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Aptamers: Update to Lateral Flow Immunoassays



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Abstract

A key driver for treatment of a disease is the time to obtain the test result. Lateral flow immunoassays are multifaceted and available for a broad range of analytes from blood proteins, mycotoxins, viral pathogens or bacterial toxins. The other key advantage of these rapid tests as compared to other immunoassays is the simplicity of the test with requirement of little sample or reagent preparation. Lateral flow assay requires assembly of a number of variants such as formats, biorecognition molecules, labels, detection systems and applications. This manuscript provides a brief overview of use of aptamers in lateral flow assay without using whole antigens or antibodies for the targets. Due to a number of important potential performance advantages, aptamers are receiving increasing attention in lateral flow applications. The identification of specific nucleic acids (aptamers) which bind to a wide range of target molecules with high affinity and specificity is done by *in vitro* selection of combinatorial libraries of RNA or DNA. These are used as new biological recognition molecules in lateral flow immunoassays assuring progress for fast and easy detection of antigens/antibodies.

Keywords: Lateral flow immunoassay; Biorecognition; Aptamer; Nucleic acid

Introduction

Lateral flow assay (LFA) based Point of Care (POC) devices are among very rapidly growing strategies for qualitative and quantitative analysis. One step rapid analysis, very low operational cost, no instrumentation, less or no interferences due to chromatographic separation, high specificity & sensitivity, and portability of the device are unique advantages related to LFA [1]. It is performed over a strip, different parts of which are assembled on a plastic (PVC) backing. These strips include sample pad, conjugate pad, nitrocellulose membrane and adsorption pad [2]. Nitrocellulose membrane is further coated with specific biorecognition molecules for test and control line. Pre-immobilized reagents at conjugate pad and nitrocellulose membrane of the strip become active upon lateral flow of liquid sample. In ELISA, immunobead assay, western blotting, microarrays and biosensors, proteins are detected mostly by specific antibodies. But, aptamers are equal to monoclonal antibodies concerning their binding affinities and their other decisive advantages [3].

Aptamers in LFA

There have been attempted many efforts for lateral flow immunoassays to improve the sensitivity, reproducibility, quantification, and multiplexing capability. Both DNA and RNA aptamers show robust binding affinities for various targets [4-6]. These are very small in size (30 to 100 nucleotides) in

comparison to other biorecognition molecules like antibodies or enzymes which allows competent immobilization at high density. Therefore, use of aptamers over antibodies in miniaturization and integration of lateral flow immunoassays can be accomplished more easily.

Aptamers are able to recognize a distinct epitope of a target molecule and can distinguish between chiral molecules [7,8]. These can be engineered completely in a test tube, can be readily produced by chemical synthesis and possess desirable storage properties. For any desired target, even non-immunogenic or toxic proteins, aptamers can be selected virtually, because they are produced in vitro by an evolutionary method called SELEX (systematic evolution of ligands by exponential enrichment) process [9,10]. The big variety of DNA/RNA library and the amplification steps of target-binding oligonucleotides during the selection process considerably facilitate the selection of ligands with highest affinity [11]. To direct the selection to aptamers with desired features, the SELEX conditions can be further modified. Also, it has been proved that one aptamer could be split into two pieces, which could simultaneously bind to the same target such as thrombin, ATP, theophylline, 17 beta-estradiol, and cocaine [12-19]. An aptamer based lateral flow strip with competitive format has been developed for on-site rapid detection of ochratoxin A (OTA) in Astragalusmembranaceus [20].

Discussion

Currently many diagnostic tests rely on antibodies, which are used as biorecognition molecules to detect a targeted molecule. The common pregnancy test, for example, detects pregnancy when an antibody indicator binds to a glycoprotein (hCG), found in urine [21]. Aptamers could be described as synthetic antibodies, single-stranded DNA or RNA molecules that can bind to pre-selected targets. They are regarded as having many advantages over antibodies. Because of their exceptionally high stability, selectivity and sensitivity, aptamer based lateral flow immunoassys have the potential to overcome the lacking functional and storage stability of most lateral flow tests.

Conclusion

The use of aptamers as new biological receptors can accelerate the development of lateral flow immunoassays of practical relevance. The more aptamers for proteins will be developed and characterized, the more aptamer based lateral flow immunoassays will be developed in the future.

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