies or aptamers is also a wide applied method in achieving multiplexed detection (Jiang et al., 2020; Shin et al., 2018; Song et al., 2016; Tian et al., 2016). Unfortunately, these designs are limited with the number of reaction chambers or antibodies, which may not facilitate the need for mass inspection. To solve this problem, continuous flow methods are applied for multiplexed detection (Li et al., 2010; Peham et al., 2011; Shu et al., 2014). In these methods, samples mixing with reaction reagents were driven towards the reaction device in turn, thus the corresponding results are read out and recorded sequentially. It is usually adapted by detection with PCR, wherein the realization of the temperature gradient relies on the design of separation of the round heating zones that the capillary containing regents and samples is wound around. Compared with other methods, the continuous flow method possesses the capability of executing a variety of detections with a large number of samples in a short time, and reduces material wastes. However, the scope of application of this method is limited mainly to the PCR method. Therefore, more methods to achieve multiplexed multiple detection need to be developed to meet the requirements of microbiological surveillance to ensure food safety.

Rapidity, accuracy, high sensitivity and industrialization have also been the research focus of food safety testing device in recent years. Integrated microfluidic device combining with sample preparation and detection is crucial for "sample-in to answer-out" to facilitate point of care diagnosis. The typical combinations include nucleic acid extraction with amplification method, the DEP for cell capture with impedance or SPR detection, and cell capture or amplification with LFD, etc. The integrated enclosed design of the chip could avoid complicated operating procedures, sample contamination, and save detection time. Besides, according to the practical demands, requirements such as more accurate detection (e.g. quantitative detection), lower costs of the device (e.g. paper-based materials) and automation should also be considered in design. In the end, the establishment of low-cost, high-efficient, high-sensitive, quantitative and automatic multiplexed detection methods will become the focus of future research.

CRediT authorship contribution statement

Xiaoying Han: Conceptualization, Investigation, Writing - original draft, Writing - review & editing. **Yuanhui Liu:** Conceptualization, Investigation, Writing - original draft, Visualization. **Juxin Yin:** Supervision, Writing - review & editing. **Min Yue:** Conceptualization, Supervision, Writing - review & editing. **Ying Mu:** Conceptualization, Supervision, Writing - review & editing, Project administration, Funding acquisition.

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgments

The authors are grateful for the financial support from the National Program on Key Research Project of China (2019YFE0103900, 2017YFC1600103) as well as European Union's Horizon 2020 Research and Innovation Programme under Grant Agreement No 861917 – SAFFI, and the Major Project of science and technology innovation 2025 in Ningbo (2018B10093).

References

Abdullah, A., Dastider, S. G., Jasim, I., Shen, Z., Yuksek, N., Zhang, S., … Almasri, M. (2019). Microfluidic based impedance biosensor for pathogens detection in food products. *Electrophoresis, 40*(4), 508–520. https://doi.org/10.1002/elps.201800405.

Ahmadi, F., Samlali, K., Vo, P. Q., & Shih, S. C. (2019). An integrated droplet-digital microfluidic system for on-demand droplet creation, mixing, incubation, and sorting. *Lab on a Chip, 19*(3), 524–535. https://doi.org/10.1039/C8LC01170B.

Amendola, V., Pilot, R., Frasconi, M., Maragò, O. M., & Iatì, M. A. (2017). Surface plasmon resonance in gold nanoparticles: A review. *Journal of Physics: Condensed Matter, 29*(20), 203002. https://doi.org/10.1088/1361-648x/aa60f3.

Asbury, C. L., Diercks, A. H., & van den Engh, G. (2002). Trapping of DNA by dielectrophoresis. *Electrophoresis, 23*(16), 2658–2666. https://doi.org/10.1002/1522-2683(200208)23:16<2658::aid-elps2658>3.0.co;2-o.

Bhardwaj, H., Sumana, G., & Marquette, C. A. (2020). A label-free ultrasensitive microfluidic surface Plasmon resonance biosensor for Aflatoxin B1 detection using nanoparticles integrated gold chip. *Food Chemistry, 307*, 125530. https://doi.org/10.1016/j.foodchem.2019.125530.

Bian, X., Jing, F., Li, G., Fan, X., Jia, C., Zhou, H., … Zhao, J. (2015). A microfluidic droplet digital PCR for simultaneous detection of pathogenic Escherichia coli O157 and Listeria monocytogenes. *Biosensors and Bioelectronics, 74*, 770–777. https://doi.org/10.1016/j.bios.2015.07.016.

Birnboim, H., & Doly, J. (1979). A rapid alkaline extraction procedure for screening recombinant plasmid DNA. *Nucleic Acid Research, 7*(6), 1513–1523. https://doi.org/10.1093/nar/7.6.1513.

Bisceglia, E., Cubizolles, M., Trainito, C. I., Berthier, J., Pudda, C., Français, O., … Le Pioufle, B. (2015). A generic and label free method based on dielectrophoresis for the continuous separation of microorganism from whole blood samples. *Sensors and Actuators B: Chemical, 212*, 335–343. https://doi.org/10.1016/j.snb.2015.02.024.

Borse, V., & Srivastava, R. (2019). Fluorescence lateral flow immunoassay based point-of-care nanodiagnostics for orthopedic implant-associated infection. *Sensors and Actuators B: Chemical, 280*, 24–33. https://doi.org/10.1016/j.snb.2018.10.034.

Cai, D., Yi, Q., Shen, C., Lan, Y., Urban, G., & Du, W. (2018). Direct enrichment of pathogens from physiological samples of high conductivity and viscosity using H-filter and positive dielectrophoresis. *Biomicrofluidics, 12*(1), 014109. https://doi.org/10.1063/1.5016413.

Carugo, D., Octon, T., Messaoudi, W., Fisher, A. L., Carboni, M., Harris, N. R., … Glynne-Jones, P. (2014). A thin-reflector microfluidic resonator for continuous-flow concentration of microorganisms: A new approach to water quality analysis using acoustofluidics. *Lab on a Chip, 14*(19), 3830–3842. https://doi.org/10.1039/C4LC00577E.

Cetin, B., & Li, D. (2011). Dielectrophoresis in microfluidics technology. *Electrophoresis, 32*(18), 2410–2427. https://doi.org/10.1002/elps.201100167.

Chen, C., Liu, P., Zhao, X., Du, W., Feng, X., & Liu, B.-F. (2017). A self-contained microfluidic in-gel loop-mediated isothermal amplification for multiplexed pathogen detection. *Sensors and Actuators B: Chemical, 239*, 1–8. https://doi.org/10.1016/j.snb.2016.07.164.

Chen, Q., Wang, D., Cai, G., Xiong, Y., Li, Y., Wang, M., … Lin, J. (2016). Fast and sensitive detection of foodborne pathogen using electrochemical impedance analysis, urease catalysis and microfluidics. *Biosensors and Bioelectronics, 86*, 770–776. https://doi.org/10.1016/j.bios.2016.07.071.

Cheng, Y. H., Barpaga, D., Soltis, J. A., Shutthanandan, V., Kargupta, R., Han, K. S., … Chatterjee, S. (2020). Metal-organic framework-based microfluidic impedance sensor platform for ultrasensitive detection of perfluorooctanesulfonate. *ACS Applied Materials & Interfaces, 12*(9), 10503–10514. https://doi.org/10.1021/acsami.9b22445.

Chiriacò, M. S., Parlangeli, I., Sirsi, F., Poltronieri, P., & Primiceri, E. (2018). Impedance sensing platform for detection of the food pathogen listeria monocytogenes. *Electronics, 7*(12), 347. https://doi.org/10.3390/electronics7120347.

Crowther, C. V., Hilton, S. H., Kemp, L., & Hayes, M. A. (2019). Isolation and identification of Listeria monocytogenes utilizing DC insulator-based

dielectrophoresis. *Analytica Chimica Acta, 1068*, 41–51. https://doi.org/10.1016/j.aca.2019.03.019.
Coarsey, C., Coleman, B., Kabir, M. A., Sher, M., & Asghar, W. (2019). Development of a flow-free magnetic actuation platform for an automated microfluidic ELISA. *RSC Advances, 9*(15), 8159–8168. https://doi.org/10.1039/c8ra07607c.
Coudray-Meunier, C., Fraisse, A., Martin-Latil, S., Guillier, L., Delannoy, S., Fach, P., & Perelle, S. (2015). A comparative study of digital RT-PCR and RT-qPCR for quantification of Hepatitis A virus and Norovirus in lettuce and water samples. *International Journal of Food Microbiology, 201*, 17–26. https://doi.org/10.1016/j.ijfoodmicro.2015.02.006.
D'Amico, L., Ajami, N., Adachi, J., Gascoyne, P., & Petrosino, J. (2017). Isolation and concentration of bacteria from blood using microfluidic membraneless dialysis and dielectrophoresis. *Lab on a Chip, 17*(7), 1340–1348. https://doi.org/10.1039/C6LC01277A.
Dao, T. N. T., Lee, E. Y., Koo, B., Jin, C. E., Lee, T. Y., & Shin, Y. (2018). A microfluidic enrichment platform with a recombinase polymerase amplification sensor for pathogen diagnosis. *Analytical Biochemistry, 544*, 87–92. https://doi.org/10.1016/j.ab.2017.12.030.
Dou, M., Sanjay, S. T., Dominguez, D. C., Zhan, S., & Li, X. (2017). A paper/polymer hybrid CD-like microfluidic SpinChip integrated with DNA-functionalized graphene oxide nanosensors for multiplex qLAMP detection. *Chemical Communications (Cambridge, England), 53*(79), 10886–10889. https://doi.org/10.1039/c7cc03246c.
Dungchai, W., Chailapakul, O., & Henry, C. S. (2009). Electrochemical detection for paper-based microfluidics. *Analytical Chemistry, 81*(14), 5821–5826. https://doi.org/10.1021/ac9007573.
El-Sharoud, W. M., Darwish, M. S., & Batt, C. A. (2013). A real-time PCR-based microfluidics platform for the detection of Cronobacter sakazakii in reconstituted milks. *International Dairy Journal, 33*(1), 67–74. https://doi.org/10.1016/j.idairyj.2013.06.010.
Euler, M., Wang, Y., Nentwich, O., Piepenburg, O., Hufert, F. T., & Weidmann, M. (2012). Recombinase polymerase amplification assay for rapid detection of Rift Valley fever virus. *Journal of Clinical Virology, 54*(4), 308–312. https://doi.org/10.1016/j.jcv.2012.05.006.
Fan, W., Chen, X., Ge, Y., Jin, Y., Jin, Q., & Zhao, J. (2019). Single-cell impedance analysis of osteogenic differentiation by droplet-based microfluidics. *Biosensors and Bioelectronics, 145*, 111730. https://doi.org/10.1016/j.bios.2019.111730.
Gal-Or, E., Gershoni, Y., Scotti, G., Nilsson, S. M., Saarinen, J., Jokinen, V., Strachan, C. J., af Gennäs, G. B., Yli-Kauhaluoma, J., & Kotiaho, T. (2019). Chemical analysis using 3D printed glass microfluidics. *Analytical Methods, 11*(13), 1802–1810. https://doi.org/10.1039/C8AY01934G.
Galvan, D. D., Parekh, V., Liu, E., Liu, E.-L., & Yu, Q. (2018). Sensitive bacterial detection via dielectrophoretic-enhanced mass transport using surface-plasmon-resonance biosensors. *Analytical Chemistry, 90*(24), 14635–14642. https://doi.org/10.1021/acs.analchem.8b05137.
Garrido-Maestu, A., Azinheiro, S., Carvalho, J., Abalde-Cela, S., Carbó-Argibay, E., Diéguez, L., Kolen'ko, Y. V., … Prado, M. (2017). Combination of microfluidic loop-mediated isothermal amplification with gold nanoparticles for rapid detection of Salmonella spp. in food samples. *Frontiers in Microbiology, 8*, 2159. https://doi.org/10.3389/fmicb.2017.02159.
Geng, T., Bao, N., Sriranganathanw, N., Li, L., & Lu, C. (2012). Genomic DNA extraction from cells by electroporation on an integrated microfluidic platform. *Analytical Chemistry, 84*(21), 9632–9639. https://doi.org/10.1021/ac3026064.
Gorgannezhad, L., Umer, M., Islam, M. N., Nguyen, N. T., & Shiddiky, M. J. A. (2018). Circulating tumor DNA and liquid biopsy: Opportunities, challenges, and recent advances in detection technologies. *Lab on a Chip, 18*(8), 1174–1196. http://doi.org/10.1039/c8lc00100f.
Green, N. G., Morgan, H., & Milner, J. J. (1997). Manipulation and trapping of sub-micron bioparticles using dielectrophoresis. *Journal of Biochemical and Biophysical Methods, 35*(2), 89–102. https://doi.org/10.1016/S0165-022X(97)00033-X.
Hakoda, M., & Shiragami, N. (2000). Separation of dead cells from culture broth by using dielectrophoresis. In I. Endo, T. Nagamune, S. Katoh, & T. Yonemoto (Eds.), *Progress in biotechnology* (pp. 53–58). Elsevier. https://doi.org/10.1016/S0921-0423(00)80012-X.
Hald, T., Aspinall, W., Devleesschauwer, B., Cooke, R., Corrigan, T., Havelaar, A. H., … Angulo, F. J. (2016). World Health Organization estimates of the relative contributions of food to the burden of disease due to selected foodborne hazards: A structured expert elicitation. *PLoS ONE, 11*(1), e0145839. https://doi.org/10.1371/journal.pone.0145839.
Han, C.-H., Ha, H. W., & Jang, J. (2019). Two-dimensional computational method for generating planar electrode patterns with enhanced volumetric electric fields and its application to continuous dielectrophoretic bacterial capture. *Lab on a Chip, 19*(10), 1772–1782. https://doi.org/10.1039/c8lc01288a.
Hao, L., Xue, L., Huang, F., Cai, G., Qi, W., Zhang, M., … Lin, J. (2020). A Microfluidic biosensor based on magnetic nanoparticle separation, quantum dots labeling and MnO2 nanoflower amplification for rapid and sensitive detection of salmonella typhimurium. *Micromachines (Basel), 11*(3). https://doi.org/10.3390/mi11030281.
Hao, X., Yeh, P., Qin, Y., Jiang, Y., Qiu, Z., Li, S., … Cao, X. (2019). Aptamer surface functionalization of microfluidic devices using dendrimers as multi-handled templates and its application in sensitive detections of foodborne pathogenic bacteria. *Analytica Chimica Acta, 1056*, 96–107. https://doi.org/10.1016/j.aca.2019.01.035.
Havelaar, A., Kirk, M., Torgerson, P., Gibb, H., Hald, T., Lake, R., Praet, N., Bellinger, D., De Silva, N., & Gargouri, N. (2010). World Health Organization Foodborne Disease Burden Epidemiology Reference Group. 2015. In World Health Organization global estimates and regional comparisons of the burden of foodborne disease in 2010.
Heo, J., Thomas, K. J., Seong, G. H., & Crooks, R. M. (2003). A microfluidic bioreactor based on hydrogel-entrapped E. coli: Cell viability, lysis, and intracellular enzyme reactions. *Analytical Chemistry, 75*(1), 22–26. https://doi.org/10.1021/ac0259717.
Hong, J. W., Chen, Y., Anderson, W. F., & Quake, S. R. (2006). Molecular biology on a microfluidic chip. *Journal of Physics: Condensed Matter, 18*(18), S691. https://doi.org/10.1088/0953-8984/18/18/S14.
Hongwarittorrn, I., Chaichanawongsaroj, N., & Laiwattanapaisal, W. (2017). Semi-quantitative visual detection of loop mediated isothermal amplification (LAMP)-generated DNA by distance-based measurement on a paper device. *Talanta, 175*, 135–142. https://doi.org/10.1016/j.talanta.2017.07.019.
Houssin, T., & Senez, V. (2014). Chapter six – Electrochemical detection. In H. Bridle (Ed.), *Waterborne pathogens* (pp. 147–188). Amsterdam: Academic Press.
Huang, A., Qiu, Z., Jin, M., Shen, Z., Chen, Z., Wang, X., & Li, J.-W. (2014). High-throughput detection of food-borne pathogenic bacteria using oligonucleotide microarray with quantum dots as fluorescent labels. *International Journal of Food Microbiology, 185*, 27–32. https://doi.org/10.1016/j.ijfoodmicro.2014.05.012.
Hwang, J., Cho, Y. H., Park, M. S., & Kim, B. H. (2019). Microchannel fabrication on glass materials for microfluidic devices. *International Journal of Precision Engineering and Manufacturing, 20*(3), 479–495. https://doi.org/10.1007/s12541-019-00103-2.
Islam, M., Shahid, A., Kuryllo, K., Li, Y., Deen, M., & Selvaganapathy, P. (2017). Electrophoretic concentration and electrical lysis of Bacteria in a microfluidic device using a nanoporous membrane. *Micromachines, 8*(2), 45. https://doi.org/10.3390/mi8020045.
Jeong, S. W., Park, Y. M., Jo, S. H., Lee, S. J., Kim, Y. T., & Lee, K. G. (2019). Smartphone operable centrifugal system (SOCS) for on-site DNA extraction from foodborne bacterial pathogen. *Biomicrofluidics, 13*(3), 034111. https://doi.org/10.1063/1.5093752.
Jiang, Y., Chen, S., Zhao, Y., Yang, X., Fu, S., McKillip, J. L., … Man, C. (2020). Multiplex loop-mediated isothermal amplification-based lateral flow dipstick for simultaneous detection of 3 food-borne pathogens in powdered infant formula. *Journal of Dairy Science*. https://doi.org/10.3168/jds.2019-17538.
Jin, Z., Ding, G., Li, G., Yang, G., Han, Y., Hao, N., … Li, W. (2020). Rapid detection of foodborne bacterial pathogens using visual high-throughput microfluidic chip. *Journal of Chemical Technology & Biotechnology, 95*(5), 1460–1466. https://doi.org/10.1002/jctb.6331.
Jung, S. H., Hahn, Y. K., Oh, S., Kwon, S., Um, E., Choi, S., & Kang, J. H. (2018). Advection flows-enhanced magnetic separation for high-throughput bacteria separation from undiluted whole blood. *Small, 14*(34), 1801731. https://doi.org/10.1002/smll.201801731.
Jung, T., Jung, Y., Ahn, J., & Yang, S. (2020). Continuous, rapid concentration of foodborne bacteria (Staphylococcus aureus, Salmonella typhimurium, and Listeria monocytogenes) using magnetophoresis-based microfluidic device. *Food Control, 107229*. https://doi.org/10.1016/j.foodcont.2020.107229.
Kaushik, A. M., Hsieh, K., & Wang, T. H. (2018). Droplet microfluidics for high-sensitivity and high-throughput detection and screening of disease biomarkers. *Wiley Interdisciplinary Reviews: Nanomedicine and Nanobiotechnology, 10*(6), e1522. https://doi.org/10.1002/wnan.1522.
Kim, J., Johnson, M., Hill, P., & Gale, B. K. (2009). Microfluidic sample preparation: Cell lysis and nucleic acid purification. *Integrative Biology, 1*(10), 574–586. https://doi.org/10.1039/b905844c.
Kim, T. H., Park, J., Kim, C. J., & Cho, Y. K. (2014). Fully integrated lab-on-a-disc for nucleic acid analysis of food-borne pathogens. *Analytical Chemistry, 86*(8), 3841–3848. https://doi.org/10.1021/ac403971h.
Ko, J.-M., Ju, J., Lee, S., & Cha, H.-C. (2006). Tobacco protoplast culture in a polydimethylsiloxane-based microfluidic channel. *Protoplasma, 227*(2–4), 237–240. https://doi.org/10.1007/s00709-005-0142-2.
Koczula, K. M., & Gallotta, A. (2016). Lateral flow assays. *Essays in Biochemistry, 60*(1), 111–120. https://doi.org/10.1042/EBC20150012.
Kwon, K., Gwak, H., Hyun, K.-A., Kwak, B.-S., & Jung, H.-I. (2019). High-throughput microfluidic chip for magnetic enrichment and photothermal DNA extraction of foodborne bacteria. *Sensors and Actuators B: Chemical, 294*, 62–68. https://doi.org/10.1016/j.snb.2019.05.007.
Lapizco-Encinas, B. H., Simmons, B. A., Cummings, E. B., & Fintschenko, Y. (2004). Dielectrophoretic concentration and separation of live and dead bacteria in an array of insulators. *Analytical Chemistry, 76*(6), 1571–1579. https://doi.org/10.1021/ac034804j.
Lee, S., Kim, B. W., Shin, H.-S., Go, A., Lee, M.-H., Lee, D.-K., … Jeong, O. C. (2019). Aptamer affinity-bead mediated capture and displacement of gram-negative bacteria using acoustophoresis. *Micromachines, 10*(11), 770. https://doi.org/10.3390/mi10110770.
Lee, S., Mehta, S., & Erickson, D. (2016). Two-color lateral flow assay for multiplex detection of causative agents behind acute febrile illnesses. *Analytical Chemistry, 88*(17), 8359–8363. https://doi.org/10.1021/acs.analchem.6b01828.
Lee, W.-C., Lien, K.-Y., Lee, G.-B., & Lei, H.-Y. (2008). An integrated microfluidic system using magnetic beads for virus detection. *Diagnostic Microbiology and Infectious Disease, 60*(1), 51–58. https://doi.org/10.1016/j.diagmicrobio.2007.07.010.
Lenshof, A., & Laurell, T. (2016). Acoustophoresis. In B. Bhushan (Ed.), *Encyclopedia of nanotechnology* (pp. 56–61). Netherlands, Dordrecht: Springer.
Li, J., Macdonald, J., & von Stetten, F. (2019). Review: A comprehensive summary of a decade development of the recombinase polymerase amplification. *The Analyst, 144*(1), 31–67. https://doi.org/10.1039/c8an01621f.
Li, M., & Anand, R. K. (2019). Integration of marker-free selection of single cells at a wireless electrode array with parallel fluidic isolation and electrical lysis. *Chemical Science, 10*(5), 1506–1513. https://doi.org/10.1039/c8sc04804e.
Li, Y., Zhang, C., & Xing, D. (2010). Fast identification of foodborne pathogenic viruses using continuous-flow reverse transcription-PCR with fluorescence detection.

Microfluidics and Nanofluidics, 10(2), 367–380. https://doi.org/10.1007/s10404-010-0675-3.

Li, Z., Bai, Y., You, M., Hu, J., Yao, C., Cao, L., & Xu, F. (2020). Fully integrated microfluidic devices for qualitative, quantitative and digital nucleic acids testing at point of care. *Biosensors and Bioelectronics, 112952*. https://doi.org/10.1016/j.bios.2020.112952.

Lillehoj, P. B., Kaplan, C. W., He, J., Shi, W., & Ho, C.-M. (2014). Rapid, electrical impedance detection of bacterial pathogens using immobilized antimicrobial peptides. *Journal of Laboratory Automation, 19*(1), 42–49. https://doi.org/10.1177/2211068213495207.

Liu, H.-T., Wen, Z.-Y., Xu, Y., Shang, Z.-G., Peng, J.-L., & Tian, P. (2017). An integrated microfluidic analysis microsystems with bacterial capture enrichment and in-situ impedance detection. *Modern Physics Letters B, 31*(25), 1750233. https://doi.org/10.1142/S0217984917502335.

Luitz, M., Risch, P., Kick, A., Reichel, M., Helmer, D., Kotz, F., & Rapp, B. E. (2020). Facile integration of electronics in glass microfluidic devices for electrochemical synthesis and analysis. In Microfluidics, BioMEMS, and medical microsystems XVIII (Vol. 11235, pp. 112350G). International Society for Optics and Photonics. https://doi.org/10.1117/12.2546353.

Mansfield, M. A. (2005). The use of nitrocellulose membranes in lateral-flow assays. In R. C. Wong, & H. Y. Tse (Eds.), *Drugs of abuse: Body fluid testing* (pp. 71–85). Totowa, NJ: Humana Press.

Mason, P., Dempsey, C., Neilson, G., & Brady, J. (2005). Nanometer-scale ion aggregates in aqueous electrolyte solutions: Guanidinium sulfate and guanidinium thiocyanate. *The Journal of Physical Chemistry B, 109*(50), 24185–24196. https://doi.org/10.1021/jp052799c.

McDonald, J. C., & Whitesides, G. M. (2002). Poly (dimethylsiloxane) as a material for fabricating microfluidic devices. *Accounts of Chemical Research, 35*(7), 491–499. https://doi.org/10.1021/ar010110q.

McGrath, J., Honrado, C., Spencer, D., Horton, B., Bridle, H., & Morgan, H. (2017). Analysis of parasitic protozoa at the single-cell level using microfluidic impedance cytometry. *Scientific Reports, 7*(1), 1–11. https://doi.org/10.1038/s41598-017-02715-y.

Moon, H.-S., Kwon, K., Kim, S.-I., Han, H., Sohn, J., Lee, S., & Jung, H.-I. (2011). Continuous separation of breast cancer cells from blood samples using multi-orifice flow fractionation (MOFF) and dielectrophoresis (DEP). *Lab on a Chip, 11*(6), 1118–1125. https://doi.org/10.1039/C0LC00345J.

Moyano, A., Serrano-Pertierra, E., Salvador, M., Martínez-García, J. C., Rivas, M., & Blanco-López, M. C. (2020). Magnetic lateral flow immunoassays. *Diagnostics (Basel, Switzerland), 10*(5), 288. https://doi.org/10.3390/diagnostics10050288.

Mu, C., Zhang, Z., Lin, M., Dai, Z., & Cao, X. (2015). Development of a highly effective multi-stage surface acoustic wave SU-8 microfluidic concentrator. *Sensors and Actuators B: Chemical, 215*, 77–85. https://doi.org/10.1016/j.snb.2015.03.038.

Muderrisoglu, C., Sargin, S., & Yesil-Celiktas, O. (2018). Application of β-glucuronidase-immobilised silica gel formulation to microfluidic platform for biotransformation of β-glucuronides. *Biotechnology Letters, 40*(5), 773–780. https://doi.org/10.1007/s10529-018-2530-7.

Muluneh, M., Shang, W., & Issadore, D. (2014). Magnetic separation: Track-etched magnetic micropores for immunomagnetic isolation of pathogens (Adv. Healthcare Mater. 7/2014), 950-950 *Advanced Healthcare Materials, 3*(7). https://doi.org/10.1002/adhm.201470034.

Naik, P., Jaitpal, S., Shetty, P., & Paul, D. (2019). An integrated one-step assay combining thermal lysis and loop-mediated isothermal DNA amplification (LAMP) in 30 min from E. coli and M. smegmatis cells on a paper substrate. *Sensors and Actuators B: Chemical, 291*, 74–80. https://doi.org/10.1016/j.snb.2019.04.044.

Nakano, A., & Ros, A. (2013). Protein dielectrophoresis: Advances, challenges, and applications. *Electrophoresis, 34*(7), 1085–1096. https://doi.org/10.1002/elps.201200482.

Ngamsom, B., Esfahani, M. M., Phurimsak, C., Lopez-Martinez, M. J., Raymond, J.-C., Broyer, P., … Pamme, N. (2016). Multiplex sorting of foodborne pathogens by on-chip free-flow magnetophoresis. *Analytica Chimica Acta, 918*, 69–76. https://doi.org/10.1016/j.aca.2016.03.014.

Ngamsom, B., Lopez-Martinez, M. J., Raymond, J.-C., Broyer, P., Patel, P., & Pamme, N. (2016). On-chip acoustophoretic isolation of microflora including S. typhimurium from raw chicken, beef and blood samples. *Journal of Microbiological Methods, 123*, 79–86. https://doi.org/10.1016/j.mimet.2016.01.016.

Nguyen, N.-V., & Jen, C.-P. (2018). Impedance detection integrated with dielectrophoresis enrichment platform for lung circulating tumor cells in a microfluidic channel. *Biosensors and Bioelectronics, 121*, 10–18. https://doi.org/10.1016/j.bios.2018.08.059.

Notomi, T., Mori, Y., Tomita, N., & Kanda, H. (2015). Loop-mediated isothermal amplification (LAMP): Principle, features, and future prospects. *Journal of Microbiology, 53*(1), 1–5. https://doi.org/10.1007/s12275-015-4656-9.

Oh, S. J., Park, B. H., Choi, G., Seo, J. H., Jung, J. H., Choi, J. S., … Seo, T. S. (2016). Fully automated and colorimetric foodborne pathogen detection on an integrated centrifugal microfluidic device. *Lab on a Chip, 16*(10), 1917–1926. https://doi.org/10.1039/c6lc00326e.

Packard, M. M., Wheeler, E. K., Alocilja, E. C., & Shusteff, M. (2013). Performance evaluation of fast microfluidic thermal lysis of bacteria for diagnostic sample preparation. *Diagnostics, 3*(1), 105–116. https://doi.org/10.3390/diagnostics3010105.

Pang, B., Ding, X., Wang, G., Zhao, C., Xu, Y., Fu, K., … Liu, Y. (2017). Rapid and quantitative detection of Vibrio parahemolyticus by the mixed-dye-based loop-mediated isothermal amplification assay on a self-priming compartmentalization microfluidic chip. *Journal of Agricultural and Food Chemistry, 65*(51), 11312–11319. https://doi.org/10.1021/acs.jafc.7b03655.

Pang, B., Fu, K., Liu, Y., Ding, X., Hu, J., Wu, W., … Li, J. (2018). Development of a self-priming PDMS/paper hybrid microfluidic chip using mixed-dye-loaded loop-mediated isothermal amplification assay for multiplex foodborne pathogens detection. *Analytica Chimica Acta, 1040*, 81–89. https://doi.org/10.1016/j.aca.2018.07.024.

Pang, Z., Al-Mahrouki, A., Berezovski, M., & Krylov, S. N. (2006). Selection of surfactants for cell lysis in chemical cytometry to study protein-DNA interactions. *Electrophoresis, 27*(8), 1489–1494. https://doi.org/10.1002/elps.200500732.

Park, B. H., Oh, S. J., Jung, J. H., Choi, G., Seo, J. H., Kim, D. H., … Seo, T. S. (2017). An integrated rotary microfluidic system with DNA extraction, loop-mediated isothermal amplification, and lateral flow strip based detection for point-of-care pathogen diagnostics. *Biosensors and Bioelectronics, 91*, 334–340. https://doi.org/10.1016/j.bios.2016.11.063.

Peham, J. R., Grienauer, W., Steiner, H., Heer, R., Vellekoop, M. J., Nohammer, C., & Wiesinger-Mayr, H. (2011). Long target droplet polymerase chain reaction with a microfluidic device for high-throughput detection of pathogenic bacteria at clinical sensitivity. *Biomed Microdevices, 13*(3), 463–473. https://doi.org/10.1007/s10544-011-9514-x.

Petchakup, C., Li, K. H. H., & Hou, H. W. (2017). Advances in single cell impedance cytometry for biomedical applications. *Micromachines, 8*(3), 87. https://doi.org/10.3390/mi8030087.

Piepenburg, O., Williams, C. H., Stemple, D. L., & Armes, N. A. (2006). DNA detection using recombination proteins. *PLoS Biology, 4*(7), e204. https://doi.org/10.1371/journal.pbio.0040204.

Pinheiro, L. B., Coleman, V. A., Hindson, C. M., Herrmann, J., Hindson, B. J., Bhat, S., & Emslie, K. R. (2012). Evaluation of a droplet digital polymerase chain reaction format for DNA copy number quantification. *Analytical Chemistry, 84*(2), 1003–1011. http://doi.org/10.1021/ac202578x.

Poon, L. L., Wong, B. W., Chan, K. H., Ng, S. S., Yuen, K. Y., Guan, Y., & Peiris, J. M. (2005). Evaluation of real-time reverse transcriptase PCR and real-time loop-mediated amplification assays for severe acute respiratory syndrome coronavirus detection. *Journal of Clinical Microbiology, 43*(7), 3457–3459. https://doi.org/10.1128/JCM.43.7.3457-3459.2005.

Puiu, M., & Bala, C. (2016). SPR and SPR imaging: Recent trends in developing nanodevices for detection and real-time monitoring of biomolecular events. *Sensors (Basel), 16*(6). https://doi.org/10.3390/s16060870.

Qiu, Y., & Park, K. (2001). Environment-sensitive hydrogels for drug delivery. *Advanced Drug Delivery Reviews, 53*(3), 321–339. https://doi.org/10.1016/S0169-409X(01)00203-4.

Qu, J. H., Dillen, A., Saeys, W., Lammertyn, J., & Spasic, D. (2020). Advancements in SPR biosensing technology: An overview of recent trends in smart layers design, multiplexing concepts, continuous monitoring and in vivo sensing. *Analytica Chimica Acta, 1104*, 10–27. https://doi.org/10.1016/j.aca.2019.12.067.

Rackus, D. G., Shamsi, M. H., & Wheeler, A. R. (2015). Electrochemistry, biosensors and microfluidics: A convergence of fields. *Chemical Society Reviews, 44*(15), 5320–5340. https://doi.org/10.1039/c4cs00369a.

Ramadan, Q., Samper, V., Poenar, D., Liang, Z., Yu, C., & Lim, T. M. (2006). Simultaneous cell lysis and bead trapping in a continuous flow microfluidic device. *Sensors and Actuators B: Chemical, 113*(2), 944–955. https://doi.org/10.1016/j.snb.2005.04.018.

Regiart, M., Fernández-Baldo, M. A., Villarroel-Rocha, J., Messina, G. A., Bertolino, F. A., Sapag, K., … Raba, J. (2017). Microfluidic immunosensor based on mesoporous silica platform and CMK-3/poly-acrylamide-co-methacrylate of dihydrolipoic acid modified gold electrode for cancer biomarker detection. *Analytica Chimica Acta, 963*, 83–92. https://doi.org/10.1016/j.aca.2017.01.029.

Ren, K., Zhou, J., & Wu, H. (2013). Materials for microfluidic chip fabrication. *Accounts of Chemical Research, 46*(11), 2396–2406. https://doi.org/10.1021/ar300314s.

Sajid, M., Kawde, A.-N., & Daud, M. (2015). Designs, formats and applications of lateral flow assay: A literature review. *Journal of Saudi Chemical Society, 19*(6), 689–705. https://doi.org/10.1016/j.jscs.2014.09.001.

San Park, T., Li, W., McCracken, K. E., & Yoon, J.-Y. (2013). Smartphone quantifies Salmonella from paper microfluidics. *Lab on a Chip, 13*(24), 4832–4840. https://doi.org/10.1039/c3lc50976a.

Shangguan, J., Li, Y., He, D., He, X., Wang, K., Zou, Z., & Shi, H. (2015). A combination of positive dielectrophoresis driven on-line enrichment and aptamer-fluorescent silica nanoparticle label for rapid and sensitive detection of Staphylococcus aureus. *Analyst, 140*(13), 4489–4497. https://doi.org/10.1039/C5AN00535C.

Sharif, S., Wang, Y., Ye, Z., Wang, Z., Qiu, Q., Ying, S., & Ying, Y. (2019). A novel impedimetric sensor for detecting LAMP amplicons of pathogenic DNA based on magnetic separation. *Sensors and Actuators B: Chemical, 301*, 127051. https://doi.org/10.1016/j.snb.2019.127051.

Shaw, K. J., Thain, L., Docker, P. T., Dyer, C. E., Greenman, J., Greenway, G. M., & Haswell, S. J. (2009). The use of carrier RNA to enhance DNA extraction from microfluidic-based silica monoliths. *Analytica Chimica Acta, 652*(1), 231–233. https://doi.org/10.1016/j.aca.2009.03.038.

Shin, J. H., Hong, J., Go, H., Park, J., Kong, M., Ryu, S., … Park, J.-K. (2018). Multiplexed detection of foodborne pathogens from contaminated lettuces using a handheld multistep lateral flow assay device. *Journal of Agricultural and Food Chemistry, 66*(1), 290–297. https://doi.org/10.1021/acs.jafc.7b03582.

Shin, J. H., & Park, J.-K. (2016). Functional packaging of lateral flow strip allows simple delivery of multiple reagents for multistep assays. *Analytical Chemistry, 88*(21), 10374–10378. https://doi.org/10.1021/acs.analchem.6b02869.

Shin, S., Kim, B., Kim, Y.-J., & Choi, S. (2019). Integrated microfluidic pneumatic circuit for point-of-care molecular diagnostics. *Biosensors and Bioelectronics, 133*, 169–176. https://doi.org/10.1016/j.bios.2019.03.018.

Shu, B., Zhang, C., & Xing, D. (2014). Segmented continuous-flow multiplex polymerase chain reaction microfluidics for high-throughput and rapid foodborne pathogen detection. *Analytica Chimica Acta, 826*, 51–60. https://doi.org/10.1016/j.aca.2014.04.017.

Song, C., Liu, C., Wu, S., Li, H., Guo, H., Yang, B., … Liu, Q. (2016). Development of a lateral flow colloidal gold immunoassay strip for the simultaneous detection of Shigella boydii and Escherichia coli O157:H7 in bread, milk and jelly samples. *Food Control, 59*, 345–351. https://doi.org/10.1016/j.foodcont.2015.06.012.

Strohmeier, O., Emperle, A., Roth, G., Mark, D., Zengerle, R., & von Stetten, F. (2013). Centrifugal gas-phase transition magnetophoresis (GTM)–a generic method for automation of magnetic bead based assays on the centrifugal microfluidic platform and application to DNA purification. *Lab on a Chip, 13*(1), 146–155. https://doi.org/10.1039/c2lc40866j.

Strohmeier, O., Keller, M., Schwemmer, F., Zehnle, S., Mark, D., von Stetten, F., … Paust, N. (2015). Centrifugal microfluidic platforms: Advanced unit operations and applications. *Chemical Society Reviews, 44*(17), 6187–6229. https://doi.org/10.1039/c4cs00371c.

Strohmeier, O., Marquart, N., Mark, D., Roth, G., Zengerle, R., & von Stetten, F. (2014). Real-time PCR based detection of a panel of food-borne pathogens on a centrifugal microfluidic "LabDisk" with on-disk quality controls and standards for quantification. *Analytical Methods, 6*(7), 2038. https://doi.org/10.1039/c3ay41822g.

Sun, W. (2010). Chapter 4 – Nucleic extraction and amplification. In W. W. Grody, R. M. Nakamura, C. M. Strom, & F. L. Kiechle (Eds.), *Molecular diagnostics* (pp. 35–47). San Diego: Academic Press.

Sun, Z., Peng, Y., Zhang, M., Wang, K., Bai, J., Li, X., … Gao, Z. (2014). Simultaneous and highly sensitive detection of six different foodborne pathogens by high-throughput suspension array technology. *Food Control, 40*, 300–309. https://doi.org/10.1016/j.foodcont.2013.11.028.

Tadmor, A. D., Ottesen, E. A., Leadbetter, J. R., & Phillips, R. (2011). Probing individual environmental bacteria for viruses by using microfluidic digital PCR. *Science, 333* (6038), 58–62. http://doi.org/10.1126/science.1200758.

Tang, Y., Zeng, X., & Liang, J. (2010). Surface plasmon resonance: An introduction to a surface spectroscopy technique. *Journal of Chemical Education, 87*(7), 742–746. https://doi.org/10.1021/ed100186y.

Tao, Z.-Y., Zhou, H.-Y., Xia, H., Xu, S., Zhu, H.-W., Culleton, R. L., … Gao, Q. (2011). Adaptation of a visualized loop-mediated isothermal amplification technique for field detection of Plasmodium vivax infection. *Parasites & Vectors, 4*(1), 115. https://doi.org/10.1186/1756-3305-4-115.

Teh, S. Y., Lin, R., Hung, L. H., & Lee, A. P. (2008). Droplet microfluidics. *Lab on a Chip, 8* (2), 198–220. https://doi.org/10.1039/b715524g.

Thissen, J. B., Be, N. A., McLoughlin, K., Gardner, S., Rack, P. G., Shapero, M. H., Rowland, R. R., Slezak, T., & Jaing, C. J. (2019). Axiom Microbiome Array, the next generation microarray for high-throughput pathogen and microbiome analysis. *PLoS ONE, 14*(2). https://doi.org/10.1371/journal.pone.0212045.

Tian, F., Lyu, J., Shi, J., Tan, F., & Yang, M. (2016). A polymeric microfluidic device integrated with nanoporous alumina membranes for simultaneous detection of multiple foodborne pathogens. *Sensors and Actuators B: Chemical, 225*, 312–318. https://doi.org/10.1016/j.snb.2015.11.059.

Tillmann, R. L., Simon, A., Müller, A., & Schildgen, O. (2007). Sensitive commercial NASBA assay for the detection of respiratory syncytial virus in clinical specimen. *PLoS ONE, 2*(12), e1357. https://doi.org/10.1371/journal.pone.0001357.

Tokel, O., Yildiz, U. H., Inci, F., Durmus, N. G., Ekiz, O. O., Turker, B., … Demirci, U. (2015). Portable microfluidic integrated plasmonic platform for pathogen detection. *Scientific Reports, 5*, 9152. https://doi.org/10.1038/srep09152.

Tomlinson, J. A., Boonham, N., & Dickinson, M. (2010). Development and evaluation of a one-hour DNA extraction and loop-mediated isothermal amplification assay for rapid detection of phytoplasmas. *Plant Pathology, 59*(3), 465–471. https://doi.org/10.1111/j.1365-3059.2009.02233.x.

Van Kessel, J. A. S., Karns, J. S., Lombard, J. E., & Kopral, C. A. (2011). Prevalence of salmonella enterica, listeria monocytogenes, and escherichia coli virulence factors in bulk tank milk and in-line filters from U.S. Dairies†. *Journal of Food Protection, 74*(5), 759–768. https://doi.org/10.4315/0362-028x.jfp-10-423.

Van Nguyen, H., Nguyen, V. D., Lee, E. Y., & Seo, T. S. (2019). Point-of-care genetic analysis for multiplex pathogenic bacteria on a fully integrated centrifugal microdevice with a large-volume sample. *Biosensors and Bioelectronics, 136*, 132–139. https://doi.org/10.1016/j.bios.2019.04.035.

Vandeventer, P. E., Mejia, J., Nadim, A., Johal, M. S., & Niemz, A. (2013). DNA adsorption to and elution from silica surfaces: Influence of amino acid buffers. *The Journal of Physical Chemistry B, 117*(37), 10742–10749. https://doi.org/10.1021/jp405753m.

Varshney, M., Li, Y., Srinivasan, B., & Tung, S. (2007). A label-free, microfluidics and interdigitated array microelectrode-based impedance biosensor in combination with nanoparticles immunoseparation for detection of Escherichia coli O157:H7 in food samples. *Sensors and Actuators B: Chemical, 128*(1), 99–107. https://doi.org/10.1016/j.snb.2007.03.045.

Verma, M. S., Tsaloglou, M.-N., Sisley, T., Christodouleas, D., Chen, A., Milette, J., & Whitesides, G. M. (2018). Sliding-strip microfluidic device enables ELISA on paper. *Biosensors and Bioelectronics, 99*, 77–84. https://doi.org/10.1016/j.bios.2017.07.034.

Wang, L., Chen, W., Ma, W., Liu, L., Ma, W., Zhao, Y., … Xu, C. (2011). Fluorescent strip sensor for rapid determination of toxins. *Chemical Communications, 47*(5), 1574–1576. https://doi.org/10.1039/C0CC04032K.

Wang, R., Xu, Y., Liu, H., Peng, J., Irudayaraj, J., & Cui, F. (2017). An integrated microsystem with dielectrophoresis enrichment and impedance detection for detection of Escherichia coli. *Biomedical Microdevices, 19*(2), 34. https://doi.org/10.1007/s10544-017-0167-2.

Wang, S., Xie, J., Jiang, M., Chang, K., Chen, R., Ma, L., … Hu, J. (2016). The development of a portable SPR bioanalyzer for sensitive detection of escherichia coli O157: H7. *Sensors, 16*(11), 1856. https://doi.org/10.3390/s16111856.

Wasey, A., Yang, J., Sun, D., He, Y., & Zhang, C. (2020). On-chip Carba NP test for accurate and high throughput detection of carbapenemase-producing Enterobacteriaceae. *Talanta, 210*, 120656. https://doi.org/10.1016/j.talanta.2019.120656.

Wei, X.-Y., Li, J.-H., Wang, L., & Yang, F. (2019). Low-voltage electrical cell lysis using a microfluidic device. *Biomedical Microdevices, 21*(1), 22. https://doi.org/10.1007/s10544-019-0369-x.

Weng, X., Jiang, H., & Li, D. (2012). Electrokinetically-controlled RNA-DNA hybridization assay for foodborne pathogens. *Microchimica Acta, 178*(3–4), 381–387. https://doi.org/10.1007/s00604-012-0853-y.

Wernecke, M., & Mullen, C. (2014). Molecular biology|Molecular biology in microbiological analysis. In C. A. Batt, & M. L. Tortorello (Eds.), *Encyclopedia of food microbiology* (2nd ed., pp. 808–814). Oxford: Academic Press.

Xia, Y., Liu, Z., Yan, S., Yin, F., Feng, X., & Liu, B.-F. (2016). Identifying multiple bacterial pathogens by loop-mediated isothermal amplification on a rotate & react slipchip. *Sensors and Actuators B: Chemical, 228*, 491–499. https://doi.org/10.1016/j.snb.2016.01.074.

Xie, S., Yuan, Y., Chai, Y., & Yuan, R. (2015). Tracing phosphate ions generated during loop-mediated isothermal amplification for electrochemical detection of nosema bombycis genomic DNA PTP1. *Analytical Chemistry, 87*(20), 10268–10274. https://doi.org/10.1021/acs.analchem.5b01858.

Xu, Y., Xie, X., Duan, Y., Wang, L., Cheng, Z., & Cheng, J. (2016). A review of impedance measurements of whole cells. *Biosensors and Bioelectronics, 77*, 824–836. https://doi.org/10.1016/j.bios.2015.10.027.

Yan, H., Zhu, Y., Zhang, Y., Wang, L., Chen, J., Lu, Y., … Xing, W. (2017). Multiplex detection of bacteria on an integrated centrifugal disk using bead-beating lysis and loop-mediated amplification. *Scientific Reports, 7*(1), 1460. https://doi.org/10.1038/s41598-017-01415-x.

Yang, L. (2009). Dielectrophoresis assisted immuno-capture and detection of foodborne pathogenic bacteria in biochips. *Talanta, 80*(2), 551–558. https://doi.org/10.1016/j.talanta.2009.07.024.

Yao, L., Wang, L., Huang, F., Cai, G., Xi, X., & Lin, J. (2018). A microfluidic impedance biosensor based on immunomagnetic separation and urease catalysis for continuous-flow detection of E. coli O157:H7. *Sensors and Actuators B: Chemical, 259*, 1013–1021. https://doi.org/10.1016/j.snb.2017.12.110.

Yasui, T., Yanagida, T., Shimada, T., Otsuka, K., Takeuchi, M., Nagashima, K., … Yonese, A. (2019). Engineering nanowire-mediated cell lysis for microbial cell identification. *ACS Nano, 13*(2), 2262–2273. https://doi.org/10.1021/acsnano.8b08959.

Yetisen, A. K., Akram, M. S., & Lowe, C. R. (2013). Paper-based microfluidic point-of-care diagnostic devices. *Lab on a Chip, 13*(12), 2210–2251. https://doi.org/10.1039/c3lc50169h.

Yin, J., Zou, Z., Hu, Z., Zhang, S., Zhang, F., Wang, B., … Mu, Y. (2020). A "sample-in-multiplex-digital-answer-out" chip for fast detection of pathogens. *Lab on a Chip, 20* (5), 979–986. https://doi.org/10.1039/c9lc01143a.

Yu, H., Meyvantsson, I., Shkel, I. A., & Beebe, D. J. (2005). Diffusion dependent cell behavior in microenvironments. *Lab on a Chip, 5*(10), 1089–1095. https://doi.org/10.1039/b504403k.

Zhang, C.-S., Li, Y.-Y., & Wang, H.-Y. (2011). Rapid amplification and detection of foodborne pathogenic rotavirus by continuous-flow reverse transcription-polymerase chain reaction integrated with online fluorescence analysis. *Chinese Journal of Analytical Chemistry, 39*(5), 645–651. https://doi.org/10.1016/s1872-2040(10)60436-7.

Zhang, H., Huang, F., Cai, G., Li, Y., & Lin, J. (2018). Rapid and sensitive detection of Escherichia coli O157: H7 using coaxial channel-based DNA extraction and microfluidic PCR. *Journal of Dairy Science, 101*(11), 9736–9746. https://doi.org/10.3168/jds.2018-14730.

Zhang, H., Xu, Y., Fohlerova, Z., Chang, H., Iliescu, C., & Neuzil, P. (2019). LAMP-on-a-chip: Revising microfluidic platforms for loop-mediated DNA amplification. *Trends in Analytical Chemistry, 113*, 44–53. https://doi.org/10.1016/j.trac.2019.01.015.

Zhang, J., Shen, Z., Xiang, Y., & Lu, Y. (2016). Integration of solution-based assays onto lateral flow device for one-step quantitative point-of-care diagnostics using personal glucose meter. *Acs Sensors, 1*(9), 1091–1096. https://doi.org/10.1021/acssensors.6b00270.

Zhao, X., & Dong, T. (2012). Multifunctional sample preparation kit and on-chip quantitative nucleic acid sequence-based amplification tests for microbial detection. *Analytical Chemistry, 84*(20), 8541–8548. https://doi.org/10.1021/ac3020609.

Zhao, X., Li, M., & Liu, Y. (2019). Microfluidic-based approaches for foodborne pathogen detection. *Microorganisms, 7*(10). https://doi.org/10.3390/microorganisms7100381.

Zhao, Y., Wang, H., Zhang, P., Sun, C., Wang, X., Wang, X., … Zhou, L. (2016). Rapid multiplex detection of 10 foodborne pathogens with an up-converting phosphor technology-based 10-channel lateral flow assay. *Scientific Reports, 6*, 21342. https://doi.org/10.1038/srep21342.

Zhu, Q., Gao, Y., Yu, B., Ren, H., Qiu, L., Han, S., … Mu, Y. (2012). Self-priming compartmentalization digital LAMP for point-of-care. *Lab on a Chip, 12*(22), 4755–4763. https://doi.org/10.1039/c2lc40774d.