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Recent Advances in Nanoparticles-based Lateral Flow Biosensors

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Abstract

Lateral flow biosensors (LFBs) provide advantages in low cost, simplicity, rapidity, stability and portability, thus making LFBs popular in biomedical, agriculture, food and environmental sciences. This article reviews recent advances of LFBs for bioassays, including the investigation of the improvements achieved by signal-amplification strategies, the application of new nanoparticle labels, novel quantitative system and simultaneous detection. We summarized the outstanding performances of LFBs such as high detection sensitivity, specificity, reproducibility and reliability.

Keywords: lateral flow biosensor, point-of-care testing, nanoparticles, bioassays

1. Introduction

In the last decades, the detecting of biothreat agents, chemical contaminants and biomolecules has attracted much attention and various new methods including real-time polymerase chain reaction (RT-PCR) [1-4], DNA microarrays [5-8], enzyme linked immunosorbent assay (ELISA) [9-13], HPLC-MS [14-17] and immunosensor

techniques [18-23] have been employed. These methods offer high accuracy, but often require time-consuming sample pretreatment, technical expertise, sophisticated and expensive instrumentation, which are inappropriate for point-of-care testing and home testing. Lateral flow biosensors (LFBs), as a promising tool for detecting analyte, have recently attracted considerable interest because of their speed, simplicity, sensitivity, specificity, easy handling, and can potentially provide "instantaneous" diagnosis near the patients [24-33]. Originally the LFBs was used in human chorionic gonadotrophin (HCG) immunoassay and called "sol particle immunoassay" (SPIA) [34,35]. Since then, the LFB was widely applied in environmental monitoring, food safety, and clinical diagnosis for visual (qualitative) and quantitative detecting of various biomolecules and chemical contaminants such as infectious agents [36-41], nucleic acids [42,30,43], proteins [44,33], cells [28], veterinary drugs [45-50], toxins [51-56] and pesticides [57,58]. The principle of LFBs is based on a solid-phase chromatographic test strip containing dry reagents those are activated by applying the fluid sample. Antibodies are immobilized on different positions of the LFB to capture target antigens and the colored detector reagents labeled on antibodies give the colored responses on the test zone and control zone on the LFB. The characteristic colored bands enable visual detection of target analyte without instrumentation. For quantitative measurements, the optical intensities of the test zone and control zone could be recorded simultaneously by corresponding instrumentation [59,60].

The aim of this article is to review major advances in recent years in LFB development and to compare the currently evaluated LFB with the traditional LFB. New strategies from these studies were presented and discussed including:

1. investigation of the improvements achieved by signal-amplification strategies;
2. application of new nanoparticle labels;
3. introducing of novel quantitative system;
4. simultaneous detection of multiple analytes.

2. LFB assay formats and principle

The LFB are designed to test the absence or presence of target analyte. The analyte might be the pathogens, hormones, drugs or metabolites. A specific labeled-antigen or labeled-antibody to the analyte is used for recognition. The LFB consists of four sections: sample application pad, conjugate pad, membrane and absorption pad. All of the components are laminated on a common sheet of plastic adhesive backing orderly using the clamshell laminator. Each pad overlaps 2 mm to allow the migration of sample solution along the LFB during the analysis.

Various possible formats have been described for LFBs, which are depending on the types of target analytes. The competitive assay and sandwich assay are the two most frequently used formats. When the target analyte is with low molecular weight or presenting single specific antigen, it is suitable for employing competitive format [61,62,48]. There are two approached based on the competitive format: 1) the specific antibody is firstly dispensed on the test zone. After applying the sample solution (containing the target antigen and the labeled antigen) to the sample application pad, the target antigen and labeled antigen would compete for binding on the specific antibody, which is immobilized on the test zone; 2) the sample solution containing target antigen is firstly incubated with the specific antibody to form the antigen-antibody complex. The antigen-antibody complex and labeled antibody would compete for binding on the specific antigen-protein conjugate, which is immobilized on the test zone [63].

As shown in Figure 1, in a typical competitive format, a monoclonal antibody (specific to target analyte-progesterone) immobilized on the test zone of a nitrocellulose (NC) membrane captures a colored reagent labeled analyte conjugate (progesterone-ovalbumin/GNPs conjugate), enabling colored reagent (colloidal gold nanoparticles (GNPs)) to accumulate on the test zone and form a characteristic red line on the test zone. The control line was immobilized by another specific protein to capture the excess colored reagent labeled conjugate. In the present (or higher than

the cutoff value) of the target analyte, the target analyte (progesterone) and colored reagent labeled analyte (progesterone-ovalbumin/GNPs conjugate) would compete for binding on the test zone. Owing to the binding affinity and steric effect, the target analyte would bind on the test zone prior to colored reagent labeled analyte. In this case, only one characteristic red band can be observed from the control zone. Conversely, in the absence (or lower than the cutoff value) of target analyte, numerous colored reagent-labeled analyte would bind on test zone, and two characteristic red lines can be observed from the test zone and control zone respectively.

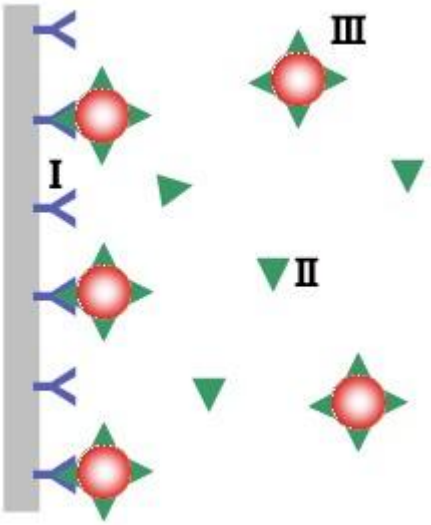

Figure 1. The principle of the competitive thin-layer immuno-affinity chromatography: Ⅰ) monoclonal antibody to progesterone immobilized to membrane; Ⅱ) progesterone; Ⅲ) colloidal gold labeled progesterone-ovalbumin conjugate; Ⅳ,nitrocellulose membrane strip.

On the other hand, when the target analyte owns more than one specific antibodies or DNA aptamers, sandwich format could be employed to test the target analyte [64-70,33,71]. Figure 2 presents the typical DNA aptamers-based sandwich format. A pair of specific DNA aptamers (two aptamer probes, which could bind target thrombin in two different sites) was employed to test target analyte (thrombin). During the detection process, a colored reagent labeled DNA aptamer (GNPs-aptamer 1) was placed on the conjugate pad (a glass-fiber membrane) to serve as the detection reagent; another DNA aptamer (aptamer 2) was immobilized on the test zone to serve as the capture reagent. On the control zone, an additional DNA probe (complementary with the detection aptamer 1) could be used to produce a control signal. When a sample solution was applied on the sample application pad, it would migrate up by capillary action and rehydrate the colored reagent labeled DNA aptamer (GNPs-aptamer 1) conjugates on the conjugate pad. Then the binding between the colored reagent labeled DNA aptamer (GNPs-aptamer 1) and target analyte (thrombin) occurred. Subsequently, the mixture passed through the test zone and the bounded target analyte was captured by capture reagent (aptamer 2). The response from the test zone is in proportion to the concentration of the target analyte. The schematic illustration is depicted in Fig. 2B.

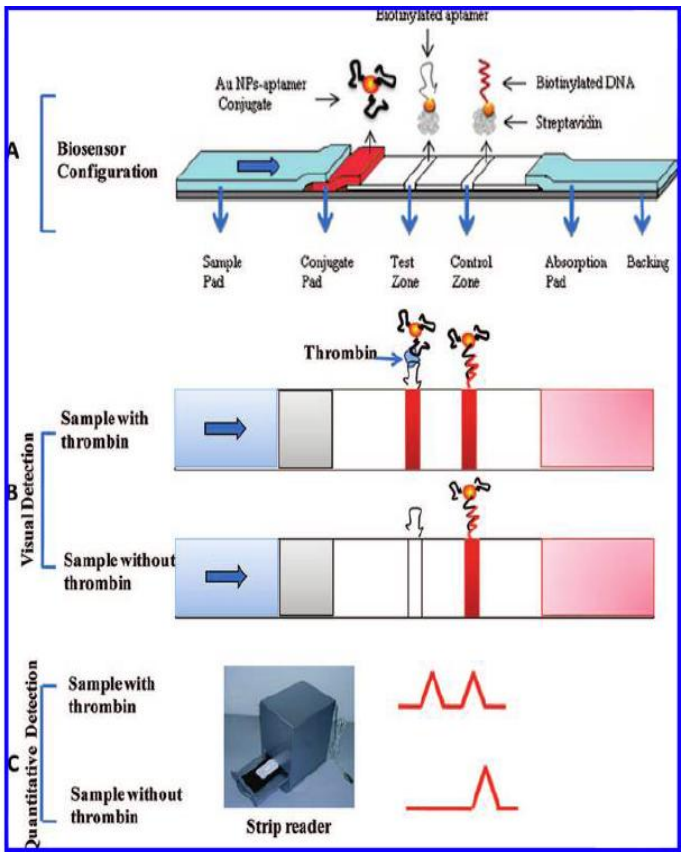

Figure 2. The sandwich format based LFB for thrombin detection [33].

To date, this LFB based technology has reached many fields of research such as medicine, agriculture, food and environmental safety. Recently, several review papers describing LFB [63,72,41,73,74], including the paper reported by Posthuma-Trumpie et al. [73] for detection of infectious agents and chemical contaminants and the paper reported by Babacar Ngom et al. [41] for the strengths, weaknesses, opportunities and threats of the LFB. Herein, we aimed to present recent advances of LFBs for biomedical detections, including the investigation of the improvements achieved by signal-amplification strategies, the application of new nanoparticle labels, novel quantitative system and the simultaneous detection.

3. Signal-amplification strategies

Colored detector reagents in LFBs such as colloidal god nanoparticle (GNPs), latex, europium, chelate-loaded silica, fluorescent immunoliposomes, green microparticle, etc. are used as labels of antibody or aptamer to detect the presence of target analyte. Among the colored particles, GNPs have been extensively employed due to their inherent advantages, such as intense optical properties, vivid color, hybridization and melting properties, fine biocompatibility, excellent chemical tailorability, distance and aggregate size-dependent optical properties. Thus, antibody or aptamer-GNPs have been used to develop LFBs for a wide variety of analytes. Due to low abundance of targets, high sensitivity is highly desired for LFBs. An effective way for improving the sensitivity of the GNPs-based bioassay is to amplify the Au signal by adopting silver enhancement technology to the traditional bioassay [75-81]. Under the action of the reducing agent, silver ions is prone to gather around the GNPs in the form of silver, the color of the test zone is greatly enhanced due to the silver deposition on the GNPs surface. Wang et al. [78] reported a silver enhancement technology-based sandwich immuno-chromatographic assay for detecting abrin-a. In a typical assay, a range of abrin-a concentrations (0, 0.1, 1.0, 10, 100 and 1000 ng/mL) were measured. The accumulation of the GNPs on the test zone and control zone enabled the visual detection within 10 min. Then, the $AgNO_3$ pad and reducer pad were orderly covered on the test zone and control zone. Followed by applying 100 μL double distilled water to infiltrate the two pads, the enhancement result of the test strips was determined within 10 min. The detection sensitivity was 0.1 ng/mL and demonstrated to be increased by 100-fold in comparison with the conventional LFB assay. Furthermore, no significant cross-reactivity can be obtained from the test strip, showed excellent detection specificity.

Another alternative approach for enhancing the detection limit is to introduce enzymes as labels into the development of biosensors and bioassays. Horseradish peroxi-dase (HRP), glucose oxidase and alkaline phosphatase are the most commonly used enzymes in the LFBs bioassays [82-84,68,85-87]. Parolo and co-workers [68] presented a HRP–GNPs dual-labeled LFB for the sensitive detection of Human IgG (HIgG). This strategy was combining the unique optical properties of GNPs and enzyme catalytic amplification, which produce darker products depositing on the GNP surface to enhance the visual effect and the response intensities. Three different HRP substrates, including 3,3,5,5-Tetramethylbenzidine (TMB), 3-Amino-9-ethylcarbazole (AEC), 3,3-Diaminobenzidinetetrahydrochloride (DAB) were tested on the LFB and the TMB resulted the most suitable for LFB applications. The study comprised two steps as follows: 1) capturing of the HRP–GNPs dual-labeled conjugate on the test zone (insensitive); 2) signal amplification by applying the HRP substrates on the test zone (sensitive). After signal amplification, the LFB gained sensitivity (up to 1 order of magnitude) without losing the simplicity and the detection reproducibility.

Recently, new advances have been made to develop ultrasensitive analyte (DNA) tests by introducing the signal amplification strategy-isothermal strand-displacement polymerase reactions (ISDPR) into the LFB bioassay. ISDPR have been developed to overcome the disadvantages of PCR. No thermal cycling is required in the ISDPR, offering distinct advantages with regard to the cost and simplicity

of instrumentation. Moreover, ISDPR permits 10^{10}-fold amplification of a DNA target sequence within 15 min, and the amplified DNA can be used and detected directly without any further purification [88,89]. A LFB combined with isothermal strand-displacement technology was proposed by Lie and co-workers for the detection of human genomic DNA in the aqoeous solutions. The ISDPR solution consisted of a biotin modified hairpin DNA probe, a strand displacement primer, target DNA probe, Klenow polymerase exo-, dNTPs and buffer solution[90]. A 41-mer DNA hairpin probe (containing the target DNA binding site; probe 1) is first immobilized on the surface of GNPs. In the present of the target DNA, it would bind on the specific binding site on the hairpin probe, and then open the hairpin probe forming the duplexes on the surface of GNPs. After introducing the primer and the tag (probe 2 and probe 3), the recognition and hybridization with probe 1 occurred. At the same time, probe 1 was undergoing a conformational change and leading to stem separation. In the presence of Klenow polymerase exo-, probe 2 would anneal with the open stem and trigger an extension reaction. During the extension reaction, the target DNA was displaced, which triggered a new round of polymerization reaction. As a result, numerous sequence tagged-duplex DNA complexes were produced throughout this repeating process. In a typical assay, the resulting complexes would be captured on the test zone of the test strip through the binding affinity between the biotin (labeled on the probe 1) and the streptavidin (SA) (immobilized on the test zone). The detection limit of the modified LFS was 0.01 fM of target DNA, which was 1000 times better than published traditional LFS methods. He et al. [91] also reported an ISDPR based LFB for visual detection of R156H-mutant DNA. The detection limit was 1-fM R156H-mutant DNA within 75 min without instrumentation. The specificity was further demonstrated by showing no cross-reactivity to distinguish of R156H- and R156C-mutant DNA on the R156 mutation site through the immobilizing of fluorescein- and biotin-modified hairpin probes in the ISDPR process. And the resulting specificity of the assay was 100%.

An interesting attempt was carried out recently by Choi and co-workers [92] upon the applying the traditional sandwich format-based LFB with the 2nd conjugate by immobilizing GNPs with different size on the different antibody, and the antibody was designed to bind specifically with the 1st GNP conjugate. In a typical assay, after the 1st GNPs conjugate was captured on the LFB, the 2nd GNP conjugate would bind on the 1st GNPs to amplify the optical signal by depositing more GNPs on the test zone. In this study, the 1st conjugate was an anti-troponin I antibody immobilized on the 10-nm GNPs blocking with bovine serum albumin (BSA), and the 2nd conjugate was an anti-BSA antibody immobilized on the 40-nm GNPs blocking with human serum albumin. The detection limit of this dual GNP conjugates-based LFB was as low as 0.01 ng / mL troponin I within 10 min, whereas 1 ng / mL was detectable by the conventional LFB method, offering a 100-fold signal amplification method than the conventional LFB.

4. Applications of new nanoparticle labels

The visible color from the test zone and control zone is generated from the integration and accumulation of the colored detector reagents. Therefore, for successful development of LFB with sufficient sensitivity, the type of colored detector reagents is very important. As mentioned above, the GNPs are the most widely used colored reagent, owing to its notable advantages. However, GNPs still suffer from some drawbacks: the sensitivity is insufficient for low abundance of targets; the signal amplification process could be costly. Therefore, it is significant to finding the suitable and new detector reagents for sensitive detection.

Magnetic nanoparticles and microparticles are attractive as nanomaterials because of their embedded magnetic entities, larger surface-to-volume ratio and they can be magnetically manipulated by using permanent magnets or electromagnets. By considering these advantages, Tang and co-workers [93] synthetised the magnetic nano-gold microspheres (MnGMs; with

nano-Fe_2O_3 particles as core and GNPs as shell) as the colored reagent and employed them for detecting aflatoxin B2 (AFB2) in food samples. The synthesis of the MnGMs comprised six steps: 1) the synthesis of the SDS–Fe_2O_3 complex; 2) immobilizing chitosan on the surface of Fe_2O_3 nanoparticles; 3) the synthesis of the chitosan/Fe_2O_3 nanocomposites; 4) cross-linking glutaraldehyde with chitosan, 5) assembling GNPs on the surface of the formed nanocomposites, 6) obtaining MnGMs by magnetic separation. In this typical competitive immuno-reaction, the test zone and control zone were immobilized with BSA-AFB2 and anti-mouse IgG, respectively. In the absence of target AFB2 (or lower than the cutoff value), two red lines can be observed from both of the test zone and control zone. In the presence of target AFB2, only one red line can be observed from the control zone. The detection limit of the LFB was as low as 0.9 ng / ml AFB2 without instrument, which was threefold lower compared to a conventional LFB test using gold nanoparticles as colored detector reagents. Moreover, results of the parallel analysis of AFB2 in blank peanut samples and in naturally contaminated samples showed good agreement rate between HPLC method and the LFIBA. Another gold magnetic nanoparticles (GMNs) based LFB was developed for treponema pallidum antibodies (Tp) detection [94]. In this study, a PAA (poly (acrylic acid))-coated GMNs (PGMNs) was prepared with recombinant Treponema pallidum antigens (r-Tp) as the detection reagent. The preparation of detection reagent included the preparation of the pure core/shell of Fe_3O_4/GNPs, coating PAA on the surface of GMNs and immobilizing the targeted moieties on the PGMNs. The purpose of coating PAA on the surface of GMNs is that PGMNs are more stable and monodispersed than GMNs. The PGMNs were further investigated and characterized by fourier transform-infrared spectroscopy (FT-IR), transmission electron microscopy, UV−visible scanning spectrophotometry, thermogravimetric analysis, and Zetasizer methodologies. The detection limit of this method was as low as 1 national clinical unit/mL (NCU/mL). The specificity of the PGMNs-based LFIA strips were determined by testing 1020 sera samples from three independent hospitals, exhibiting high values of the specificity for all clinical tests (> 97%).

An immuno-nanoparticle composed of a silver core and a gold shell (Ag/Au) was synthesized by Liao et al. [95] and applied for the fast screening of aflatoxin B_1 (AFB_1) in food samples. The synthesis of the core-shell Ag/Au nanocomposites comprised four steps: 1) The synthesis of Ag NPs; 2) the reduction of $HAuCl_4$ by introducing $NaBH_4$ solution; 3) the coverage of the Au^0 on the surface of Ag surface. In this study, the Ag/Au nanocomparticle was immobilized with monoclonal anti-AFB_1 primary antibodies, and the resulting conjugates were used as detection reagent in the lateral flow strip test. The detection limit (cutoff value) of the AFB_1 was observed at 0.1 ng / mL within 15 min. High sensitivity was also achieved when a blue dye doped latex beads were used as the colored reagent in the LFB for sensitive DNA detection in aqueous solutions and plasma [96]. The strategy was based on the sandwich format using the target DNA and a pair of DNA probes (capture probe and detection probe). The blue dye doped latex beads was immobilized on the detection probe as the detection reagent, and the capture probe was immobilized on the test zone as the capture reagent. The detection limit was 0.1 nM in aqueous solutions and 3.75 fmol in 50 μL of human plasma.

A carbon nanoparticle based competitive LFB was reported by Sua´rez-Pantaleo´n and co-workers [97] for the detection synthetic phytoregulator forchlorfenuron (CPPU). The detection limit of the carbon nanoparticle based LFB was 89 ng / L, and the results of the parallel analysis of fruit samples with incurred residues showed good agreement rate of two reference methods (ELISA and HPLC).

In addition, the use of fluorescent technique [98-105], up-converting phosphorus [106,107,59,74] and technology also allowed an increase in sensitivity and simplicity compared with that for visual detection of traditional colored labels.

5. New quantitative system

Quantity system is significant for the target analysis, because suitable quantity system can avoid the loss of signal which should also mean a consequent increase of assay sensitivity. Recently, Qin and co-workers [108] introduced the concept of thermal contrast into the LFB based bioassay to improve detection sensitivity. After addition of the targets, the captured GNP-antibody-antigen complex on the test zone enabled the visual detection of the target antigen. At low concentrations of antigen, less GNPs was captured on the test zone, resulting in insufficient bound GNPs for visual detection. A laser or light-emitting diode (LED) and an infrared temperature gun were used to irradiate the GNPs on the test zone and control zone. The thermal contrast can extend the analytical sensitivity of the LFB by 32-fold. They further modification of the assay by combination of higher-absorbing nanoparticles and low-absorbing LFB backing materials. As a result, a 1000-fold improvement in sensitivity was obtained. The use of thermal contrast is a promising novel detection strategy for signal amplification of the LFB bioassay.

Li et al. [109] also introduced a high power LED (520-540 nm) into the LFB for detection of clenbuterol (CLE) in swine urine. The LED (520-540 nm) was used as light emitter to irradiate the captured GNPs. The reflective optical signal from GNPs was filter and converted to photoelectrical signals. Because GNPs have an affinity for the absorbance in the green region, therefore, the more GNPs were capture on the test strip, the less photoelectrical responses can be obtained. The detection limit of the CLE by the new quantitative system based LFB was 220 pg / mL.

6. Simultaneous detection

Recently, research in the laboratory was focused on developing new LFBs for simultaneous detection [110-113]. Compared with the single detection, simultaneous detection offers many advantages such as simple, timesaving, sample saving, and no need for checking the suspected parameters [114,115,71]. A combination of colloidal gold nanoparticles and oligonucleotide LFB was described by Yuichi Oku et al. [116] for the simultaneous detection of antigens (HBs antigen) and antibodies (Treponema pallidum, TP) in specimens. The assay was based on sandwich format. Four labeled antigens were used in the study: GNPs-labeled anti-antigen A, oligonucleotide 1-labeled anti-antigen A, GNPs-labeled antigen B and oligonucleotide 1-labeled antigen B were firstly applied on the application pad. After introducing the sample solution (containing antigen A and antibody B) onto the sample application pad, specific bindings including antigen A/anti-antigen A and antibody B/ antigen B occurred. At the same time, two sandwich typed complexes: oligonucleotide 1/antigen A/GNPs and oligonucleotide 1/antibody B/GNPs were formed on the LFB. Subsequently, the solution containing two complexes continued to migrate along the strip. When they reached the two test zone, the two complexes were captured by the complementary oligonucleotide 1 and the complementary oligonucleotide 2 via the DNA–DNA interaction, respectively. Therefore, in the presence of the two target analytes in a specimen, two characteristic red bands can be observed from the two test zones. In contrary, in the absence of target analyte, no red band can be observed. As a result, more than two types of reactions can be detected on a single assay device. The detection limit was 1 / 8 mg / ml for the TP sensing and 5 ng / ml for the HBs sensing.

Zhu et al. [115] detected cardiac troponin I (hs-cTnl) and myoglobin simultaneously to exclude acute myocardial infarction (AMI) via GNPs based LFB. In this study, the specimens of cardiac troponin I (hs-cTnl) and myoglobin were collected from 173 patients with AMI symptoms. The assay was then compared with the commercial LFIA test, and the agreement rates between the two methods were 100%. In the typical assay, two conjugate pads were introduced to the LFB. In the second conjugate pad, monoclonal anti-hs-cTnI detecting antibody (anti-hs-cTnI mAb1) were labeled with GNPs (13 nm), and then coated with biotinylated DNA. Meanwhile, the monoclonal anti-myoglobin detecting antibody (anti-myoglobin mAb1) were

also labeled with GNPs (13 nm). Another GNPs with size of 41 nm was coated with streptavidin and then bound with anti-hs-cTnImAb1-labeled GNPs (13 nm) on the second conjugate pad to block the first conjugate pad as an intensifier. After the modified LFB test for the simultaneous detection of cardiac troponin I (hs-cTnl) and myoglobin, two characteristic red bands can be observed from the two test zones. The sensitivity of the modified LFB and the commercial LFB were 100% and 80%, respectively, by studying 20 samples, and the both specificities were 100%. A lateral flow strip for simultaneous detection of cyromazine (CA) and melamine (MA) in foods of animal origin was developed by Le et al. [111]. The assay was based on a competitive binding bioassay using GNPs as label. In a typical LFB assay, anti-CA mAbs and GNPs-coated anti-MA mAbs were firstly added on the conjugate pad. The two test zones and control zone were respectively dispensed with CA–BSA, MA–BSA and goat anti-mouse IgG. In the absence of target analyte, the CG-mAb 2.2.4 / 6A11 or CG-mAb 4.1.4 / 5D8 would be captured by the two test zones which were dispensed with CA–BSA and MA–BSA. In the presence of target analyte (concentration more than 25 ng / ml), no red band can be observed in the corresponding position. A sensitivities limit at 0.22 ng / ml and 0.26 ng / ml were obtained from CA and MA in matrix sample, respectively. The recoveries for CA and MA at three concentration levels (50, 100, and 150 ng / g) were ranged from 73.9% to 104.2%. Using HPLC as confirmatory method, the validation of the ICA test was achieved by analyzing CA and MA in real samples (muscle, liver of swine, cattle and sheep, and milk). The agreement rates between LFBs and HPLC were 100%.

Kolosova et al. [110] developed a multi-analyte lateral-flow technique using GNPs-labeled monoclonal antibodies for the simultaneous detection of deoxynivalenol (DON) and zearalenone (ZEA). The sensitivities of the tests were estimated to be 1500 μL / kg and 100 μL / kg for DON and ZEA, respectively, and the detection time was less than 10 min. Results from these studies have proved the success of test strip applications.

A combination of GNPs based LFB and logic gates ("OR" and "AND" functions) for proteins and small molecules was described by Chen and co-workers [117]. In this study, thrombin and ATP were used as the inputs, split/integrated aptamers were used as the molecular recognition elements, and the GNPs were used as a tracer. After a complete assay, the output signals could be observed from the color change of the test zone. For the "OR" gate detection, in the absence of any inputs (0,0), no color change can be observed from the test zone; in the presence of either (1,0; 0,1) or both inputs (1,1), color change can be observed from the test zone. For the "AND" gate detection, in the presence of both inputs (1,1), color change can be observed from the test zone; in the absence of any (0,0) or either inputs (1,0; 0,1), no color change can be observed from the test zone. This is the first application of logic gates in LFB detection, opening the new possibility for the analyte analysis in bioassays.

7. Conclusions and outlook

This review has summarized the recent progress on LFBs. Major advantages found on LFBs are portable, inexpensive, simple, robust, short assay time (generally several minutes), and do not require complicated equipment and skilled technicians. The unique and remarkable properties of LFBs have paved the way and opened the possibility for the detecting disease biomarkers, infectious agents and biothreat agents in medicine, agriculture, food and environmental safety. The studies described above demonstrate the recent progress of LFB in improving sensitivity including the signal-amplification strategies and new nanoparticle labels for achieving lower detection limits. In addition, the LFBs are promising in simultaneous detection of several analytes in contaminated or hazardous samples effectively outside the laboratory. Furthermore, recent breakthroughs in research in the LFB bioassays could also encourage more innovation in the design of novel quantitative system, which would overcome the loss of signal and extend the analytical sensitivity. However, the challenges for current research are 1) "How will the LFB technologies work without

complicated sample pretreatment?" and 2) "Will it be possible to achieve low volume detection in LFB analysis?" Therefore, future innovative research is expected to lead to advanced LFB by simplicity and practicability improvement, including sample pad pretreatment strategy and introducing with other major technological advances, such as microfluidic technology.

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References

1. Chen C, Ridzon DA, Broomer AJ, Zhou Z, Lee DH, Nguyen JT, Barbisin M, Xu NL, Mahuvakar VR, Andersen MR. Real-time quantification of microRNAs by stem–loop RT–PCR. *Nucleic Acids Research* 2005; 33 (20):e179-e179. DOI: 10.1093/nar/gni178
2. Pfaffl MW. A new mathematical model for relative quantification in real-time RT–PCR. *Nucleic Acids Research* 2001; 29 (9):e45-e45. DOI: 10.1093/nar/29.9.e45
3. Schmittgen TD, Livak KJ. Analyzing real-time PCR data by the comparative CT method. *Nature protocols* 2008; 3 (6):1101-1108. DOI: 10.1038/nprot.2008.73
4. Vandesompele J, De Preter K, Pattyn F, Poppe B, Van Roy N, De Paepe A, Speleman F. Accurate normalization of real-time quantitative RT-PCR data by geometric averaging of multiple internal control genes. *Genome biology* 2002; 3 (7):research0034.
5. Barad O, Meiri E, Avniel A, Aharonov R, Barzilai A, Bentwich I, Einav U, Gilad S, Hurban P, Karov Y. MicroRNA expression detected by oligonucleotide microarrays: system establishment and expression profiling in human tissues. *Genome research* 2004; 14 (12):2486-2494. DOI: 10.1101/gr.2845604
6. Kerr MK, Martin M, Churchill GA. Analysis of variance for gene expression microarray data. *Journal of computational biology* 2000; 7 (6):819-837. DOI: 10.1089/10665270050514954
7. Yang YH, Dudoit S, Luu P, Lin DM, Peng V, Ngai J, Speed TP. Normalization for cDNA microarray data: a robust composite method addressing single and multiple slide systematic variation. *Nucleic Acids Research* 2002; 30 (4):e15-e15. DOI: 10.1093/nar/30.4.e15
8. Yin JQ, Zhao RC, Morris KV. Profiling microRNA expression with microarrays. *Trends in biotechnology* 2008; 26 (2):70-76. DOI: 10.1016/j.tibtech.2007.11.007
9. Elshal MF, McCoy JP. Multiplex bead array assays: performance evaluation and comparison of sensitivity to ELISA. *Methods* 2006; 38 (4):317-323. DOI: 10.1016/j.ymeth.2005.11.010
10. Eteshola E, Leckband D. Development and characterization of an ELISA assay in PDMS microfluidic channels. *Sensors and Actuators B: Chemical* 2001; 72 (2):129-133. DOI: 10.1016/S0925-4005(00)00640-7
11. Sblattero D, Berti I, Trevisiol C, Marzari R, Tommasini A, Bradbury A, Fasano A, Ventura A, Not T. Human recombinant tissue transglutaminase ELISA: an innovative diagnostic assay for celiac disease. *The American journal of gastroenterology* 2000; 95 (5):1253-1257. DOI: 10.1111/j.1572-0241.2000.02018.x
12. Sehr P, Zumbach K, Pawlita M. A generic capture ELISA for recombinant proteins fused to glutathione< i> S</i>-transferase: validation for HPV serology. *Journal of immunological methods* 2001; 253 (1):153-162. DOI: 10.1016/S0022-1759(01)00376-3
13. Vanmechelen E, Vanderstichele H, Davidsson P, Van Kerschaver E, Van Der Perre B, Sjögren M, Andreasen N, Blennow K. Quantification of tau phosphorylated at threonine 181 in human cerebrospinal fluid: a sandwich ELISA with a synthetic

phosphopeptide for standardization. *Neuroscience letters* 2000; 285 (1):49-52. DOI: 10.1016/S0304-3940(00)01036-3

14. Chandra A, Rana J, Li Y. Separation, identification, quantification, and method validation of anthocyanins in botanical supplement raw materials by HPLC and HPLC-MS. *Journal of agricultural and food chemistry* 2001; 49 (8):3515-3521. DOI: 10.1021/jf010389p
15. Gika HG, Theodoridis GA, Wingate JE, Wilson ID. Within-day reproducibility of an HPLC-MS-based method for metabonomic analysis: application to human urine. *Journal of proteome research* 2007; 6 (8):3291-3303. DOI: 10.1021/pr070183p
16. Wilson ID, Plumb R, Granger J, Major H, Williams R, Lenz EM. HPLC-MS-based methods for the study of metabonomics. *Journal of Chromatography B* 2005; 817 (1):67-76. DOI: 10.1016/j.jchromb.2004.07.045
17. Ye X, Kuklenyik Z, Needham LL, Calafat AM. Automated on-line column-switching HPLC-MS/MS method with peak focusing for the determination of nine environmental phenols in urine. *Analytical Chemistry* 2005; 77 (16):5407-5413. DOI: 10.1021/ac050390d
18. Baxter G, Ferguson J, O'Conno M, Elliott C. Detection of streptomycin residues in whole milk using an optical immunobiosensor. *Journal of agricultural and food chemistry* 2001; 49 (7):3204-3207. DOI: 10.1021/jf001484l
19. Huang K-J, Niu D-J, Sun J-Y, Zhu X-L, Zhu J-J. Label-free amperometric immunobiosensor based on a gold colloid and Prussian blue nanocomposite film modified carbon ionic liquid electrode. *Analytical and bioanalytical chemistry* 2010; 397 (8):3553-3561. DOI: 10.1007/s00216-010-3868-4
20. Johansson MA, Hellenäs K-E. Matrix effects in immunobiosensor determination of clenbuterol in urine and serum. *Analyst* 2004; 129 (5):438-442. DOI: 10.1039/b316723b
21. Johnsson L, Baxter GA, Crooks SR, Brandon DL, Elliott CT. Reduction of sample matrix effects-The analysis of benzimidazole residues in serum by immunobiosensor. *Food and agricultural immunology* 2002; 14 (3):209-216. DOI: 10.1080/09540100220145000a
22. Marquette CA, Blum LcJ. Regenerable immunobiosensor for the chemiluminescent flow injection analysis of the herbicide 2, 4-D. *Talanta* 2000; 51 (2):395-401. DOI: 10.1016/S0039-9140(99)00298-2
23. Traynor IM, Plumpton L, Fodey TL, Higgins C, Elliott CT. Immunobiosensor detection of domoic acid as a screening test in bivalve molluscs: comparison with liquid chromatography-based analysis. *Journal of AOAC International* 2006; 89 (3):868-872.
24. Bührer-Sékula S, Smits HL, Gussenhoven GC, Van Leeuwen J, Amador S, Fujiwara T, Klatser PR, Oskam L. Simple and fast lateral flow test for classification of leprosy patients and identification of contacts with high risk of developing leprosy. *Journal of Clinical Microbiology* 2003; 41 (5):1991-1995. DOI: 10.1128/JCM.41.5.1991-1995.2003
25. Delmulle BS, De Saeger SM, Sibanda L, Barna-Vetro I, Van Peteghem CH. Development of an immunoassay-based lateral flow dipstick for the rapid detection of aflatoxin B1 in pig feed. *Journal of agricultural and food chemistry* 2005; 53 (9):3364-3368. DOI: 10.1021/jf0404804
26. Fenton EM, Mascarenas MR, López GP, Sibbett SS. Multiplex lateral-flow test strips fabricated by two-dimensional shaping. *ACS Applied Materials & Interfaces* 2008; 1 (1):124-129. DOI: 10.1021/am800043z
27. He Y, Zhang X, Zeng K, Zhang S, Baloda M, Gurung AS, Liu G. Visual detection of Hg< sup> 2+</sup> in aqueous solution using gold nanoparticles and thymine-rich hairpin DNA probes. *Biosensors and Bioelectronics* 2011; 26 (11):4464-4470. DOI: 10.1016/j.bios.2011.05.003
28. Liu G, Mao X, Phillips JA, Xu H, Tan W, Zeng L. Aptamer− Nanoparticle Strip Biosensor for Sensitive Detection of Cancer

Cells. *Analytical Chemistry* 2009; 81 (24):10013-10018. DOI: 10.1021/ac901889s
29. Liu J, Mazumdar D, Lu Y. A Simple and Sensitive "Dipstick" Test in Serum Based on Lateral Flow Separation of Aptamer‐Linked Nanostructures. *Angewandte Chemie* 2006; 118 (47):8123-8127. DOI: 10.1002/ange.200603106
30. Mao X, Ma Y, Zhang A, Zhang L, Zeng L, Liu G. Disposable nucleic acid biosensors based on gold nanoparticle probes and lateral flow strip. *Analytical Chemistry* 2009; 81 (4):1660-1668. DOI: 10.1021/ac8024653
31. Wozniak RA, Waldor MK. Integrative and conjugative elements: mosaic mobile genetic elements enabling dynamic lateral gene flow. *Nature Reviews Microbiology* 2010; 8 (8):552-563. DOI: 10.1038/nrmicro2382
32. Xia X, Xu Y, Zhao X, Li Q. Lateral flow immunoassay using europium chelate-loaded silica nanoparticles as labels. *Clinical chemistry* 2009; 55 (1):179-182. DOI: 10.1373/clinchem.2008.114561
33. Xu H, Mao X, Zeng Q, Wang S, Kawde A-N, Liu G. Aptamer-functionalized gold nanoparticles as probes in a dry-reagent strip biosensor for protein analysis. *Analytical Chemistry* 2008; 81 (2):669-675. DOI: 10.1021/ac8020592
34. Leuvering JH. Metal sol particle immunoassay. *Google Patents* 1982.
35. Leuvering JH, Thal P, Waart Mvd, Schuurs A. Sol particle immunoassay (SPIA). *Journal of immunoassay* 1980; 1 (1):77-91. DOI: 10.1080/01971528008055777
36. Carter DJ, Cary RB. Lateral flow microarrays: a novel platform for rapid nucleic acid detection based on miniaturized lateral flow chromatography. *Nucleic Acids Research* 2007; 35 (10):e74. DOI: 10.1093/nar/gkm269
37. Cheek BJ, Steel AB, Torres MP, Yu Y-Y, Yang H. Chemiluminescence detection for hybridization assays on the flow-thru chip, a three-dimensional microchannel biochip. *Analytical Chemistry* 2001; 73 (24):5777-5783. DOI: 10.1021/ac0108616
38. Fisher M, Atiya‐Nasagi Y, Simon I, Gordin M, Mechaly A, Yitzhaki S. A combined immunomagnetic separation and lateral flow method for a sensitive on‐site detection of Bacillus anthracis spores–assessment in water and dairy products. *Letters in applied microbiology* 2009; 48 (4):413-418. DOI: 10.1111/j.1472-765X.2008.02542.x
39. Jung BY, Jung SC, Kweon CH. Development of a rapid immunochromatographic strip for detection of Escherichia coli O157. *Journal of Food Protection*® 2005; 68 (10):2140-2143.
40. Nakasone N, Toma C, Lu Y, Iwanaga M. Development of a rapid immunochromatographic test to identify enteropathogenic and enterohemorrhagic< i> Escherichia coli</i> by detecting EspB. *Diagnostic microbiology and infectious disease* 2007; 57 (1):21-25. DOI: 10.1016/j.diagmicrobio.2006.05.012
41. Ngom B, Guo Y, Wang X, Bi D. Development and application of lateral flow test strip technology for detection of infectious agents and chemical contaminants: a review. *Analytical and bioanalytical chemistry* 2010; 397 (3):1113-1135. DOI: 10.1007/s00216-010-3661-4
42. He Y, Zhang S, Zhang X, Baloda M, Gurung AS, Xu H, Zhang X, Liu G. Ultrasensitive nucleic acid biosensor based on enzyme–gold nanoparticle dual label and lateral flow strip biosensor. *Biosensors and Bioelectronics* 2011; 26 (5):2018-2024. DOI: 10.1016/j.bios.2010.08.079
43. Mao X, Xu H, Zeng Q, Zeng L, Liu G. Molecular beacon-functionalized gold nanoparticles as probes in dry-reagent strip biosensor for DNA analysis. *Chemical Communications* 2009; (21):3065-3067. DOI: 10.1039/b822582f
44. Liu G, Lin Y-Y, Wang J, Wu H, Wai CM, Lin Y. Disposable electrochemical immunosensor diagnosis device based on nanoparticle probe and immunochromatographic strip. *Analytical*

Chemistry 2007; 79 (20):7644-7653. DOI: 10.1021/ac070691i

45. Huo T, Peng C, Xu C, Liu L. Immumochromatographic assay for determination of hexoestrol residues. *European Food Research and Technology* 2007; 225 (5-6):743-747. DOI: 10.1007/s00217-006-0477-8
46. Liu L, Peng C, Jin Z, Xu C. Development and evaluation of a rapid lateral flow immunochromatographic strip assay for screening 19 - nortestosterone. *Biomedical chromatography* 2007; 21 (8):861-866. DOI: 10.1002/bmc.832
47. Posthuma-Trumpie GA, Korf J, van Amerongen A. Development of a competitive lateral flow immunoassay for progesterone: influence of coating conjugates and buffer components. *Analytical and bioanalytical chemistry* 2008; 392 (6):1215-1223. DOI: 10.1007/s00216-008-2362-8
48. Qian S, Bau HH. Analysis of lateral flow biodetectors: competitive format. *Analytical biochemistry* 2004; 326 (2):211-224. DOI: 10.1016/j.ab.2003.12.019
49. Wang X, Li K, Shi D, Xiong N, Jin X, Yi J, Bi D. Development of an immunochromatographic lateral-flow test strip for rapid detection of sulfonamides in eggs and chicken muscles. *Journal of agricultural and food chemistry* 2007; 55 (6):2072-2078. DOI: 10.1021/jf062523h
50. Zhang G, Wang X, Yang J, Yang Y, Xing G, Li Q, Zhao D, Chai S, Guo J. Development of an immunochromatographic lateral flow test strip for detection of β-adrenergic agonist clenbuterol residues. *Journal of immunological methods* 2006; 312 (1):27-33. DOI: 10.1016/j.jim.2006.02.017
51. Cha YJ, Kwon SY, Kim TY, Kim JR, Kim HS, Park MH, Park SH, Park AJ, Bai JH, Son HC. Annual Report on External Quality Assessment in Immunoserology in Korea (2009). *Journal of Laboratory Medicine and Quality Assurance* 2010; 32 (1):45-68.
52. Kawatsu K, Kumeda Y, Taguchi M, Yamazaki-Matsune W, Kanki M, Inoue K. Development and evaluation of immunochromatographic assay for simple and rapid detection of Campylobacter jejuni and Campylobacter coli in human stool specimens. *Journal of Clinical Microbiology* 2008; 46 (4):1226-1231. DOI: 10.1128/JCM.02170-07
53. Khreich N, Lamourette P, Boutal H, Devilliers K, Créminon C, Volland H. Detection of< i> Staphylococcus</i> enterotoxin B using fluorescent immunoliposomes as label for immunochromatographic testing. *Analytical biochemistry* 2008; 377 (2):182-188. DOI: 10.1016/j.ab.2008.02.032
54. Klewitz T, Gessler F, Beer H, Pflanz K, Scheper T. Immunochromatographic assay for determination of botulinum neurotoxin type D. *Sensors and Actuators B: Chemical* 2006; 113 (2):582-589. DOI: 10.1016/j.snb.2005.07.007
55. Lyoo Y, Kleiboeker S, Jang K-Y, Shin N, Kang J-M, Kim C-H, Lee S-J, Sur J-H. A simple and rapid chromatographic strip test for detection of antibody to porcine reproductive and respiratory syndrome virus. *Journal of veterinary diagnostic investigation* 2005; 17 (5):469-473. DOI: 10.1177/104063870501700512
56. Yan Z, Zhou L, Zhao Y, Wang J, Huang L, Hu K, Liu H, Wang H, Guo Z, Song Y. Rapid quantitative detection of< i> Yersinia pestis</i> by lateral-flow immunoassay and up-converting phosphor technology-based biosensor. *Sensors and Actuators B: Chemical* 2006; 119 (2):656-663. DOI: 10.1016/j.snb.2006.01.029
57. Guo Y-R, Liu S-Y, Gui W-J, Zhu G-N. Gold immunochromatographic assay for simultaneous detection of carbofuran and triazophos in water samples. *Analytical biochemistry* 2009; 389 (1):32-39. DOI: 10.1016/j.ab.2009.03.020
58. Kranthi K, Davis M, Mayee C, Russell D, Shukla R, Satija U, Kshirsagar M, Shiware D, Kranthi S. Development of a colloidal-gold based lateral-flow immunoassay kit for 'quality-control'assessment of pyrethroid and endosulfan formulations in a novel single strip format. *Crop protection* 2009;

28 (5):428-434. DOI: 10.1016/j.cropro.2009.01.003
59. Hampl J, Hall M, Mufti NA, Yao Y-mM, MacQueen DB, Wright WH, Cooper DE. Upconverting phosphor reporters in immunochromatographic assays. *Analytical biochemistry* 2001; 288 (2):176-187. DOI: 10.1006/abio.2000.4902
60. Takanashi S, Okame M, Shiota T, Takagi M, Yagyu F, Tung PG, Nishimura S, Katsumata N, Igarashi T, Okitsu S. Development of a rapid immunochromatographic test for noroviruses genogroups I and II. *Journal of virological methods* 2008; 148 (1):1-8. DOI: 10.1016/j.jviromet.2007.10.010
61. Laitinen MP, Vuento M. Immunochromatographic assay for quantitation of milk progesterone. *Acta Chemica Scandinavica* 1996; 50 (2):141-145. DOI: 10.3891/acta.chem.scand.50-0141
62. O'Keeffe M, Crabbe P, Salden M, Wichers J, Van Peteghem C, Kohen F, Pieraccini G, Moneti G. Preliminary evaluation of a lateral flow immunoassay device for screening urine samples for the presence of sulphamethazine. *Journal of immunological methods* 2003; 278 (1):117-126. DOI: 10.1016/S0022-1759(03)00207-2
63. Anfossi L, Baggiani C, Giovannoli C, D'Arco G, Giraudi G. Lateral-flow immunoassays for mycotoxins and phycotoxins: a review. *Analytical and bioanalytical chemistry* 2013; 405 (2-3):467-480. DOI: 10.1007/s00216-012-6033-4
64. Bamrungsap S, Apiwat C, Chantima W, Dharakul T, Wiriyachaiporn N. Rapid and sensitive lateral flow immunoassay for influenza antigen using fluorescently-doped silica nanoparticles. *Microchimica Acta* 2014; 181 (1-2):223-230. DOI: 10.1007/s00604-013-1106-4
65. Drygin YF, Blintsov AN, Grigorenko VG, Andreeva IP, Osipov AP, Varitzev YA, Uskov AI, Kravchenko DV, Atabekov JG. Highly sensitive field test lateral flow immunodiagnostics of PVX infection. *Applied microbiology and biotechnology* 2012; 93 (1):179-189. DOI: 10.1007/s00253-011-3522-x
66. Hou S-Y, Hsiao Y-L, Lin M-S, Yen C-C, Chang C-S. MicroRNA detection using lateral flow nucleic acid strips with gold nanoparticles. *Talanta* 2012; 99:375-379. DOI: 10.1016/j.talanta.2012.05.067
67. Jin W, Yamada K, Ikami M, Kaji N, Tokeshi M, Atsumi Y, Mizutani M, Murai A, Okamoto A, Namikawa T. Application of IgY to sandwich enzyme-linked immunosorbent assays, lateral flow devices, and immunopillar chips for detecting staphylococcal enterotoxins in milk and dairy products. *Journal of microbiological methods* 2013; 92 (3):323-331. DOI: 10.1016/j.mimet.2013.01.001
68. Parolo C, de la Escosura-Muñiz A, Merkoçi A. Enhanced lateral flow immunoassay using gold nanoparticles loaded with enzymes. *Biosensors and Bioelectronics* 2013; 40 (1):412-416. DOI: 10.1016/j.bios.2012.06.049
69. Pöhlmann C, Dieser I, Sprinzl M. A lateral flow assay for identification of Escherichia coli by ribosomal RNA hybridisation. *Analyst* 2014; 139 (5):1063-1071. DOI: 10.1039/c3an02059b
70. Xiang T, Jiang Z, Zheng J, Lo C, Tsou H, Ren G, Zhang J, Huang A, Lai G. A novel double antibody sandwich-lateral flow immunoassay for the rapid and simple detection of hepatitis C virus. *International journal of molecular medicine* 2012; 30 (5):1041.
71. Zhu J, Zou N, Zhu D, Wang J, Jin Q, Zhao J, Mao H. Simultaneous Detection of High-Sensitivity Cardiac Troponin I and Myoglobin by Modified Sandwich Lateral Flow Immunoassay: Proof of Principle. *Clinical chemistry* 2011; 57 (12):1732-1738. DOI: 10.1373/clinchem.2011.171694
72. Karakus C, Salih BA. Comparison of the lateral flow immunoassays (LFIA) for the diagnosis of< i> Helicobacter pylori</i> infection. *Journal of immunological methods* 2013; 396 (1):8-14. DOI: 10.1016/j.jim.2013.08.010

73. Posthuma-Trumpie GA, Korf J, van Amerongen A. Lateral flow (immuno) assay: its strengths, weaknesses, opportunities and threats. A literature survey. *Analytical and bioanalytical chemistry* 2009; 393 (2):569-582. DOI: 10.1007/s00216-008-2287-2
74. Tjon Kon Fat EM, Abrams WR, Niedbala RS, Corstjens PL. Lateral Flow Sandwich Assay Utilizing Upconverting Phosphor (UCP) Reporters. *Methods in Cell Biology* 2012; 112:203-234. DOI: 10.1016/B978-0-12-405914-6.00011-1
75. Anfossi L, Di Nardo F, Giovannoli C, Passini C, Baggiani C. Increased sensitivity of lateral flow immunoassay for ochratoxin A through silver enhancement. *Analytical and bioanalytical chemistry* 2013; 405 (30):9859-9867. DOI: 10.1007/s00216-013-7428-6
76. Lai W, Tang D, Que X, Zhuang J, Fu L, Chen G. Enzyme-catalyzed silver deposition on irregular-shaped gold nanoparticles for electrochemical immunoassay of alpha-fetoprotein. *Analytica chimica acta* 2012; 755:62-68. DOI: 10.1016/j.aca.2012.10.028
77. Liu C-C, Yeung C-Y, Chen P-H, Yeh M-K, Hou S-Y. < i> Salmonella</i> detection using 16S ribosomal DNA/RNA probe-gold nanoparticles and lateral flow immunoassay. *Food chemistry* 2013; 141 (3):2526-2532. DOI: 10.1016/j.foodchem.2013.05.089
78. Wang W, Wu W-Y, Zhong X, Miao Q, Zhu J-J. Aptamer-based PDMS–gold nanoparticle composite as a platform for visual detection of biomolecules with silver enhancement. *Biosensors and Bioelectronics* 2011; 26 (7):3110-3114. DOI: 10.1016/j.bios.2010.10.034
79. Wang Z, Duan N, Li J, Ye J, Ma S, Le G. Ultrasensitive chemiluminescent immunoassay of Salmonella with silver enhancement of nanogold labels. *Luminescence* 2011; 26 (2):136-141. DOI: 10.1002/bio.1196
80. Wen J, Zhou S, Yuan Y. Graphene oxide as nanogold carrier for ultrasensitive electrochemical immunoassay of< i> Shewanella oneidensis</i> with silver enhancement strategy. *Biosensors and Bioelectronics* 2014; 52:44-49. DOI: 10.1016/j.bios.2013.08.022
81. Yang W, Li X-b, Liu G-w, Zhang B-b, Zhang Y, Kong T, Tang J-j, Li D-n, Wang Z. A colloidal gold probe-based silver enhancement immunochromatographic assay for the rapid detection of abrin-a. *Biosensors and Bioelectronics* 2011; 26 (8):3710-3713. DOI: 10.1016/j.bios.2011.02.016
82. Fung K-K, Chan CP-Y, Renneberg R. Development of enzyme-based bar code-style lateral-flow assay for hydrogen peroxide determination. *Analytica chimica acta* 2009; 634 (1):89-95. DOI: 10.1016/j.aca.2008.11.064
83. Kiba Y, Otani Y, Yasukawa T, Mizutani F. Electrochemical detection of redox species flowing in a nitrocellulose membrane and application to quantitative immunochromatography. *Electrochimica Acta* 2012; 81:14-19. DOI: 10.1016/j.electacta.2012.07.074
84. Lönnberg M, Carlsson J. Quantitative detection in the attomole range for immunochromatographic tests by means of a flatbed scanner. *Analytical biochemistry* 2001; 293 (2):224-231. DOI: 10.1006/abio.2001.5130
85. TufanáÍz M. Nanoparticle embedded enzymes for improved lateral flow sensors. *Analyst* 2013; 138 (15):4255-4259. DOI: 10.1039/c3an00733b
86. Yamaguchi M, Matsuda Y, Sasaki S, Sasaki M, Kadoma Y, Imai Y, Niwa D, Shetty V. Immunosensor with fluid control mechanism for salivary cortisol analysis. *Biosensors and Bioelectronics* 2013; 41:186-191. DOI: 10.1016/j.bios.2012.08.016
87. Zhang C, Zhang Y, Wang S. Development of multianalyte flow-through and lateral-flow assays using gold particles and horseradish peroxidase as tracers for the rapid determination of carbaryl and endosulfan in agricultural products. *Journal of agricultural and food chemistry* 2006; 54 (7):2502-2507. DOI: 10.1021/jf0531407

88. Dong H, Zhang J, Ju H, Lu H, Wang S, Jin S, Hao K, Du H, Zhang X. Highly sensitive multiple microRNA detection based on fluorescence quenching of graphene oxide and isothermal strand-displacement polymerase reaction. *Analytical Chemistry* 2012; 84 (10):4587-4593. DOI: 10.1021/ac300721u
89. He Y, Zeng K, Zhang S, Gurung AS, Baloda M, Zhang X, Liu G. Visual detection of gene mutations based on isothermal strand-displacement polymerase reaction and lateral flow strip. *Biosensors and Bioelectronics* 2012; 31 (1):310-315. DOI: 10.1016/j.bios.2011.10.037
90. Lie P, Liu J, Fang Z, Dun B, Zeng L. A lateral flow biosensor for detection of nucleic acids with high sensitivity and selectivity. *Chemical Communications* 2012; 48 (2):236-238. DOI: 10.1039/c1cc15878c
91. He Y, Zeng K, Zhang X, Gurung AS, Baloda M, Xu H, Liu G. Ultrasensitive electrochemical detection of nucleic acid based on the isothermal strand-displacement polymerase reaction and enzyme dual amplification. *Electrochemistry Communications* 2010; 12 (7):985-988. DOI: 10.1016/j.elecom.2010.05.007
92. Choi DH, Lee SK, Oh YK, Bae BW, Lee SD, Kim S, Shin Y-B, Kim M-G. A dual gold nanoparticle conjugate-based lateral flow assay (LFA) method for the analysis of troponin I. *Biosensors and Bioelectronics* 2010; 25 (8):1999-2002. DOI: 10.1016/j.bios.2010.01.019
93. Tang D, Sauceda J, Lin Z, Ott S, Basova E, Goryacheva I, Biselli S, Lin J, Niessner R, Knopp D. Magnetic nanogold microspheres-based lateral-flow immunodipstick for rapid detection of aflatoxin B< sub> 2</sub> in food. *Biosensors and Bioelectronics* 2009; 25 (2):514-518. DOI: 10.1016/j.bios.2009.07.030
94. Yang D, Ma J, Zhang Q, Li N, Yang J, Raju PA, Peng M, Luo Y, Hui W, Chen C. Polyelectrolyte-Coated Gold Magnetic Nanoparticles for Immunoassay Development: Toward Point of Care Diagnostics for Syphilis Screening. *Analytical Chemistry* 2013; 85 (14):6688-6695. DOI: 10.1021/ac400517e
95. Liao J-Y, Li H. Lateral flow immunodipstick for visual detection of aflatoxin B1 in food using immuno-nanoparticles composed of a silver core and a gold shell. *Microchimica Acta* 2010; 171 (3-4):289-295. DOI: 10.1007/s00604-010-0431-0
96. Mao X, Wang W, Du TE. Dry-reagent nucleic acid biosensor based on blue dye doped latex beads and lateral flow strip. *Talanta* 2013; 114:248-253. DOI: 10.1016/j.talanta.2013.04.044
97. Suárez-Pantaleón C, Wichers J, Abad-Somovilla A, van Amerongen A, Abad-Fuentes A. Development of an immunochromatographic assay based on carbon nanoparticles for the determination of the phytoregulator forchlorfenuron. *Biosensors and Bioelectronics* 2013; 42:170-176. DOI: 10.1016/j.bios.2012.11.001
98. Berlina AN, Taranova NA, Zherdev AV, Vengerov YY, Dzantiev BB. Quantum dot-based lateral flow immunoassay for detection of chloramphenicol in milk. *Analytical and bioanalytical chemistry* 2013; 405 (14):4997-5000. DOI: 10.1007/s00216-013-6876-3
99. Chen R, Li H, Zhang H, Zhang S, Shi W, Shen J, Wang Z. Development of a lateral flow fluorescent microsphere immunoassay for the determination of sulfamethazine in milk. *Analytical and bioanalytical chemistry* 2013; 405 (21):6783-6789. DOI: 10.1007/s00216-013-7150-4
100. Dudek MM, Kent NJ, Gu P, Fan ZH, Killard AJ. Development of a fluorescent method for detecting the onset of coagulation in human plasma on microstructured lateral flow platforms. *Analyst* 2011; 136 (9):1816-1825. DOI: 10.1039/c0an00907e
101. Juntunen E, Myyryläinen T, Salminen T, Soukka T, Pettersson K. Performance of fluorescent europium (III) nanoparticles and colloidal gold reporters in lateral flow bioaffinity assay. *Analytical biochemistry*

2012; 428 (1):31-38. DOI: 10.1016/j.ab.2012.06.005
102. Leonardi GP, Wilson AM, Zuretti AR. Comparison of conventional lateral-flow assays and a new fluorescent immunoassay to detect influenza viruses. *Journal of virological methods* 2013; 189 (2):379-382. DOI: 10.1016/j.jviromet.2013.02.008
103. Li X, Lu D, Sheng Z, Chen K, Guo X, Jin M, Han H. A fast and sensitive immunoassay of avian influenza virus based on label-free quantum dot probe and lateral flow test strip. *Talanta* 2012; 100:1-6. DOI: 10.1016/j.talanta.2012.08.041
104. Nabatiyan A, Baumann MA, Parpia Z, Kelso D. A lateral flow-based ultra-sensitive p24 HIV assay utilizing fluorescent microparticles. *JAIDS Journal of Acquired Immune Deficiency Syndromes* 2010; 53 (1):55-61. DOI: 10.1097/QAI.0b013e3181c4b9d5
105. Wang Y, Nugen SR. Development of fluorescent nanoparticle-labeled lateral flow assay for the detection of nucleic acids. *Biomedical microdevices* 2013; 15 (5):751-758. DOI: 10.1007/s10544-013-9760-1
106. Corstjens P, Zuiderwijk M, Brink A, Li S, Feindt H, Niedbala RS, Tanke H. Use of up-converting phosphor reporters in lateral-flow assays to detect specific nucleic acid sequences: a rapid, sensitive DNA test to identify human papillomavirus type 16 infection. *Clinical chemistry* 2001; 47 (10):1885-1893.
107. Corstjens PL, Zuiderwijk M, Nilsson M, Feindt H, Sam Niedbala R, Tanke HJ. Lateral-flow and up-converting phosphor reporters to detect single-stranded nucleic acids in a sandwich-hybridization assay. *Analytical biochemistry* 2003; 312 (2):191-200. DOI: 10.1016/S0003-2697(02)00505-5
108. Qin Z, Chan WC, Boulware DR, Akkin T, Butler EK, Bischof JC. Significantly improved analytical sensitivity of lateral flow immunoassays by using thermal contrast. *Angewandte Chemie* 2012; 124 (18):4434-4437. DOI: 10.1002/ange.201200997
109. Li C, Luo W, Xu H, Zhang Q, Xu H, Aguilar ZP, Lai W, Wei H, Xiong Y. Development of an immunochromatographic assay for rapid and quantitative detection of clenbuterol in swine urine. *Food Control* 2013; 34 (2):725-732. DOI: 10.1016/j.foodcont.2013.06.021
110. Kolosova AY, De Saeger S, Sibanda L, Verheijen R, Van Peteghem C. Development of a colloidal gold-based lateral-flow immunoassay for the rapid simultaneous detection of zearalenone and deoxynivalenol. *Analytical and bioanalytical chemistry* 2007; 389 (7-8):2103-2107. DOI: 10.1007/s00216-007-1642-z
111. Le T, Yan P, Xu J, Hao Y. A novel colloidal gold-based lateral flow immunoassay for rapid simultaneous detection of cyromazine and melamine in foods of animal origin. *Food chemistry* 2013; 138 (2):1610-1615. DOI: 10.1016/j.foodchem.2012.11.077
112. Renger F, Bang H, Feist E, Fredenhagen G, Natusch A, Backhaus M, Burmester G-R, Egerer K. Research article Immediate determination of ACPA and rheumatoid factor-a novel point of care test for detection of anti-MCV antibodies and rheumatoid factor using a lateral-flow immunoassay. 2010.
113. Taranova NA, Byzova NA, Zaiko VV, Starovoitova TA, Vengerov YY, Zherdev AV, Dzantiev BB. Integration of lateral flow and microarray technologies for multiplex immunoassay: application to the determination of drugs of abuse. *Microchimica Acta* 2013; 180 (11-12):1165-1172. DOI: 10.1007/s00604-013-1043-2
114. Wang Y-K, Yan Y-X, Ji W-H, Wang H-a, Li S-Q, Zou Q, Sun J-H. Rapid Simultaneous Quantification of Zearalenone and Fumonisin B1 in Corn and Wheat by Lateral Flow Dual Immunoassay. *Journal of agricultural and food chemistry* 2013; 61 (21):5031-5036. DOI: 10.1021/jf400803q
115. Zhu J, Zou N, Mao H, Wang P, Zhu D, Ji H, Cong H, Sun C, Wang H, Zhang F.

Evaluation of a modified lateral flow immunoassay for detection of high-sensitivity cardiac troponin I andmyoglobin. *Biosensors and Bioelectronics* 2013; 42:522-525. DOI: 10.1016/j.bios.2012.10.016

116. Oku Y, Kamiya K, Kamiya H, Shibahara Y, Ii T, Uesaka Y. Development of oligonucleotide lateral-flow immunoassay for multi-parameter detection. *Journal of immunological methods* 2001; 258 (1):73-84. DOI: 10.1016/S0022-1759(01)00470-7

117. Chen J, Fang Z, Lie P, Zeng L. Computational lateral flow biosensor for proteins and small molecules: a new class of strip logic gates. *Analytical Chemistry* 2012; 84 (15):6321-6325. DOI: 10.1021/ac301508b

