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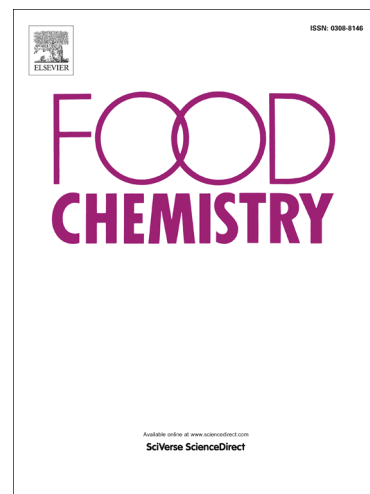
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FRET-based aptamer biosensor for selective and sensitive detection of aflatoxin B1 in peanut and rice

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ABSTRACT

Aflatoxins are potential food pollutants produced by fungi. Among them, Aflatoxin B1 (AFB1) is the most toxic. Therefore, a great deal of concern is associated with AFB1 toxicity. In this work, utilizing a FRET-based method, we have developed a nanobiosensor for detection of AFB1 in agricultural foods. Aptamer-conjugated Quantum dots (QDs) are adsorbed to Au nanoparticles (AuNPs) due to interaction of aptamers with AuNPs leading to quenching effect on QDs fluorescence. Upon the addition of AFB1, the specific aptamers are attracted to AFB1, getting distance from AuNPs which result in fluorescence recovery. Under optimized conditions the detection limit of proposed nanobiosensor was 3.4 nM with linear range of 10 – 400 nM. Selectivity test demonstrates that the nanobiosensor could be a promising tool for specific evaluation of food stuff. This method was successfully applied for the analysis of AFB1 in rice and peanut samples.

Keywords: Aflatoxin B1, Aptasensor, FRET, Sensitive Detection, Fluorescence, Quantum dots, Nanobiosensor, Food, Au nanoparticle

1. Introduction

Aflatoxins (AFs) are toxic compounds produced as secondary metabolites by the fungi namely *Aspergillus flavus* and *Aspergillus parasiticus* (Yabe et al., 1988). Among more than 20 identified aflatoxins which contaminate several agriculture crops, Aflatoxin B1 (AFB1) is considered the most toxic (Nonaka et al., 2009). AFB1 has been classified as a Group I carcinogen in humans while having mutagenic, hepatotoxic and teratogenic effects in animal species (Bacaloni et al., 2008). There has been several reports on AFB1 outbreaks, in which the most shocking was reported in 2004 with 125 deaths in Kenya (Khayoon et al., 2010; Nonaka et al., 2009).

Considering its potential threat, many analytical methods have been developed for determination of AFB1 in various matrices including liquid chromatography (LC), thin-layer chromatography (TLC), high-performance liquid chromatography (HPLC) immunoaffinity chromatography (IAC), enzyme-linked immunosorbent assay (ELISA) and electrochemical immunosensor (Amaya-González et al., 2013; Bacaloni et al., 2008; Khayoon et al., 2010; Nonaka et al., 2009; Piermarini et al., 2007). However due to limitations associated with these methods, including extensive sample preparation, expensive procedure and unavailability for on-site screening, increasing demand has been emerged especially in developing countries for more simple and cost-effective methods.

Semiconductor quantum dots (QDs) as a new type of fluorescent probes, have unique optical characteristics such as photostability and high quantum yield originated from “quantum size” effect, and have been proven to be of many use in biosensing application (Zhang et al., 2012; Alivisatos, 1996).

Aptamers are artificially-synthesized single-stranded DNA or RNA sequences which can take secondary and tertiary structures allowing them to bind to specific targets with high

affinity (Iliuk et al., 2011). Since their report in early 1990s (Ellington & Szostak, 1990; Robertson & Joyce, 1990; Tuerk & Gold, 1990)., aptamers have gained increasing interest due to their advantages including no limitation for their target and ease of reproduction (Kim et al., 2016). In this work, utilizing a QD-aptamer-AuNP FRET system we have developed a sensitive nanobiosensor for detection of AFB1 in rice and peanut samples.

2. Experimental

2.1. Reagents

Hydrogen tetrachloroaurate (III) tetrahydrate, cadmium chloride hydrate, tellurium powder, N-hydroxysuccinimide (NHS), 1-ethyl-3-(3-dimethylaminopropyl) carbodiimide (EDC) and sodium citrate were purchased from Merck and all other commercially available substances were purchased from Aldrich and Acros, and used without further purification. The 50-mer 5'-amino-modified AFB1 aptamer oligonucleotide (5'-GTTGGGCACGTGTTGTCTCTCTGTGTCTCGTGCCCTTCGCTAGGCCAC A-3') was synthesized by Shanghai Generay Biotech Co (Shanghai, China) and was based on a patent obtained by NeoVentures Biotechnology Inc. The aflatoxin B1 (AFB1) standard sample was purchased from Sigma-Aldrich, Co. All other reagents were of analytical reagent grade and ultra-pure water (Milli-Q plus, Millipore Inc., Bedford, MA) was used throughout the reactions.

2.2. Apparatus

Absorption spectra were studied on a Perkin-Elmer lambda 25 spectrometer and all the fluorescence measurements were carried out on a Perkin-Elmer LS55 luminescence spectrometer.

2.3. Synthesis of CdTe quantum dots

The experimental procedure was based on (Silva et al., 2012). In summary, Cd solution (0.4 mmol) and thioglycolic acid (TGA) (1.4 mmol) were solvated in 80 mL distilled water with pH adjusted to 10.0 using NaOH solution. Next, sodium borohydrate (0.8 mmol) and Te powder were diluted in 10 ml distilled water in a flask, with vigorous stirring under argon flow. The mixture was heated to 80°C to get a clear red NaHTe solution. Cd-TGA solution was heated at 100°C under argon flow in a 250mL three-neck flask. Then the freshly prepared NaHTe solution (4.0 mL) was added to the flask, and the resulting solution was refluxed at 100 °C for 2 hours. The characterization of CdTe quantum dots was carried out through transmission electron microscopy and spectrofluorometer. (Fig. S1).

(Fig. S1)

2.4. Conjugation of ssDNA aptamers to CdTe QDs

Amine-modified aflatoxin aptamers were dissolved in TE Buffer (1X). Immobilization was carried out similar to Kim et al (Kim & Jurng, 2011). Firstly, the activation of carboxyl-functionalized QDs was conducted using EDC (1 mM) and NHS (1mM) for 30 minutes. Freshly activated QDs were incubated with amine modified aptamers for 2 hours. Non-reacted COOH groups on the QDs were blocked using ethanolamine solution (10 mM). Eventually, purification of aptamer/QDs conjugates was conducted by centrifugation and resuspension in phosphate buffer.

2.5. The synthesis of citrate-protected AuNP solution

The synthesis was carried out as follows: 50 ml of $\text{HAuCl}_4 \cdot 4\text{H}_2\text{O}$ aqueous solution (1 mM) was heated in a flask with until reaching boiling state. Then 10 mL trisodium citrate (38.8 mM) was added to boiling solution while stirring. The mixture was boiled for another 10 min. The yellow color of mixture changed to wine red. The heating was stopped and the

solution was cooled down in room temperature while being stirred. (Borghei et al., 2016; Hosseini et al., 2015). After that, the resulting AuNPs solution was stored at 4 °C. The characterization of synthesized AuNPs was conducted by TEM. The concentration of AuNPs was calculated using the extinction coefficient ($2.7 \times 10^8 \text{ M}^{-1} \text{ cm}^{-1}$) at 520 nm and Beer's law to be about 10 nM. As shown in Fig.S2 the diameter of AuNPs was about 13 nm.

2.6. Assay procedure

Unmodified AuNPs were added to aptamer conjugated QDs solution followed by a 15 min mild shaking. Then, sample solutions containing different concentrations of AFB1 were added into the mixture and incubated for 30 min at room temperature. The fluorescent intensity of the solution was measured by a Luminescence Spectrometer using excitation wavelength of 350 nm.

2.7. Preparation of peanuts and rice samples

The fresh peanut and rice samples purchased from a local market were finely ground with a laboratory mill. 5 g of sample in 15 mL of methanol–water (60:40 v/v) was added to a 50-mL centrifuge tube. Extraction of the samples carried out by shaking for 100 min followed by 5 min centrifugation at 3500 rpm at room temperature. The supernatant was transferred to a 1.5-mL tube and diluted with pure water which subsequently was used as blank sample, and the spiked samples with various concentrations of AFB1 were used as analytical samples. Eventually the results of the proposed nanobiosensor were validated using HPLC method.

3. Results and Discussion

3.1. Principles of FRET based aptasensor

In this work, we have developed an aptasensor for detection of AFB1 based on FRET mechanism, using CdTe QDs and AuNPs as donor and acceptor respectively. Scheme 1 depicts the principle mechanism of proposed method (scheme 1). Aflatoxin specific aptamers

were covalently linked to CdTe QDs beforehand which has been confirmed by Gel-electrophoresis (data not shown). The non-covalent interaction of ssDNA and AuNPs has been reported before (Li & Rothberg, 2004), so as shown in Fig. 1(a) during incubation in the absence of aflatoxin B1, Aptamer-conjugated QDs were absorbed to AuNPs due to electrostatic interaction between exposed bases of aptamers and AuNPs. In addition, because of suitable overlap between fluorescence emission of QDs and the absorption spectrum of AuNPs (Fig. 1b), the fluorescence of QDs can be substantially quenched in close proximity of AuNPs (Zhang et al., 2010). Therefore when there is no target available, the fluorescence signal of QDs is quenched dramatically (FRET phenomena). On the other hand, in the presence of the target aflatoxin B1, QDs-aptamers prefer to bind with their specific target and form a QD-aptamer-target complex which lead to ssDNAs to get distance from AuNPs resulting in recovery of QDs fluorescence.

Scheme 1

Fig. 1

3.2. Optimization

In order to obtain the best performance, some effective conditions such as concentration ratio of aptamer to QDs, AuNPs concentration and incubation time were optimized. As shown in Fig. 2(a), high molar ratio of aptamer to QDs led to requirement of too much aflatoxin to release the conjugates from quantum dots (ineffective recovery of fluorescence) and low sensitivity of biosensor. On the other hand, weak FRET effect was seen upon implementation of low molar ratio of those. Molar ratio 8 was chose as the optimum ratio. After conjugation of aptamers, in order to block remaining carboxyl group of QDs, they were treated with ethanolamine, because these negative charges can induce a repulsion effect due to proximity of negatively charged AuNPs.

As expected, increasing concentrations of AuNPs had a decreasing effect on the fluorescence intensity of QDs-Aptamers (Fig. 2b). The fluorescence signal was almost completely quenched after incubating with AuNPs when the concentration of which was 140 pM. Therefore 350 pM was considered as the optimum concentration.

Finally the effect of incubation time for QDs-aptamers and AuNPs over the range of 0-30 min was studied. As it can be seen in the Fig. 2(c), incubation times which were less than 15 min resulted in incomplete quenching of QDs fluorescence signal. On the other hand, those more than 15 min had no significant difference compared to incubation time of 15 min. Therefore, 15 min was regarded as the best choice.

Fig. 2

3.3. Analytical performance of Aptasensor

As mentioned before, in the absence of target AFB1 the high fluorescence of QD-aptamer conjugates was weakened. On the other hand in the presence of aflatoxin B1 aptamers were attracted to their specific target and would not adsorb on the surface of AuNPs which led to a stronger fluorescence signal. The higher the aflatoxin concentration is, the higher the fluorescence signal is obtained. That being so, the fluorescence intensities were measured against the AFB1 concentrations (Fig. 3). Under optimized conditions, there was a linear relationship between fluorescence signal and aflatoxin B1 concentration in the range between 10 – 400 nM (Inset, Fig. 3). The regression equation was $I=0.43C+66.89$ and this equation had a correlation coefficient of 0.989, which indicated that there was a good linear relationship between fluorescence intensity and aflatoxin B1 concentration. The detection limit for aflatoxin B1 determined at 3.4 nM based on $S/N=3$. The relative standard deviation was 2.4% for the determination of 100 nM aflatoxin B1 ($n=4$).

Table 1 shows the comparison of the sensitivity, rapidity and simplicity in this work with other aptamer, enzyme or antibody based methods. Some aptasensors has been reported for AFB1 analysis. Recent study conducted by Zhang et al. showed that electrochemical immunosensor could detect 11.2 pM of AFB1 but required time-consuming (> 3 h) and complicated steps (> 6; pre-incubation, washing, etc). Also our work has comparable detection limit with the Lu et al. aptasensor. The sensitivity of FRET based aptasensor suggested in this work was also comparable to those of other immunosensors except for direct competitive fluorescence-linked immunosorbent assay electrochemical immunosensor based methods which require complicated experimental steps (6 steps). However, compared with the currently immunosensors, the results in this work clearly indicated an excellent sensitivity for detection of AFB1.

Fig. 3

Table 1

3.4. Selectivity

In order to confirm the selectivity of the aptasensor for aflatoxin B1, some other mycotoxins including Aflatoxin B2, Aflatoxin M2 and Aflatoxin G2 were tested. As shown in Fig. 4, compared to aflatoxin B1, no significant increase in fluorescence intensity was observed for common interferes and blank sample except for a minor increase for aflatoxin M2. This data confirm that the FRET-based aptasensor has suitable selectivity for aflatoxin B1.

Fig. 4

3.5. Real Sample Assay

The proposed method was used to detect AFB1 in rice and peanut and the accuracy was studied by recovery experiment. The prepared blank and spiked samples were tested by

replacing AFB1 standard samples and the concentration of AFB1 in peanut and rice samples was determined from the calibration curve. The results were compared with those obtained by the HPLC method. As shown in Table 2, percentage recovery of the spiked AFB1 samples in the range of 103.0% to 108.0% was obtained, indicating acceptable accuracy of the proposed detection sensor for AFB1 in real samples. This analysis demonstrated that the established biosensing system could be applied for AFB1 determination in real agriculture products.

Table 2

4. Conclusion

This study offered a new nanobiosensor for detection of AFB1, involving aptamer as specific recognition element and CdTe QDs as fluorescence indicators using FRET phenomena. This signal-on method showed a suitable sensitivity towards AFB1 while common interferences such as aflatoxin B2, aflatoxin G2 and aflatoxin M2 didn't show significant signal. After optimization of some conditions, the linear range of aptasensor covered a large concentration range from 10 to 400 nM with a detection limit of 3.4 nM.

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References

- Alivisatos, A. P. (1996). Semiconductor clusters, nanocrystals, and quantum dots. *Science*, 271, 933.
- Amaya-González, S., de-los-Santos-Álvarez, N., Miranda-Ordieres, A. J., & Lobo-Castañón, M. J. (2013). Aptamer-based analysis: A promising alternative for food safety control. *Sensors*, 13, 16292-16311.
- Bacaloni, A., Cavaliere, C., Cucci, F., Foglia, P., Samperi, R., & Laganà, A. (2008). Determination of aflatoxins in hazelnuts by various sample preparation methods and liquid chromatography–tandem mass spectrometry. *Journal of Chromatography A*, 1179, 182-189.
- Borghei, Y. S., Hosseini, M., Dadmehr, M., Hosseinkhani, S., Ganjali, M. R., & Sheikhnejad, R. (2016). Visual detection of cancer cells by colorimetric aptasensor based on aggregation of gold nanoparticles induced by DNA hybridization. *Analytica chimica acta*, 904, 92-97.
- Ellington, A. D., & Szostak, J. W. (1990). In vitro selection of RNA molecules that bind specific ligands. *Nature*, 346, 818-822.
- Hosseini, M., Khabbaz, H., Dadmehr, M., Ganjali, M. R., Mohamadnejad, J. (2015). Aptamer-based Colorimetric and Chemiluminescence Detection of Aflatoxin B1 in Foods Samples," *Acta Chimica Slovenica*, 62, 721-728.
- Illiuk, A. B., Hu, L., & Tao, W. A. (2011). Aptamer in bioanalytical applications. *Analytical chemistry*, 83, 4440-4452.
- Khayoon, W. S., Saad, B., Yan, C. B., Hashim, N. H., Ali, A. S. M., Salleh, M. I., et al., (2010). Determination of aflatoxins in animal feeds by HPLC with multifunctional column clean-up. *Food Chemistry*, 118, 882-886.
- Kim, Y. S., & Jurng, J. (2011). Gold nanoparticle-based homogeneous fluorescent aptasensor for multiplex detection. *Analyst*, 136, 3720-4.

- Kim, Y. S., Raston, N. H., & Gu, M. B. (2016). Aptamer-based nanobiosensors. *Biosensors and Bioelectronics*, 76, 2-19.
- Li, H., & Rothberg, L. J. (2004). Label-free colorimetric detection of specific sequences in genomic DNA amplified by the polymerase chain reaction. *Journal of the American Chemical Society*, 126, 10958-10961.
- Lu, Z., Chen, X., Wang, Y., Zheng, X., & Li, C. M. (2014). Aptamer based fluorescence recovery assay for aflatoxin B1 using a quencher system composed of quantum dots and graphene oxide. *Microchimica Acta*, 182(3-4), 571-578.
- Nonaka, Y., Saito, K., Hanioka, N., Narimatsu, S., & Kataoka, H. (2009). Determination of aflatoxins in food samples by automated on-line in-tube solid-phase microextraction coupled with liquid chromatography–mass spectrometry. *Journal of Chromatography A*, 1216, 4416-4422.
- Piermarini, S., Micheli, L., Ammida, N., Palleschi, G., & Moscone, D. (2007). Electrochemical immunosensor array using a 96-well screen-printed microplate for aflatoxin B 1 detection. *Biosensors and Bioelectronics*, 22, 1434-1440.
- Robertson, D. L., & Joyce, G. F. (1990). Selection in vitro of an RNA enzyme that specifically cleaves single-stranded DNA. *Nature*, 344, 467-8.
- Silva, F. O., Carvalho, M. S., Mendonca, R., Macedo, W. A., Balzuweit, K., Reiss, P., et al., (2012). Effect of surface ligands on the optical properties of aqueous soluble CdTe quantum dots. *Nanoscale Research Letters*, 7, 536.
- Soldatkin, O. O., Burdak, O. S., Sergeyeva, T. A., Arkhypova, V. M., Dzyadevych, S. V., & Soldatkin, A. P. (2013). Acetylcholinesterase-based conductometric biosensor for determination of aflatoxin B1. *Sensors and Actuators, B: Chemical*, 188, 999-1003.
- Tuerk, C., & Gold, L. (1990). Systematic evolution of ligands by exponential enrichment: RNA ligands to bacteriophage T4 DNA polymerase. *Science*, 249, 505-10.
- Yabe, K., Ando, Y., & Hamasaki, T. (1988). Biosynthetic relationship among aflatoxins B1, B2, G1 and G2. *Applied and environmental microbiology*, 54, 2101-2106.
- Ye, Y., Zhou, Y., Mo, Z., Cheng, W., Yang, S., Wang, X., et al., (2010). Rapid detection of aflatoxin B1 on membrane by dot-immunogold filtration assay. *Talanta*, 81, 792-798.
- Zhang, C., Xu, J., Zhang, S., Ji, X., & He, Z. (2012). One-Pot Synthesized DNA–CdTe Quantum Dots Applied in a Biosensor for the Detection of Sequence-Specific Oligonucleotides. *Chemistry – A European Journal*, 18, 8296-8300.

- Zhang, J., Wang, L., Zhang, H., Boey, F., Song, S., Fan, C. (2010). Aptamer-Based Multicolor Fluorescent Gold Nanoprobes for Multiplex Detection in Homogeneous Solution. *Small*, 6, 201-204.
- Zhang, X., Li, C. R., Wang, W. C., Xue, J., Huang, Y. L., Yang, X. X. et al., (2016). A novel electrochemical immunosensor for highly sensitive detection of aflatoxin B1 in corn using single-walled carbon nanotubes/chitosan. *Food Chemistry*, 92, 197-202.
- Zhang, X., Li, C. R., Wang, W. C., Xue, J., Huang, Y. L., Yang, X. X., et al., (2015). A novel electrochemical immunosensor for highly sensitive detection of aflatoxin B1 in corn using single-walled carbon nanotubes/chitosan. *Food Chemistry*, 192, 197-202.
- Zhang, Z., Li, Y., Li, P., Zhang, Q., Zhang, W., Hu, X. et al., (2014). Monoclonal antibody-quantum dots CdTe conjugate-based fluoroimmunoassay for the determination of aflatoxin B1 in peanuts. *Food Chemistry*, 146, 314-319.

Figures and legends:

Scheme 1. Schematic illustration of detection procedure by FRET-based nanobiosensor.

Fig. 1. (a) Overlap between fluorescence emission of CdTe QDs and the absorption spectrum of AuNPs. (b) Fluorescence emission of aptamer-conjugated QDs, quenching effect of AuNPs and recovery of fluorescence after addition of AFB1.

Fig. 2. Optimization assay conditions: (a) Effect of different ratios of Aptamer to QDs. (b) Effect of different concentrations of AuNPs. (c) Optimization of incubation time.

Fig. 3. Fluorescence recovery of aptasensor with different concentrations of AFB1 (10-500 nM). Inset: calibration curves for detection of aflatoxin B1.

Fig. 4. Selectivity evaluation of aptasensor using common mycotoxins

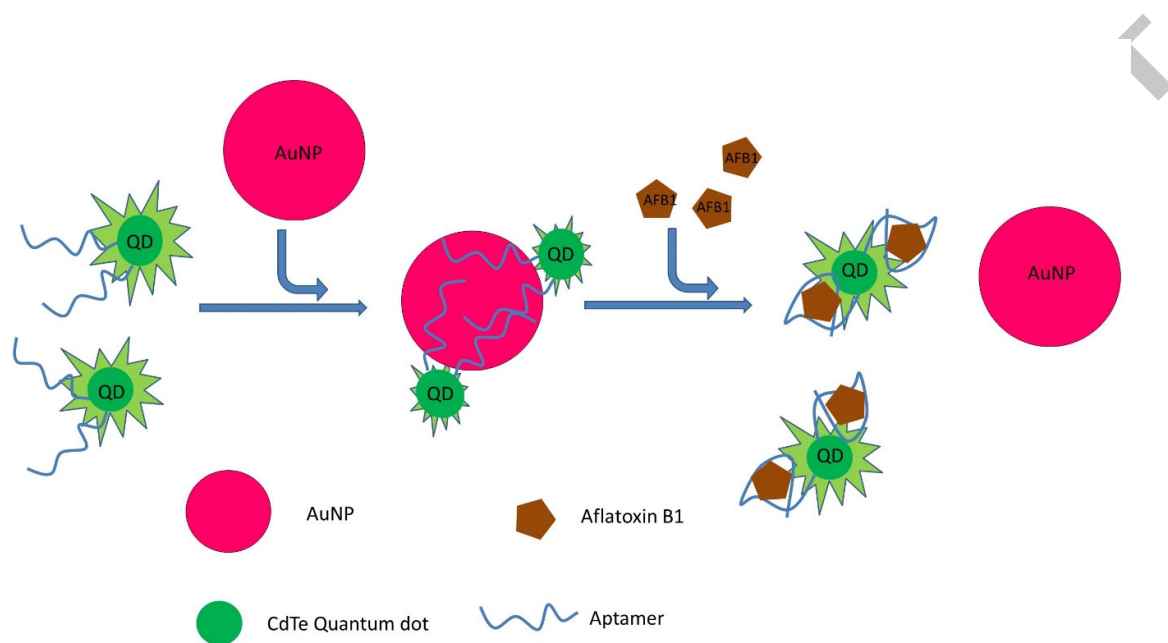
Table 1. Comparisons of the characteristics of the proposed sensor with those of the previously AFB1

Detection technique	Recognition probe	Step required	Time	Limit of Detection	Ref.
electrochemical immunosensor	Antibody	> 6	> 3 h	11.2×10^{-12} M	(Zhang et al., 2016)
direct competitive fluorescence-linked immunosorbent assay	Antibody	> 5	> 4 h	50×10^{-12} M	(Zhang et al., 2014)
electrochemical immunosensor	Antibody	6	> 3 h	11×10^{-12} M	(Zhang et al., 2015)
Immunofiltration assay	Antibody	> 6	> 2 h	6.41×10^{-9} M	(Ye et al., 2010)
acetylcholinesterase -based conductometric	Enzyme	-	-	0.16×10^{-6} M	(Soldatkin et al., 2013)
QD-Aptamer-GO system	Aptamer	3	<1 h	1×10^{-9} M	(Lu, et al., 2014)
FRET-based Aptasensor	Aptamer	3	< 1 h	3.4×10^{-9} M	This Work

Table 2. Analytical performance of aptasensor on peanut real samples and validation by HPLC

Sample	Spiked (nM)	Found aptasensor ^a (nM)	Recovery(%)	HPLC	Recovery(%)
Rice	5.0	(5.40 ^a ± 0.20)	108	(5.20 ± 0.10)	104
	10.0	(10.30 ^a ± 0.10)	103	(10.20 ^a ± 0.10)	102
	20.0	(21.00 ± 0.10)	104.7	(21.57 ± 0.10)	102.7
Peanut	5.0	(5.30 ± 0.1)	106	5.20 ± 0.10	104
	10.0	(10.80 ± 0.30)	108	(10.30 ± 0.10)	103
	20.0	(20.90 ± 0.10)	104.5	(20.85 ± 0.20)	104.2

^a Results are based on four measurements



Scheme1

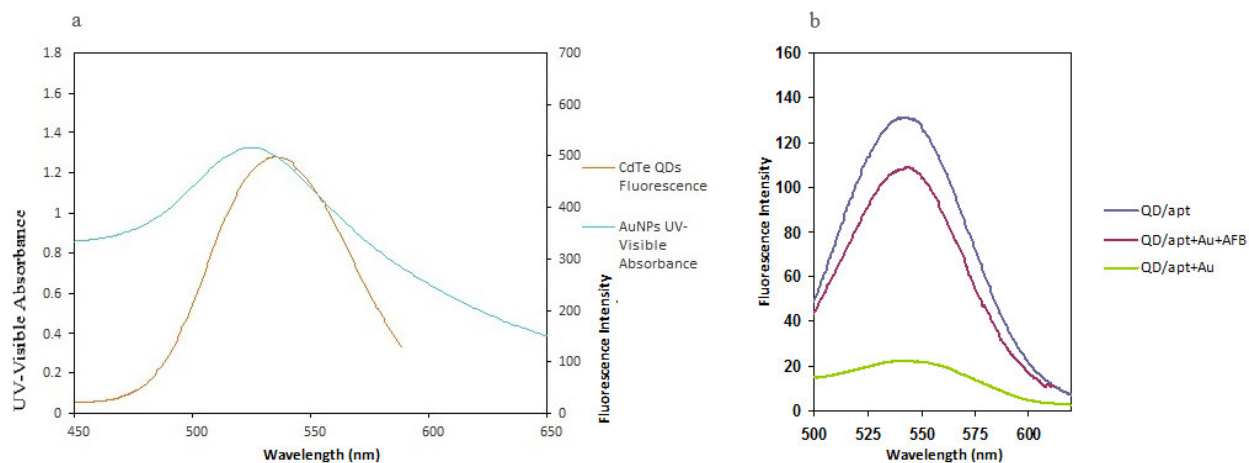


Fig. 1

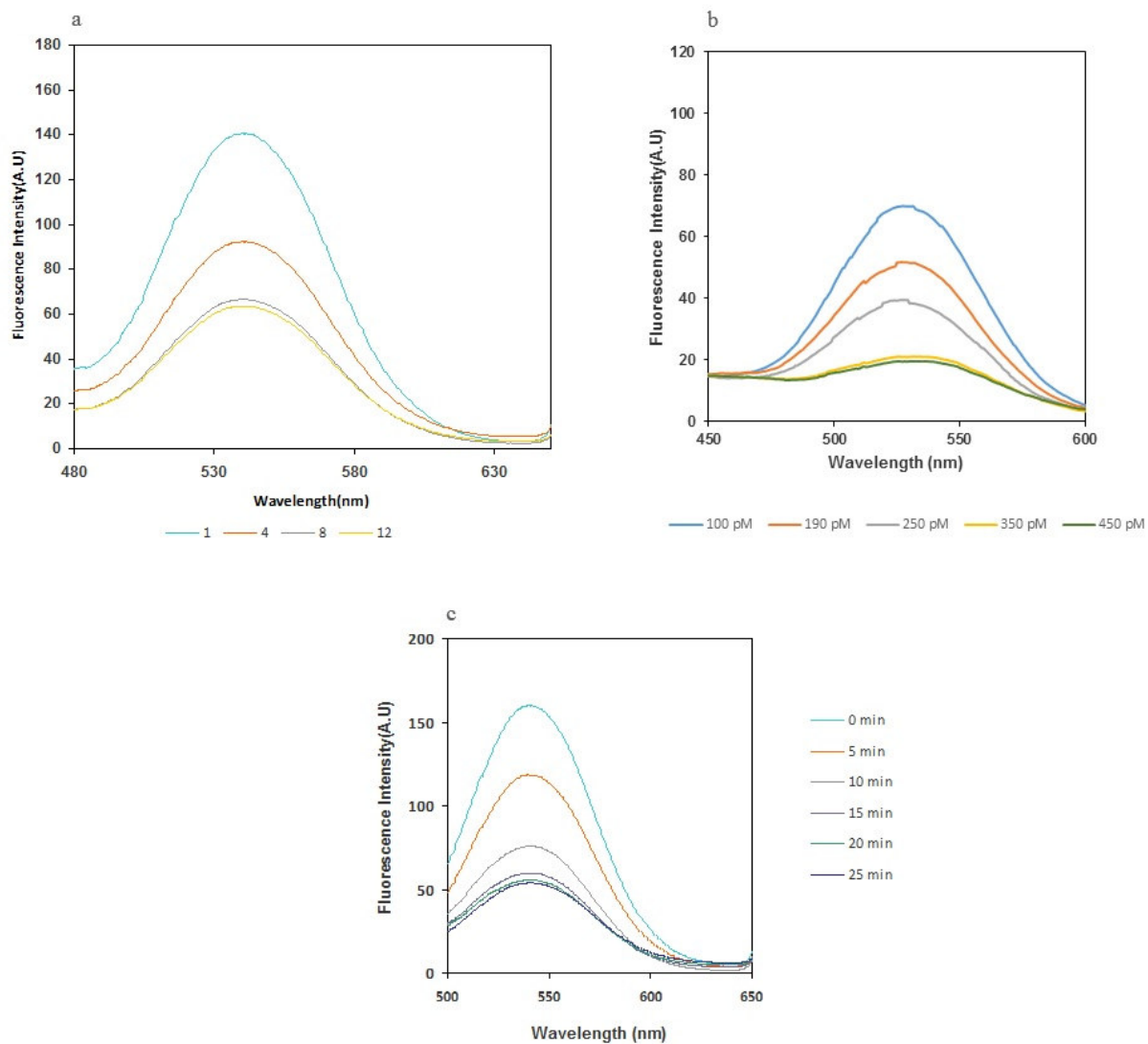


Fig. 2

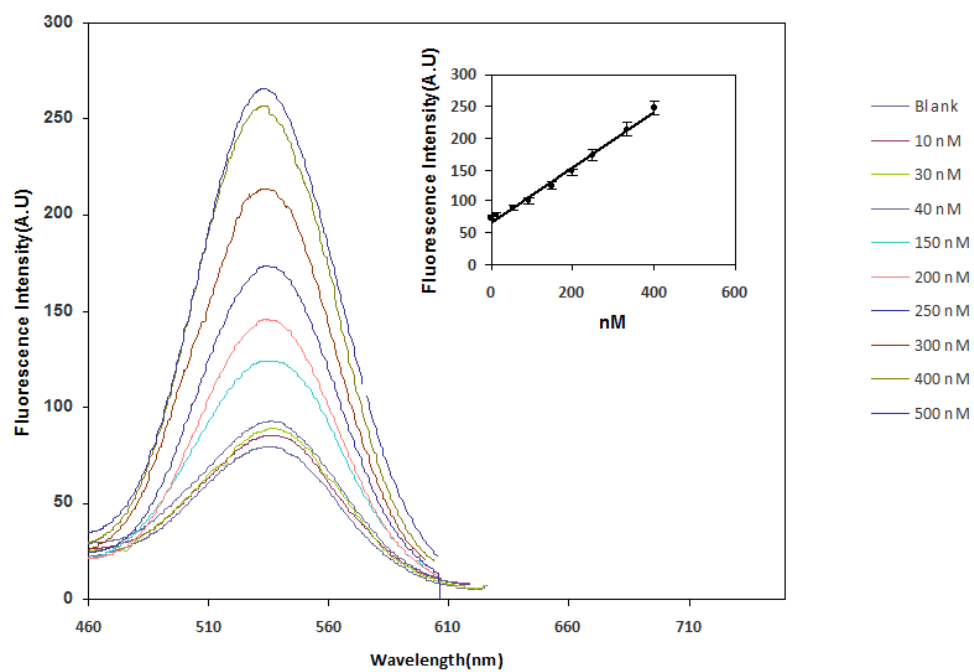
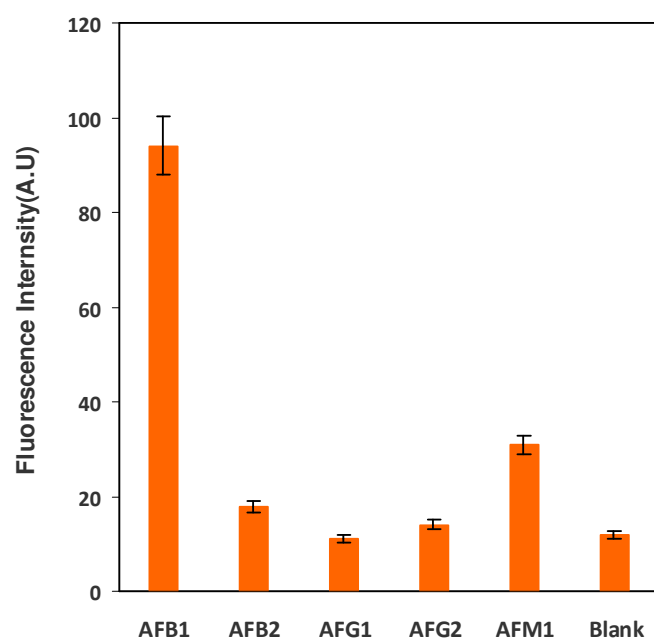


Fig. 3

**Fig. 4**

Supplementary data

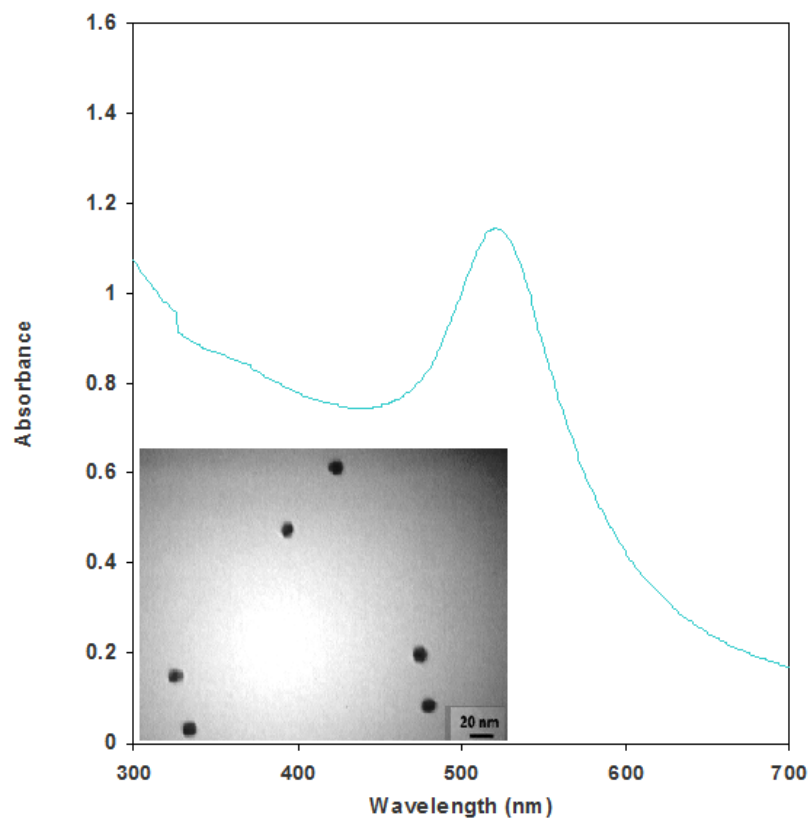
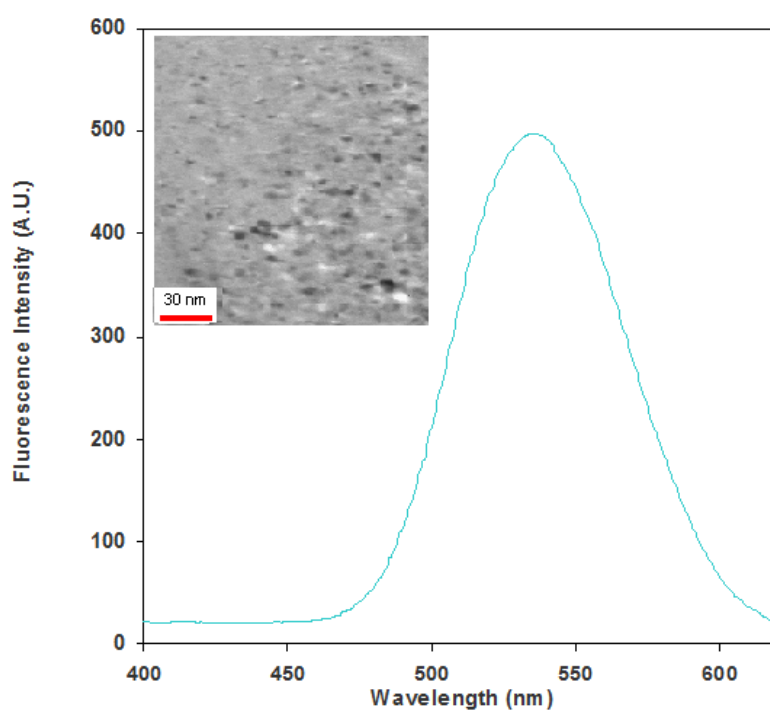


Fig.S1

**Fig.S2**

Highlights

- We present a simple FRET-based assay for Aflatoxin B1 detection.
- In this assay, aptamer-conjugated CdTe quantum dots were interacted with Au nanoparticles which led to fluorescence quenching of quantum dots.
- The aptasensor showed a linear range between 10 – 400 nM.
- This method was successfully applied for the analysis of AFB1 in rice and peanut samples.