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ABSTRACT

Slip-driven microfluidic devices can manipulate fluid by the relative movement of microfluidic plates that are in close contact. Since the demonstration of the first SlipChip device, many slip-driven microfluidic devices with different form factors have been developed, including SlipPAD, SlipDisc, sliding stripe, and volumetric bar chart chip. Slip-driven microfluidic devices can be fabricated from glass, quartz, polydimethylsiloxane, paper, and plastic with various fabrication methods: etching, casting, wax printing, laser cutting, micromilling, injection molding, etc. The slipping operation of the devices can be performed manually, by a micrometer with a base station, or autonomously, by a clockwork mechanism. A variety of readout methods other than fluorescence microscopy have been demonstrated, including both fluorescence detection and colorimetric detection by mobile phones, direct visual detection, and real-time fluorescence imaging. This review will focus on slip-driven microfluidic devices for nucleic acid analysis, including multiplex nucleic acid detection, digital nucleic acid quantification, real-time nucleic acid amplification, and sample-in-answer-out nucleic acid analysis. Slip-driven microfluidic devices present promising approaches for both life science research and clinical molecular diagnostics.

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I. INTRODUCTION

Partitioning fluid is a critical step for many biological and chemical methods, such as multiplex analysis, high-throughput screening, and the study of single cells or single molecules. Partitioning can be performed with pipetting steps with tubes or microtiter plates and often involves many cumbersome manual operations or costly liquid-handling robotic systems. Moreover, the throughput is generally limited by the number of test tubes or wells on 96- or 384-well plates. Because these methods generally handle liquid in the microliter and milliliter regimes, performing a screening panel may consume a large amount of reagents, which can sometimes be cost prohibitive.

Due to the rapid development of microfluidic technology, partitioning can now be scaled down to much smaller volumes, such as nanoliters, picoliters, and subpicoliters, with much higher throughput. Microdroplets with nanoliter or smaller volumes can be generated and manipulated by several different microfluidic approaches. For example, flow-driven droplet formation can generate droplets in a high-throughput manner;^{1–5} however, achieving

narrow volume distributions of the droplets generally requires precise control of the fluidic flow of both the carrier and dispersed liquid. Pneumatic microvalves embedded in a microfluidic device can be used to compartmentalize the liquid in the fluidic channel,^{6,7} but this system still relies on the pneumatic control system. Devices with microwells were developed for partitioning liquid in small volumes,^{8–10} and electrowetting-based microfluidic devices can also generate and manipulate droplets.^{11–14} Moreover, arrays of droplets can be directly printed on a solid substrate with the assistance of pumps or robotic systems.^{15–17}

Since the demonstration of the SlipChip concept in 2009,¹⁸ there have been different slip-driven microfluidic devices presented for a broad spectrum of applications, especially nucleic acid analysis. These microfluidic devices generally consist of at least two modules that can change relative position by a slipping motion to connect and disconnect fluidic paths (Fig. 1). Several different materials such as glass,¹⁸ polydimethylsiloxane (PDMS),¹⁹ paper,²⁰ and plastic²¹ have been applied to fabricate these devices. These devices can control and manipulate a small amount of fluid in an

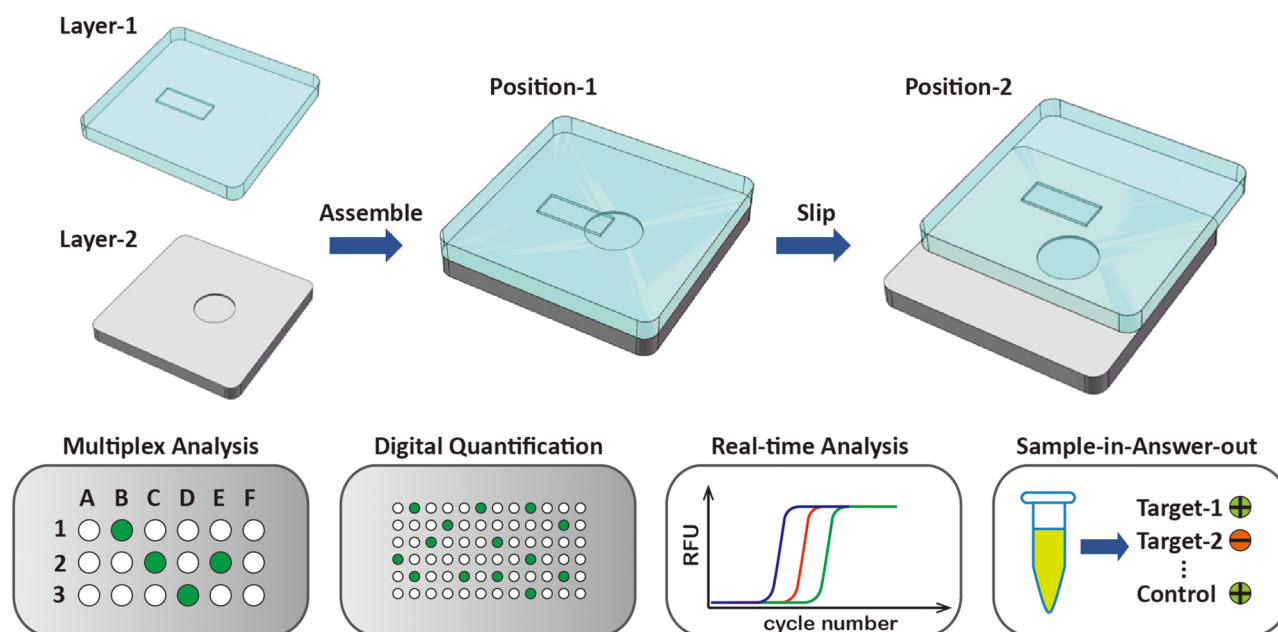


FIG. 1. Slip-driven microfluidic device for nucleic acid analysis. Layer-1 and layer-2 can be assembled into position-1, with rectangular features on layer-1 and circular features on layer-2 partially overlapping. Layer-1 is slipped against layer-2 to position-2 to disconnect the rectangular feature from the circular feature. The slip-driven microfluidic devices can be applied for multiplex analysis, digital quantification, real-time analysis, and sample-in-answer-out analysis.

“instrument-free” fashion. To demonstrate the robustness and easy-to-use nature of a SlipChip device, a six-year-old volunteer performed an experiment to generate thousands of nanoliter microdroplets on an injection-molded plastic SlipChip with simple manual operations.²¹

The use of these slip-driven microfluidic devices has been demonstrated in multiplex nucleic acid analysis for the identification of target molecules,^{22–24} digital nucleic acid quantification of target molecules,^{25–31} real-time nucleic acid analysis,^{32–34} and sample-in-answer-out nucleic acid analysis.^{20,23} In addition to nucleic acid analysis, slip-driven microfluidic devices have also been applied to the study of protein crystallization,^{35,36} immunoassays,^{37–40} cell and bacterial cultivation,^{19,41,42} bacterial chemotaxis,⁴³ separation by isoelectric focusing^{44,45} or electrophoresis,⁴⁶ antimicrobial susceptibility testing (AST),⁴⁷ sample storage,⁴⁸ etc. In this review, we will focus on the progress in slip-driven microfluidic devices for nucleic acid analysis, including their materials, fabrication methods, and related applications. We will also provide our view of the opportunities and challenges for the use of these devices in related molecular diagnostics and scientific research.

II. GENERAL PRINCIPLE OF SLIP-DRIVEN MICROFLUIDIC TECHNOLOGY

A. Materials and fabrication

Slip-driven microfluidic devices have been made from a variety of base materials such as glass, PDMS, paper, and plastic through different fabrication processes. The selection of material

and fabrication methods depends primarily on the specific applications and use cases. For instance, applications that rely on a fluorescence readout will require the material to have relatively good light transmission near the target excitation and emission wavelengths. In another scenario, diagnostic applications targeting resource-limited settings require the device to be inexpensively fabricated, and the device should be disposable. Very often, the selection of materials and fabrication methods can determine the ease of transforming the concept of slip-driven microfluidic devices into manufacturable products at scale. Therefore, it is important to evaluate the selection of materials and fabrication methods from a systematic and application-driven perspective.

1. Glass for slip-driven microfluidic devices

A wet etching process with glass or quartz material is commonly used for the preparation of slip-driven microfluidic devices. Glass materials, such as soda-lime glass or borofloat glass, and quartz have superior optical properties. They have high transparency over a broad range of light wavelengths. Therefore, these materials are favorable for applications that require a fluorescence readout or other light-dependent analyses. Glass material can provide a flat surface, which is critical for slip-driven microfluidic devices, such as SlipChips. The glass material is also relatively rigid (low elasticity), which makes the microfluidic plates less likely to deform during the slipping process. Furthermore, the glass material is chemically inert and can also provide a good barrier to prevent water evaporation during the incubation process.

Photolithography and hydrofluoric acid (HF)-based wet etching can be applied to fabricate slip-driven microfluidic devices from glass or quartz materials. Du *et al.*¹⁸ applied this method to produce a glass-based SlipChip device. In brief, a glass plate coated with chromium and photoresist can be aligned with a photomask with designed patterns and then exposed to ultraviolet (UV) light. The exposed area can be removed by NaOH solution, and the underlying chromium layer can be etched off in chromium etchant. Then, HF-based wet etching can be performed in a temperature-controlled shaking water bath. This etching process is isotropic, which will make the pattern expand in both the x and y directions while etching occurs downward in the z direction. This method can be used to generate features down to tens of micrometers in size and can provide relatively smooth finishing. By utilizing multiple photolithography processes, Shen *et al.*²⁶ also demonstrated SlipChip devices with microscale features of different depths from wet etching of glass materials [Fig. 2(a)].

2. Paper for slip-driven microfluidic devices

Paper-based microfluidics have been demonstrated for many applications, particularly low-cost diagnostics in resource-limited settings.^{14,40,49–52} The merits of being lightweight, low in cost, flexible, foldable, easy-to-fabricate, and easy-to-dispose make paper a very attractive material for slip-driven microfluidic devices. Furthermore, paper microfluidics can drive aqueous fluid by wicking, which makes the device favorable for “instrument-free” operation. Connelly *et al.*²⁰ demonstrated a slip-driven “paper machine” that integrates sample

preparation and loop-mediated isothermal amplification (LAMP) for nucleic acid analysis [Fig. 2(b)]. Li *et al.*⁵³ presented a paper-based SlipPAD device integrated with an electrochemical sensor for hepatitis B virus (HBV) DNA analysis.

3. PDMS for slip-driven microfluidic devices

PDMS casting is a method commonly used to fabricate microfluidic devices, and it was also applied to prepare slip-driven microfluidic devices. PDMS is a polymeric organosilicon compound that is commonly referred to as a silicone and is optically clear and chemically inert. Since the introduction of rapid prototyping of microfluidic devices in PDMS by Dr. George Whitesides in 1998,⁵⁴ PDMS has been applied in the fabrication of a broad spectrum of microfluidic systems. Chang *et al.*¹⁹ demonstrated a PDMS SlipChip device for *in vitro* cell culture, cell cocultures, and consequent analysis. Performing nucleic acid amplification generally requires an elevated temperature, and there is a risk of water loss through the porous structure of PDMS. Moreover, compared to glass materials, PDMS is more elastomeric. During the slipping operation, there can be a risk of deformation. These issues may be potentially addressed by binding the PDMS layer to a more rigid, less water-permeable supporting layer.

4. Plastic for slip-driven microfluidic devices

Various types of plastic materials with different fabrication processes were demonstrated for the preparation of slip-driven

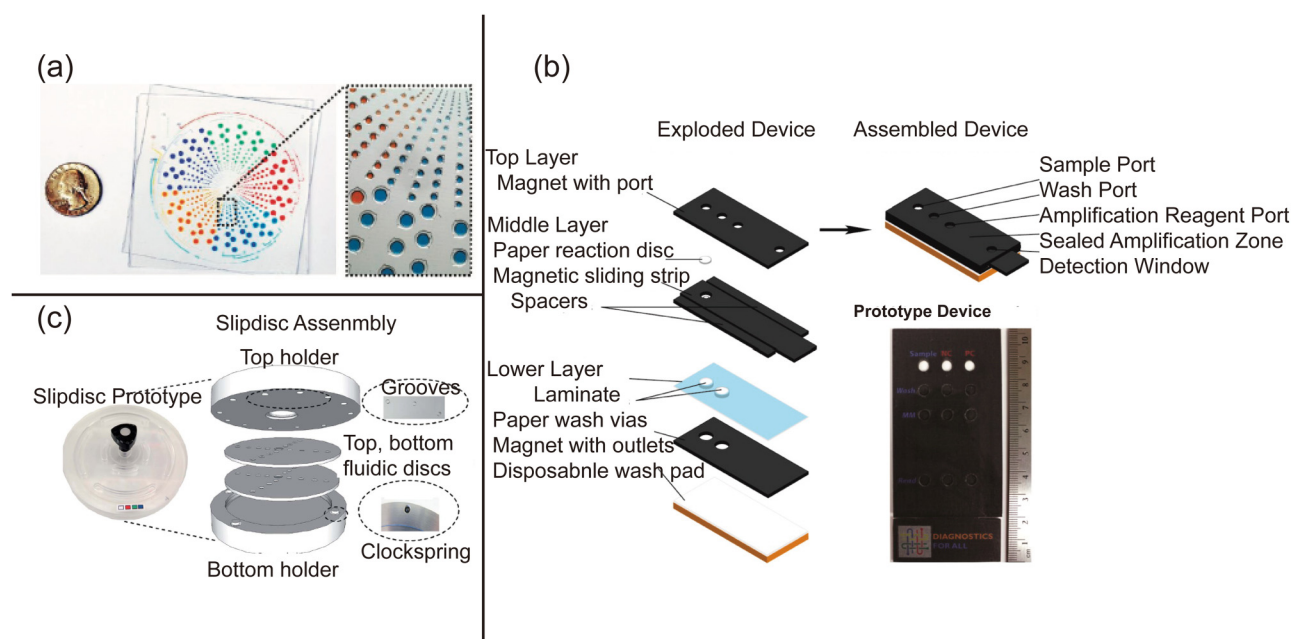


FIG. 2. Examples of slip-driven microfluidic devices fabricated from different materials. (a) A rotational SlipChip device fabricated from glass. Reprinted with permission from Shen *et al.*, *J. Am. Chem. Soc.* **133**(44), 17705–17712 (2011). Copyright 2011 American Chemical Society. (b) A sliding paper-based microfluidic device. Reprinted with permission from Connelly *et al.*, *Anal. Chem.* **87**(15), 7595–7601 (2015). Copyright 2015 American Chemical Society. (c) A plastic SlipDisc. Reprinted with permission from Banerjee *et al.*, *RSC Adv.* **7**(56), 35048–35054 (2017). Copyright 2017 The Royal Society of Chemistry.

microfluidic devices. Tsaloglou *et al.*³² reported a SlipChip device for recombinase polymerase amplification (RPA) with a microfluidic plate of poly(methyl methacrylate) (PMMA) fabricated by micromilling. Banerjee *et al.*⁵⁵ demonstrated a SlipDisc fabricated from laser-cut polyethylene terephthalate (PET) films bound to optically transparent polycarbonate (PC) disks [Fig. 2(c)]. Schoepp *et al.*⁵⁶ presented digital LAMP quantification for rapid pathogen-specific phenotypic antibiotic susceptibility testing using disposable injection-molded SlipChip devices. Injection molding is a common manufacturing process used to produce parts and products on a large scale, and its cost per unit can be relatively low. Furthermore, injection molding can introduce additional 3-dimensional structures to microfluidic devices. The development of an injection-molded SlipChip⁵⁶ was a significant milestone for this type of device to become a viable commercial product [Fig. 3(a)].

B. Operation and readout of slip-driven microfluidic devices

1. Slipping operation

Slip-driven microfluidic devices rely on the relative movement of different modules to establish or disconnect the fluidic path and

to form partitions for further analysis. A controlled and user-friendly method to perform this slipping operation is critical to make these slip-driven microfluidic devices available for a broad range of use cases outside research laboratories. This slipping motion can be achieved manually with the assistance of embedded slipping guidance²¹ [Fig. 3(a)] or a stereomicroscope.²² This slipping operation can also be driven by a screw gauge on a base holder to precisely move one plate against the other [Fig. 3(b)].³² Furthermore, this process can also be performed in an autonomous manner by implementing a hand-wound clockwork mechanism for precise rotational movement of the SlipDisc [Fig. 3(c)].⁵⁵

2. Reagent loading and fluidic control in slip-driven microfluidic devices

The slip-driven microfluidic devices generally integrate with easy-loading features that do not require fluidic pumps or other complex fluidic control systems. In addition, several internal on-device fluidic control features can further simplify the loading process. For instance, Li *et al.*⁵⁷ reported a dead-end filling feature on the SlipChip device that allows filling with an aqueous solution without leaking by balancing the loading pressure, capillary pressure,

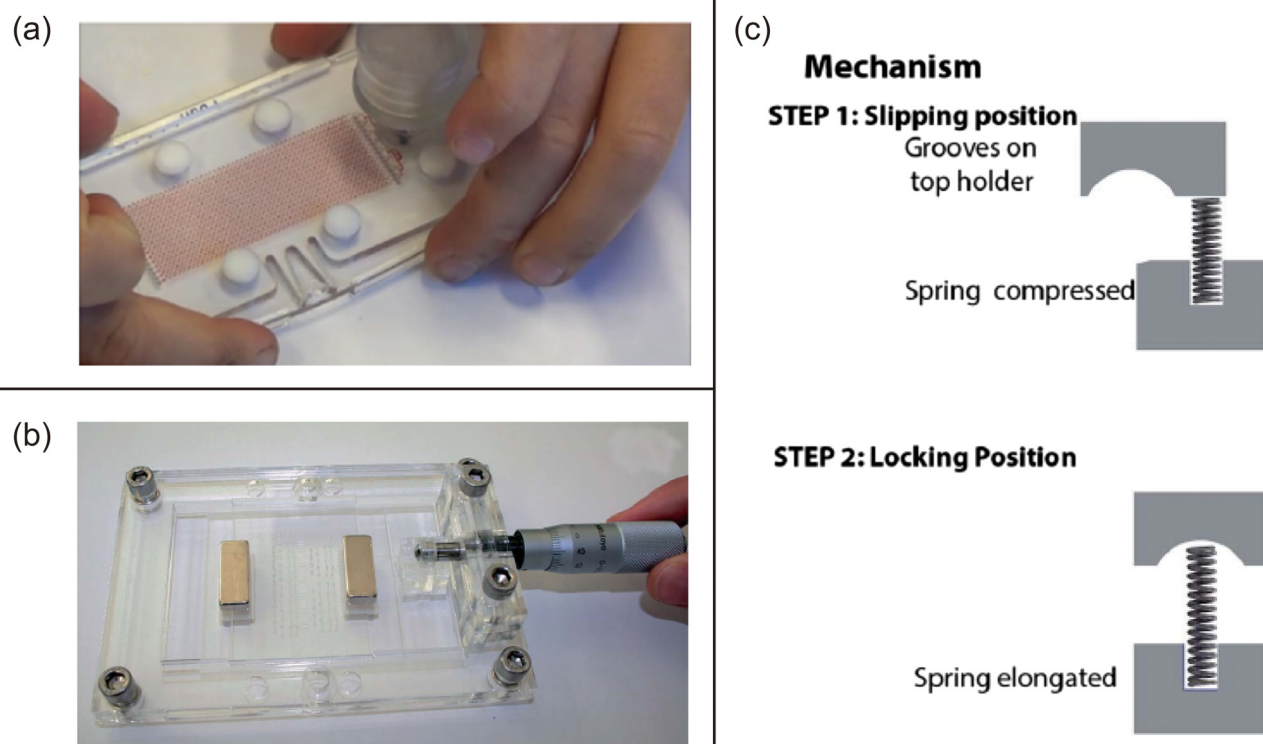


FIG. 3. Examples of different operating mechanisms of slip-driven microfluidic devices. (a) Manual slipping with embedded slipping guidance on the device. Reprinted with permission from Begolo *et al.*, *Lab Chip* **14**(24), 4616–4628 (2014). Copyright 2014 The Royal Society of Chemistry. (b) Slipping by a micrometer on a base frame. Reprinted with permission from Tsaloglou *et al.*, *Analyst* **140**(1), 258–264 (2015). Copyright 2015 The Royal Society of Chemistry. (c) Clockwork mechanism for precise rotational movement of the SlipDisc. Reprinted with permission from Banerjee *et al.*, *RSC Adv.* **7**(56), 35048–35054 (2017). Copyright 2017 The Royal Society of Chemistry.

and sealing pressure. Pompano *et al.*⁵⁸ also described a method driven by capillary pressure to initiate and control the fluidic flow in the microfluidic channels on the SlipChip device. Many slip-driven devices have been presented with simple manual pipette loading under research conditions.^{22,25,32} The pipetting pressure can provide positive loading pressure that drives aqueous solution into the fluidic channels. This approach is very convenient for research laboratories where pipetting steps are performed routinely; however, it can be less practical for reagent loading in resource-limited settings. To bridge this gap, Begolo *et al.*²¹ demonstrated a pumping lid method that can provide positive or negative pressure to introduce the solution into the SlipChip devices.

3. Readout of the slip-driven microfluidic devices

Various readout methods were developed for the slip-driven microfluidic devices for different applications and use cases. In research laboratories, fluorescence microscopes, which can vary magnification, exposure conditions, and wavelengths, offer great flexibility for the investigation of new devices and/or new assays.

Apart from that, Selck and Ismagilov³³ reported an optical system for real-time fluorescence imaging on the SlipChip device, and the system can be used for mechanistic investigation and optimization of digital assays [Fig. 4(a)]. However, for scenarios outside of laboratories, such as experiments in resource-limited settings, a portable reader or direct visual readout would be greatly favorable. Selck *et al.*²⁹ developed a mobile phone-based portable fluorescence imaging device that images digital isothermal amplification results on SlipChip devices. Xia *et al.*²⁴ presented a rotational SlipChip device that uses LAMP and calcein, and a pattern of green fluorescence was directly visible under a UV flashlight in the dark [Fig. 4(b)]. Rodriguez-Manzano *et al.*⁵⁹ applied an unmodified cell phone to image digital amplification of DNA and RNA on a SlipChip device with colorimetric amplification-indicator dyes [Fig. 4(c)]. Song *et al.*⁶⁰ developed a multistage volumetric bar chart chip (MV-Chip) that integrates cascade amplification steps controlled by multistep slipping to detect target DNA with visual bar chart advancements [Fig. 4(d)]. Li *et al.*⁵³ also integrated electrochemical sensors on a paper-based SlipPAD device for the quantitative detection of HBV DNA.

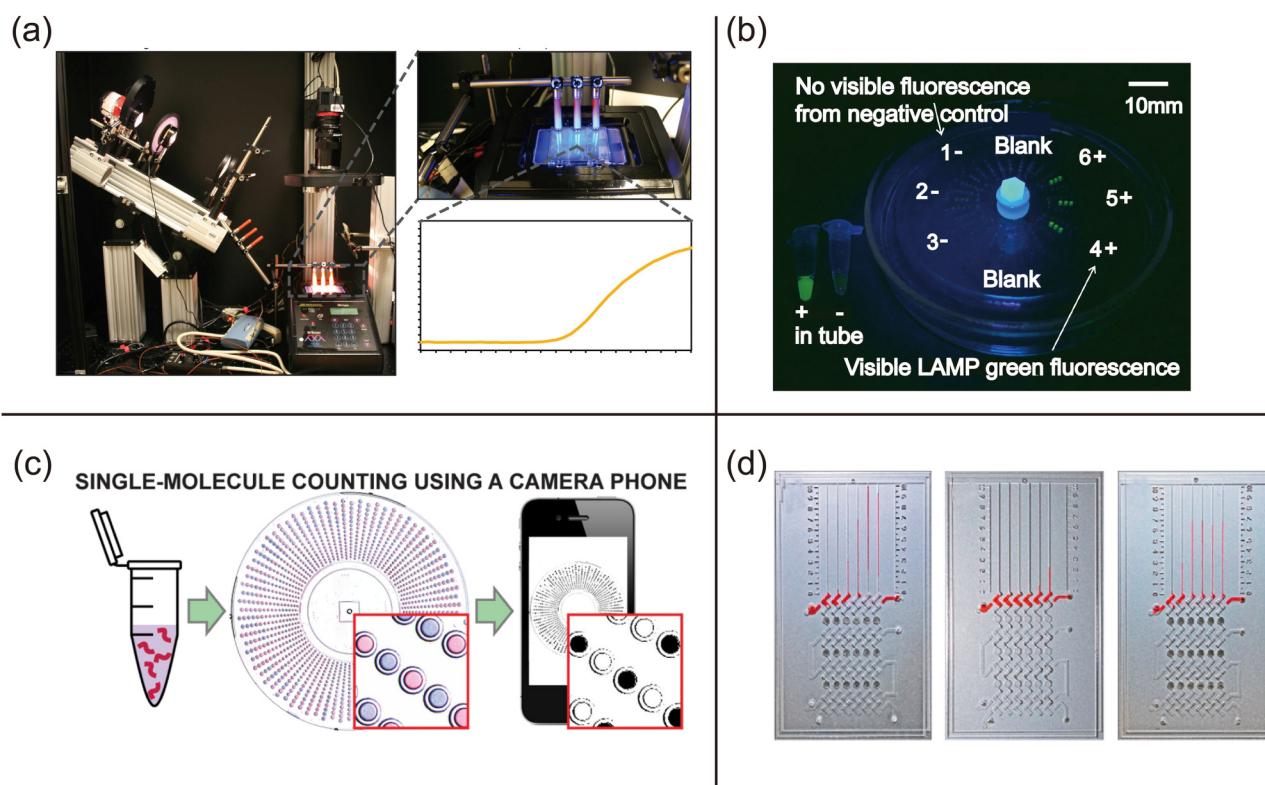


FIG. 4. Examples of detection and analysis methods for slip-driven microfluidic devices. (a) A real-time fluorescence imaging system. Reprinted with permission from Selck and Ismagilov, PLoS One 11(10), e0163060 (2016). Copyright 2016 Selck and Ismagilov. (b) Visual analysis of LAMP with calcein on a SlipDisc. Reprinted with permission from Xia *et al.*, Sens. Actuators B Chem. 228, 491–499 (2016). Copyright 2016 Elsevier B.V. (c) Colorimetric analysis of a SlipChip device with an unmodified mobile phone. Reprinted with permission from Rodriguez-Manzano *et al.*, ACS Nano 10(3), 3102–3113 (2016). Copyright 2016 American Chemical Society. (d) Visual bar chart advancements for result analysis. Reprinted with permission from Song *et al.*, J. Am. Chem. Soc. 135(45), 16785–16788 (2013). Copyright 2013 American Chemical Society.

III. NUCLEIC ACID ANALYSIS IN SLIP-DRIVEN MICROFLUIDIC DEVICES

A. Multiplex nucleic acid analysis in slip-driven microfluidic devices

The first demonstration of nucleic acid analysis using a slip-driven microfluidic device was the nanoliter multiplex polymerase chain reaction (PCR) on a SlipChip system developed by Shen *et al.*²² The device was fabricated from glass by the wet etching method. The glass provided good optical transparency for the detection of the PCR amplification product fluorescence. The surface of this SlipChip device was silanized to be hydrophobic, and mineral oil was placed between the two plates of the SlipChip device. Therefore, aqueous solutions, such as PCR mix, could form nanoliter droplets in the device without wetting the surface or leaking into the gap. The low water permeability of glass and the presence of mineral oil prevented the evaporation of nanoliter droplets during thermal cycling. The amplification product could be recovered from the SlipChip device for further analysis, such as gel electrophoresis, to confirm the specificity of target amplification. Shen *et al.*²² developed a SlipChip device that contains 384 nanoliter wells for screening up to 384 different target genes with a single FAM fluorescence channel on a $1 \times 3 \text{ in.}^2$ glass slide [Fig. 5(a)]. The device was demonstrated with multiplex colony PCR for 20 sepsis-related pathogens, including *Escherichia coli*, *Pseudomonas aeruginosa*, *Staphylococcus aureus*, methicillin-resistant *Staphylococcus aureus* (MRSA), and *Candida albicans*, with both positive and negative amplification controls in approximately 2 h.

In clinical diagnostic settings, it is preferred to identify causative pathogens in clinical specimens without time-consuming cultivation

and sample preparation. Cai *et al.*²³ utilized dielectrophoresis to directly extract pathogens from blood samples and then identified the pathogens by multiplex nanoliter PCR arrays with end point fluorescence detection on a SlipChip device [Fig. 5(b)]. Dielectrophoresis allows broad-spectrum separation of bacterial and fungal species in physiological fluids. The multiplex PCR array could identify up to 20 pathogenic species from blood samples within 3 h.

Furthermore, Xia *et al.*²⁴ developed a rotational SlipChip device for the visual identification of multiple bacterial pathogens by LAMP with calcein. Positive LAMP with calcein generates bright green fluorescence under UV light that can be utilized for direct visual readout. This device identified pathogens among common digestive bacteria, such as *Salmonella enterica*, *Bacillus cereus*, *E. coli*, and *Vibrio parahaemolyticus*, in approximately 1 h.

B. Slip-driven microfluidic devices for digital nucleic acid analysis

Digital nucleic acid analysis is considered a sensitive and absolute quantification method for target nucleic acids. It divides a solution into a large number of partitions, some of which contain one or more of the target nucleic acid templates and others that do not. After nucleic acid amplification, the partitions containing the target nucleic acid exhibit greater fluorescence than the partitions that do not. Based on the fluorescence intensity, these partitions can be classified into groups “1” and “0.” Analyzing the data with Poisson statistics can precisely determine the concentration of target nucleic acid in the sample.²⁸ Digital nucleic acid analysis has been demonstrated in droplet, microchamber, and microchip

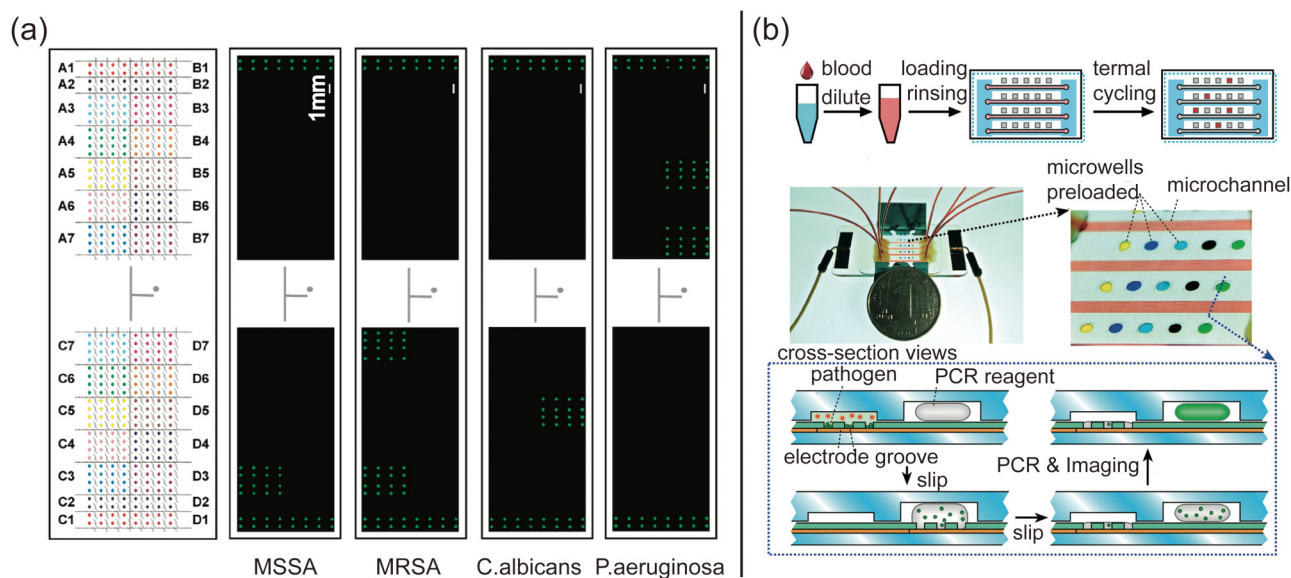


FIG. 5. Examples of slip-driven microfluidic devices for multiplex nucleic acid analysis. (a) A SlipChip device with a panel of preloaded primers for multiplex nanoliter PCR, including methicillin-sensitive *S. aureus* (MSSA), methicillin-resistant *S. aureus* (MRSA), *C. albicans*, and *P. aeruginosa*. Reprinted with permission from Shen *et al.*, *Anal. Chem.* **82**(11), 4606–4612 (2010). Copyright 2010 American Chemical Society. (b) A device that integrates dielectrophoresis with multiplex nanoliter PCR. Reprinted with permission from Cai *et al.*, *Lab Chip* **14**, 3917–3924 (2014). Copyright 2014 The Royal Society of Chemistry.

TABLE I. Specification of representative slip-driven microfluidic devices for digital nucleic acid analysis.

Target	Assay	Droplet volume (nl)	Number of partitions	Slipping step	Reference
<i>S. aureus nuc</i> gene	PCR	2.6	1280	1	25
HIV and HCV viral RNA	RT-PCR	625	80	1	26
		125	160		
		25	160		
		5	160		
		1	160		
		0.2	160		
Methicillin-resistant <i>S. aureus</i> genomic DNA	RPA	9	1550	2	27
HIV viral RNA	RT-LAMP	...	...	2	30
<i>E. coli</i> DNA	LAMP	2.4	5376	1	56
HBV viral DNA	LAMP	12	576	8	31

formats, and it has been previously summarized and discussed in other review articles.^{61–64}

SlipChips can compartmentalize aqueous solution into a large number of small-volume partitions by relative movement of two microfluidic plates that are in close contact (Table I). They do not require fluidic pumps or complex fluidic control systems. Shen *et al.*²⁵ demonstrated a digital PCR SlipChip to perform quantification of the *S. aureus nuc* gene. The device consisted of 1280 of 2.6 nl microwells on a 1.5×1 in.² area [Fig. 6(a)].

Many biochemical and clinical applications, such as viral load analysis, require nucleic acid quantification over a large dynamic range. To extend the dynamic range, instead of increasing the well density and the total well number, Shen *et al.*²⁶ developed a multi-volume approach to perform digital nucleic acid quantification with a dynamic range of over 5 orders of magnitude. To achieve the same dynamic range, this multivolume SlipChip requires significantly fewer microwells and much smaller device footprints than the single-volume design.²⁸ Different designs of the multivolume SlipChip were shown to achieve the desired dynamic range, sensitivity, and quantification resolution. Shen *et al.*²⁶ demonstrated a rotational SlipChip device containing 160 wells of five volumes each (200 pl, 1 nl, 5 nl, 25 nl, and 125 nl) and 80 wells of 625 nl to simultaneously quantify five samples for HIV and hepatitis C virus (HCV) viral load analysis [Fig. 6(b)]. Yu *et al.*³¹ also utilized serial dilution nanoliter arrays generated by a multistep SlipChip device to achieve a quantification dynamic range of 7 orders of magnitude with fewer than 600 wells in total, and this method was applied for HBV viral load quantification [Fig. 6(c)].

Furthermore, several digital isothermal amplifications on SlipChip devices were also developed to demonstrate the feasibility of nucleic acid quantification in resource-limited settings and to evaluate the performance of digital isothermal assays. For diagnostics in resource-limited settings, isothermal amplification of nucleic acids could be more favorable than PCR methods since it can amplify at a constant temperature and does not require thermal cycling. Some isothermal reactions, such as RPA, have optimum reaction temperatures close to room temperature. Even at room temperature, some background amplification occurs. It takes time to generate partitions for digital amplification, which may cause overestimation of the target nucleic acid concentration. Shen *et al.*²⁷

demonstrated a two-step SlipChip device that first divided the sample into nanoliter partitions and then utilized an additional slipping step to add a chemical initiator, such as magnesium acetate, to simultaneously initiate over 1000 RPA reactions for digital quantification of MRSA genes at approximately 37 °C [Fig. 6(d)]. Moreover, Sun *et al.*³⁰ utilized this two-step digital process to evaluate and optimize the performance of digital reverse transcription LAMP for quantification of HIV viral RNA. They improved the reaction efficiency by partitioning the viral RNA molecules first to produce cDNA through reverse transcription and then using a second slipping step to perform LAMP amplification on individual cDNA molecules. Schoepp *et al.*⁵⁶ also reported a digital LAMP SlipChip method to determine the phenotypic antibiotic susceptibility of *E. coli* from urine samples within 30 min, which provides a promising method for fast antimicrobial susceptibility testing (AST).

C. Real-time slip-driven microfluidic systems for nucleic acid analysis

Real-time detection and analysis systems were also developed for slip-driven microfluidic devices for the quantification of nucleic acids and the investigation of reaction mechanisms. Tsaloglou *et al.*³² developed a real-time RPA system for quantitative measurement of the *Clostridium difficile* toxin B gene with exonuclease fluorescence probes from six replicate 500 nl reaction wells and two sets of 500 nl control wells. Selck and Ismagilov³³ presented an instrument for the analysis of digital nucleic acid amplification on the SlipChip device in real time, and the system was validated for both digital real-time PCR and digital real-time LAMP. Khorosheva *et al.*³⁴ utilized a real-time digital analysis system to investigate the correlation between reaction speed and analytical sensitivity in digital RT-LAMP and to optimize the efficiency of digital RT-LAMP on SlipChip devices.

D. Slip-driven microfluidic device for sample-in-answer-out nucleic acid analysis

Slip-driven microfluidic devices can manipulate and control fluid without complex auxiliary equipment, and this advantage makes it promising for biochemical and diagnostic applications in resource-limited settings. Outside of research laboratories, user-friendly and integrated solutions without specialized skilled

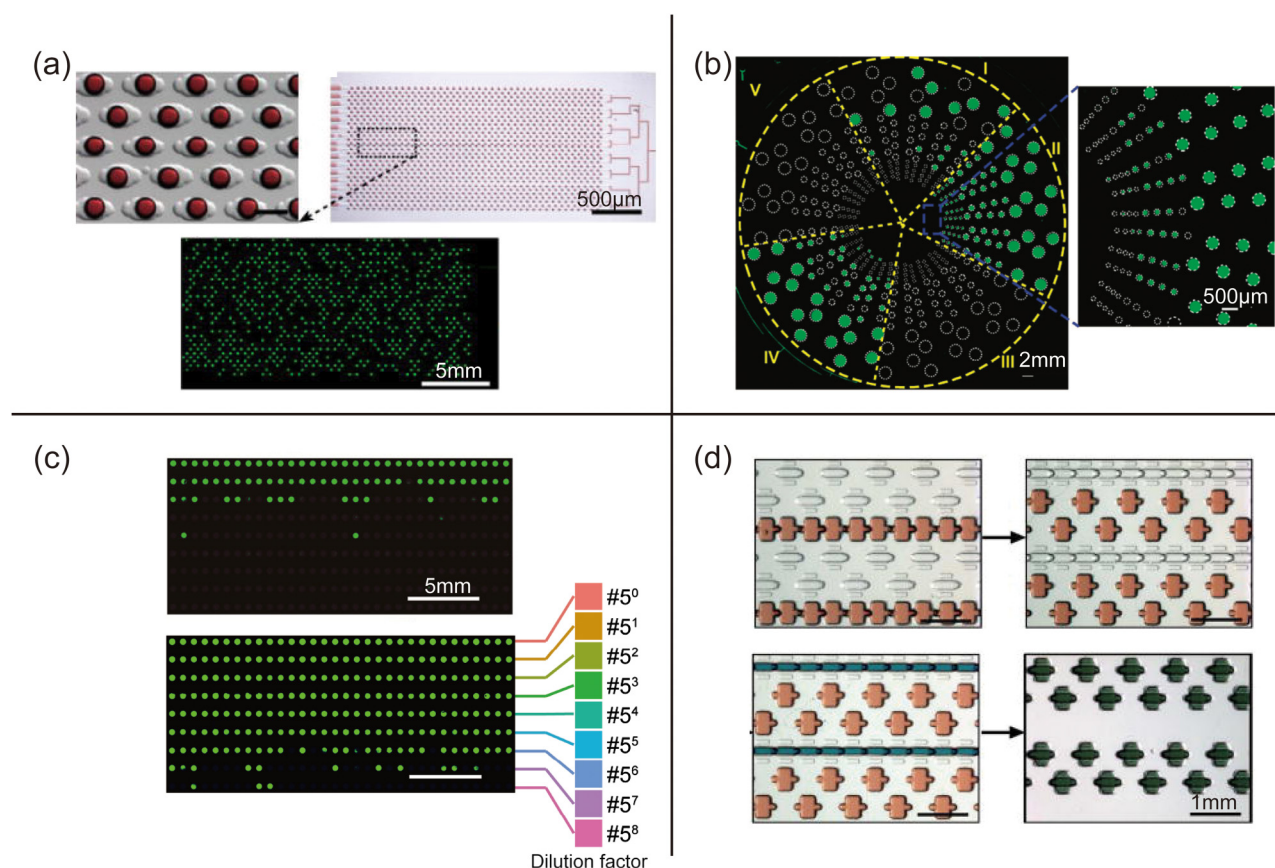


FIG. 6. Examples of slip-driven microfluidic devices for digital nucleic acid analysis. (a) A SlipChip device for digital PCR quantification of an *S. aureus* gene. Reprinted with permission from Shen *et al.*, Lab Chip 10(20), 2666–2672 (2010). Copyright 2010 The Royal Society of Chemistry. (b) A multivolume SlipChip device for simultaneous quantification of 5 samples with a large dynamic range for analysis of HIV and HCV viral loads. Reprinted with permission from Shen *et al.*, J. Am. Chem. Soc. 133(44), 17705–17712 (2011). Copyright 2011 American Chemical Society. (c) A multistep SlipChip for the generation of serial dilution nanoliter arrays and HBV viral load quantification by digital LAMP; Reprinted with permission from Yu *et al.*, Anal. Chem. (in press). Copyright 2019 American Chemical Society. (d) A two-step SlipChip device for digital RPA for isothermal quantification of the MRSA *mecA* gene. Reprinted with permission from Shen *et al.*, Anal. Chem. 83(9), 3533–3540 (2011). Copyright 2011 American Chemical Society.

technical personnel are preferred. A large number of slip-driven microfluidic devices are focused on downstream nucleic acid analysis, and it could be greatly impactful to integrate these devices with sample preparation processes, including raw sample preparation, nucleic acid extraction, and nucleic acid purification and concentration. To date, only very limited work has been demonstrated on this front. Cai *et al.*²³ integrated a dielectrophoresis module to extract pathogens from whole blood samples and then identified pathogens by multiplex PCR on the same SlipChip device. Connelly *et al.*²⁰ reported a paper-based sliding-strip device that integrates sample preparation and LAMP for nucleic acid analysis and allows the introduction of samples, wash buffers, amplification reagents, and detection reagents with serial slipping steps.

IV. DISCUSSION AND PERSPECTIVES

Here, we have reviewed the recent development of slip-driven microfluidic devices for nucleic acid analysis. Built with different

materials, fabrication methods, and form factors, these devices shared one thing in common: they can manipulate fluid with simple slipping movements. This merit makes them very appealing both for analysis in research laboratories and for molecular diagnostics in clinical settings, especially in resource-limited or point-of-care settings. Some SlipChip devices also perform parallel manipulation of droplets in a high throughput manner by multiple slipping steps, such as the addition of a second reagent to individual droplets. This capability makes slip-driven microfluidic devices very attractive for the investigation of multistep reactions in small volumes. The SlipChip device can also be applied for the simultaneous generation of droplets with different predesigned volumes for biochemical analysis with a large dynamic range. Despite these advantages, several areas remain to be further developed.

First, inexpensively manufacturing slip-driven microfluidic devices on a large scale is important for these devices to be widely applied in research and diagnostic settings. The first SlipChip device was fabricated from a glass substrate by wet etching

methods. Materials such as glass and PDMS have good physical and chemical properties and are great for testing new designs and concepts with fast turnaround times. However, it could be cost prohibitive to manufacture glass or PDMS devices at a large scale. Large-scale industrial manufacturing methods, such as injection molding or roll-to-roll manufacturing,^{65,66} would be great for manufacturing slip-driven microfluidic devices at low cost. Making SlipChip devices from plastic material by injection molding was a significant milestone,⁵⁶ and it supports the feasibility of manufacturing the devices on a large scale. In addition to scaling up with low unit cost, manufacturing methods should also demonstrate reproducibility, predictability, and robustness in device fabrication.

Second, for diagnostic applications, especially in point-of-care settings, it is critical to integrate the nucleic acid sample preparation step with the nucleic acid analysis device. It is intriguing that several concepts for slip-driven microfluidic devices integrating sample preparation with nucleic acid amplification have been reported. However, more could be done for nucleic acid sample preparation from complex sample types, such as blood, saliva, urine, stool, and swab samples. To ensure the high quality of nucleic acid analysis performance on slip-driven microfluidic devices, the sample preparation module needs to be effective and efficient. There have been several microfluidic approaches for on-chip nucleic acid sample preparation,^{67,68} and it will be interesting to see if these existing methods or any new methods can be seamlessly integrated with the slip-driven microfluidic devices.

Third, for many applications, it will be greatly informative if the partitioned droplets can be recovered after amplification for further analysis, such as nucleic acid sequencing. The amplification product can be recovered from these devices in a pooled format for gel electrophoresis.²² However, the position information provided by the device has not yet been fully utilized. The capability to recover and further investigate each individual partition would be very helpful for many single-molecule and single-cell analysis applications.

Finally, the current slip-driven microfluidic devices are mostly two layers with two-dimensional movements, such as movement in the x and y directions and rotation. Three-dimensional movement, such as that obtained by the addition of movement in the z direction, could further extend the capability of the platform. In addition, multilayer devices that can perform separate functions in each layer but can still be connected or disconnected through slipping motion are another potential direction for further development.

V. CONCLUSION

In this review, we have summarized the recent development in slip-driven microfluidic devices for nucleic acid analysis. We have discussed the use of different materials and fabrication methods to prepare the devices and described the operation and readout methods for the slip-driven microfluidic devices. Slip-driven microfluidic devices have been reported for multiplex nucleic acid detection, digital nucleic acid quantification, real-time nucleic acid amplification, and sample-in-answer-out nucleic acid analysis with physiological samples. We envision that slip-driven microfluidic devices can serve as promising platforms for biochemical and medical studies, food and environmental testing, and clinical molecular diagnostics.

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