

Supplementary Materials

Antibody and Aptamer Based Competitive Fluorescence Polarization/Anisotropy Assays for Ochratoxin A with Tetramethylrhodamine Labeled Ochratoxin A as a Fluorescent Probe

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Table S1 Comparison of competitive FA/FP assays for OTA using antibody or aptamers as affinity ligands and fluorophore labeled OTA as fluorescent probe.

Methods	Affinity ligands	Fluorophore	Temp.	Maximum signal change	LOD	Ref.
FP assay	Antibody	FAM	NA	~0.08 (P)	0.7 nM	[14]
FA assay	Antibody	TMR	25 °C	0.259 (P), 0.228 (r)	2.4 nM	this work
FA assay with streptavidin as mass enhancer	Aptamer	TMR	10 °C	0.101 (P), 0.078 (r)	2.4 nM	this work
FP assay	Antibody	FAM	NA	~0.068 (P)	1.7 nM	[11]
FP assay	Antibody	FAM	NA	NA	2 nM	[12]
FP assay using anchor protein modules	Aptamer	FAM	4 °C	~0.08 (P)	3.6 nM	[24]
FA assay using streptavidin to amplify signal	Aptamer	Lissamine Rhodamine B	10 °C	0.198 (P), 0.156 (r)	2.5 nM	[25]
FP assay	Antibody	FAM	NA	~0.05 (P)	3.7 nM	[15]
FP assay	Antibody	FAM	NA	0.05 (P)	7.4 nM	[13]

LOD: detection limit; r: fluorescence anisotropy; P: fluorescence polarization; Temp.: Temperature for detection; FAM: fluorescein;

TMR: tetramethylrhodamine; NA : not available

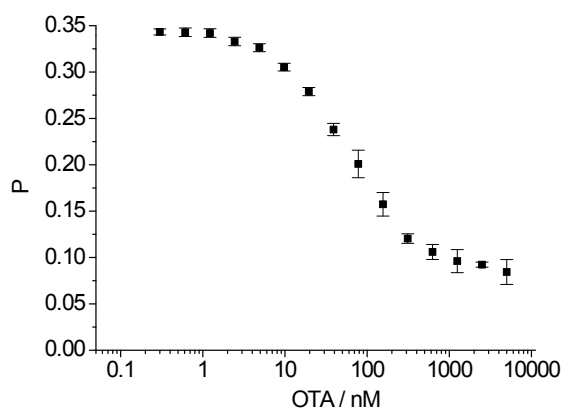


Fig. S1 Detection of OTA with the antibody based competitive FP assay. FP (P) responses to OTA. Varying concentrations of OTA were incubated with the antibody (2 nM) and Probe 1 (2 nM).

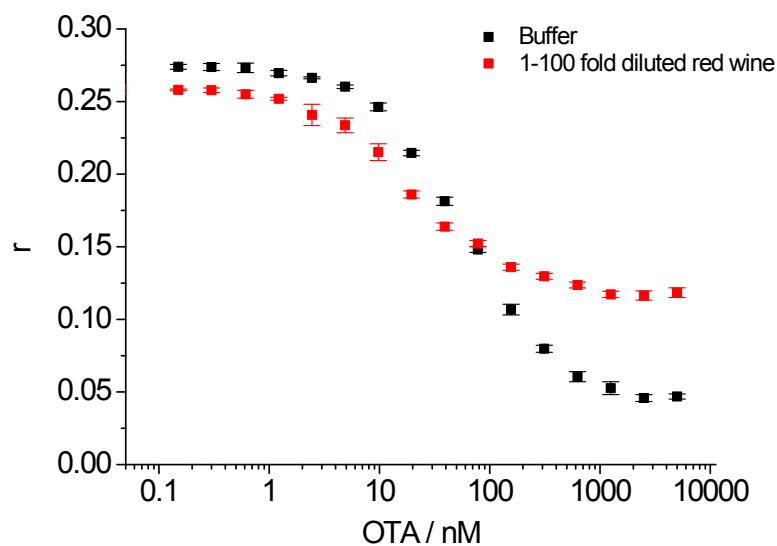


Fig. S2. Detection of OTA in diluted red wine sample by the antibody based competitive FA assay.

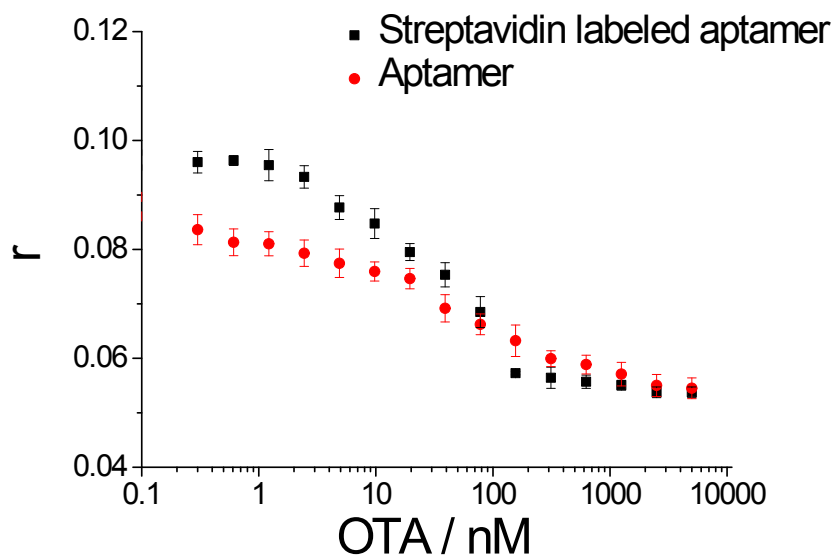


Fig. S3 Detection of OTA by the aptamer based competitive FA assay. TMR-labeled OTA (Probe 1, 20 nM), the aptamer or the streptavidin labeled aptamer (50 nM) and varying concentrations of OTA were incubated and measured at 25 °C.

The maximum FA change was about 0.033 in the FA assay using unlabeled aptamer at 25 °C, while the maximum FA change was about 0.042 for the aptamer FA assay using streptavidin labeled aptamer.

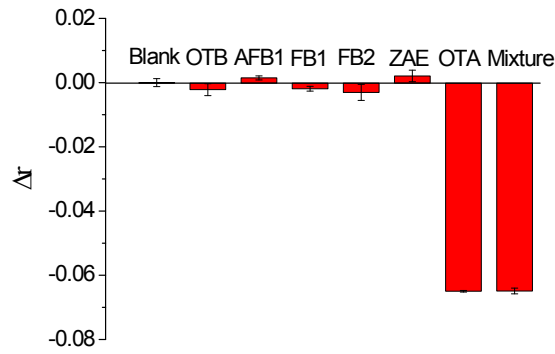


Fig. S4. Selectivity of the aptamer based competitive FA assay for OTA using the streptavidin-conjugated aptamer and Probe 1 at 10 °C. Several different mycotoxins were tested (OTB, AFB1, FB1, FB2, ZAE (each of 100 nM)). Mixture contained OTB, AFB1, FB1, FB2, ZAE and OTA (each of 100 nM).

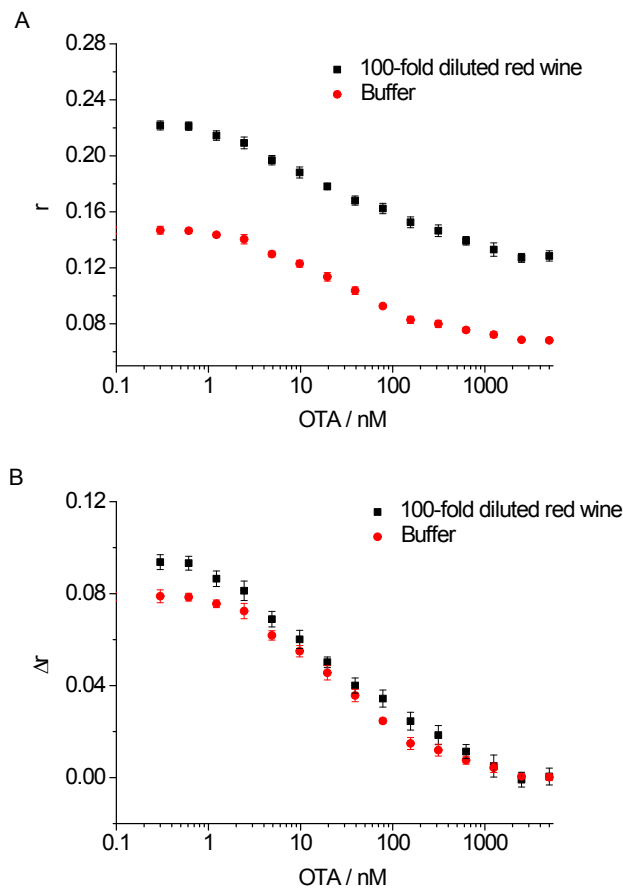


Fig. S5 Detection of OTA in diluted red wine sample by the aptamer based competitive FA assay. The (A) fluorescence anisotropy and (B) net fluorescence anisotropy responses of OTA spiked in 100-fold diluted red wine at 10 °C. 20 nM

probe 1, 50 nM SA-conjugated O36 and varying concentrations of OTA spiked in 100-fold diluted red wine samples.