Electronic Supplementary Material (ESI) for Analytical Methods.

Supporting Information

Graphene oxide-mediated fluorescence turn-ON GO-FAM-FRET aptasensor for detection of sterigmatocystin

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Table S1: Primers and oligos used in the present study

Name	Sequence	
Forward primer	CTCGTCTCGTTCTCTCAGTC	[1]
Reverse Primer	TCCTCCTTCTTCTTCGTGTC	

Flow cytometry analysis:

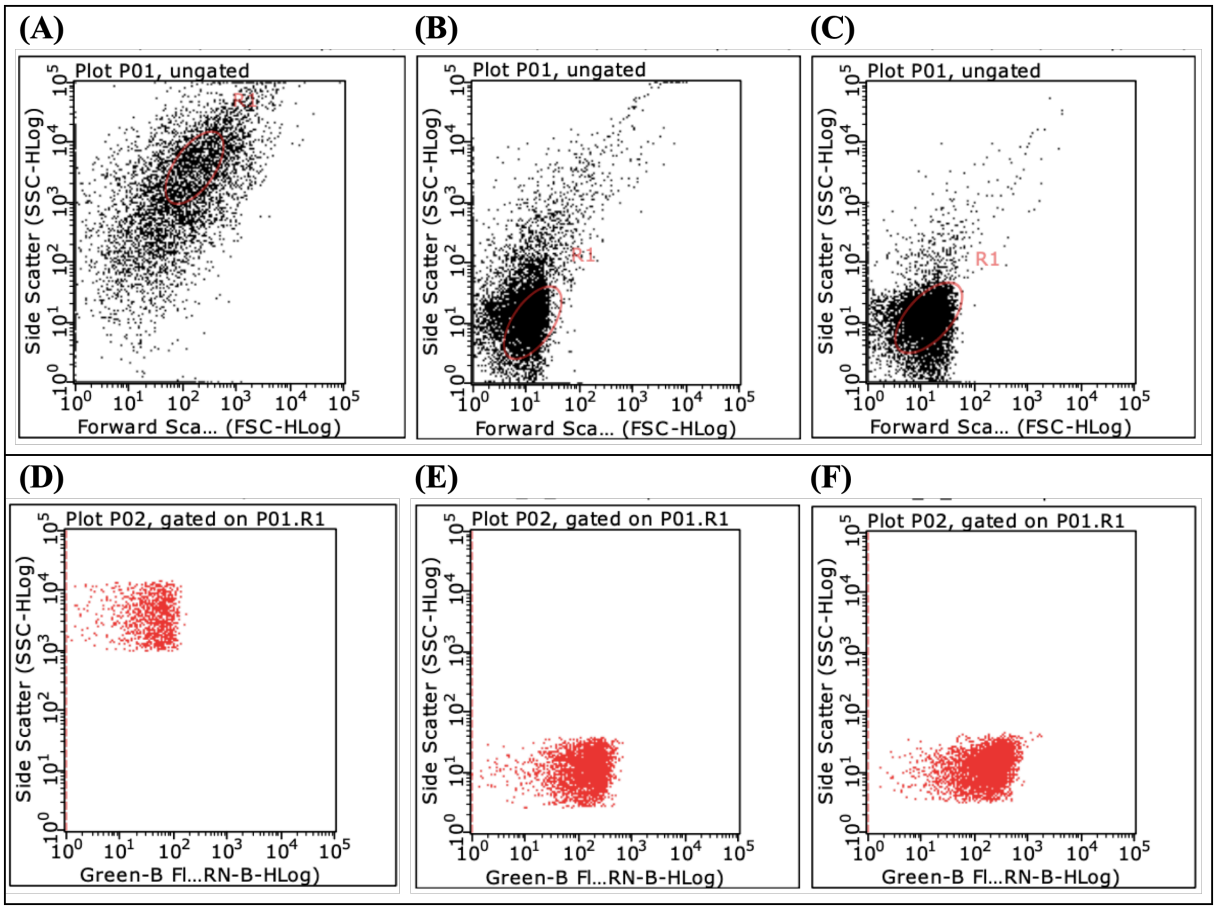
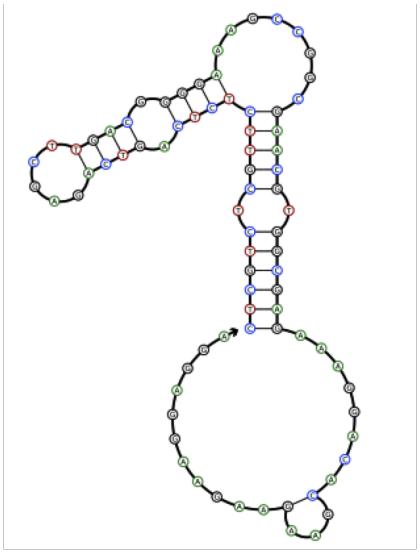


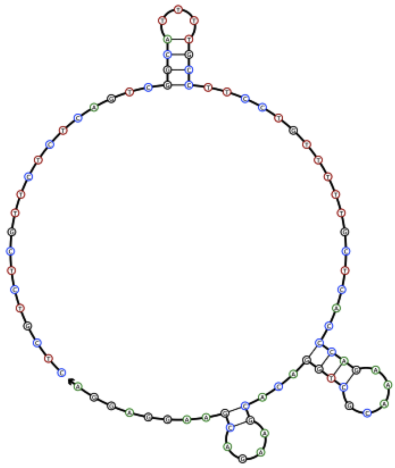
Figure S1: Flow cytometry binding analysis of the FAM labelled aptamer and STC. Forward and side scattering of the STC **(A)**, MSA-C6 **(B)** and complex **(C)**. The fluorescence in the green region vs side scattering of STC **(D)**, MSA-C6 **(E)** and complex **(F)**.

Secondary structure prediction:



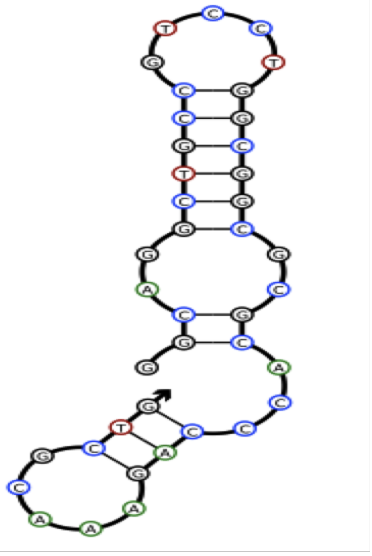
ΔG -11.8

(a)



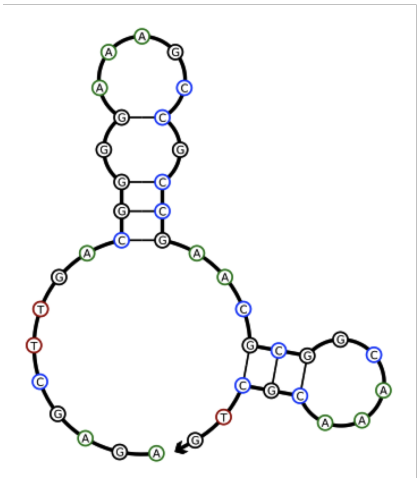
ΔG -7.9

(b)



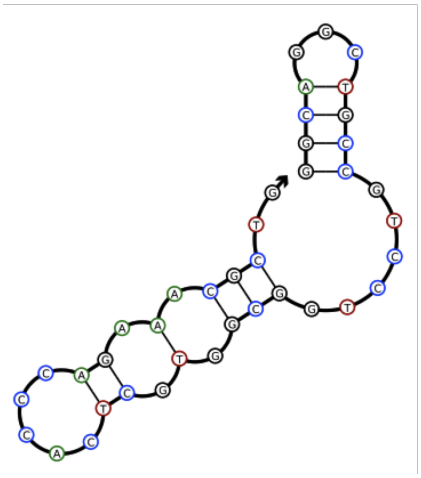
ΔG -7,68

(c)



ΔG -7.48

(d)



ΔG -5.55

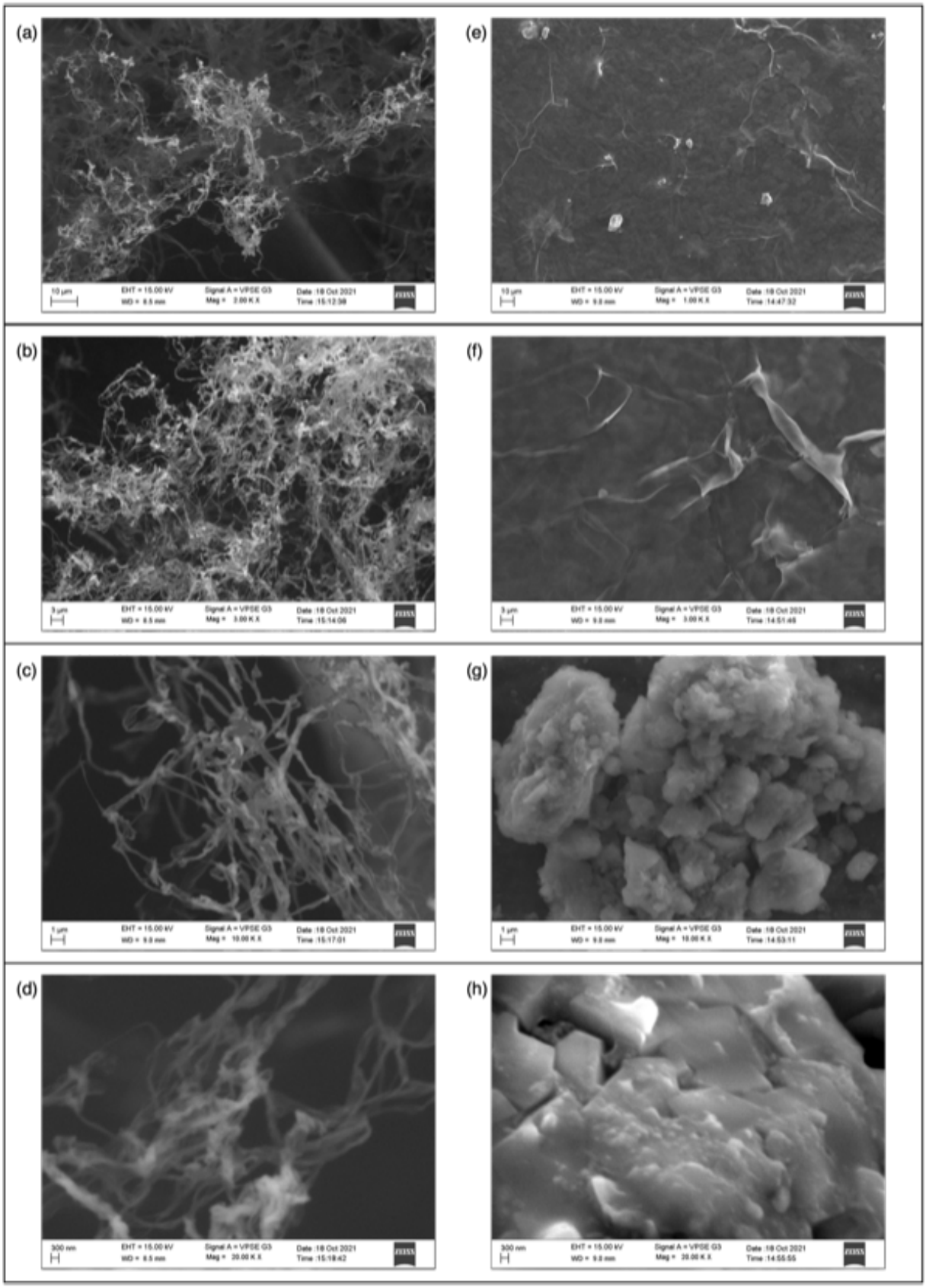
(e)

Figure S2: Secondary structure prediction. Five sequences were obtained after cloning and sequencing, Homology was performed and secondary structure was predicted using the UNAfold server.

Table S2: Comparison of different sensors developed for Sterigmatocystin.

Sr No	Analytical technique	LOD	Source	References
1	Liquid chromatography (UPLC) coupled with triple quadrupole tandem mass spectrometry (MS/MS).	0.10 µg/kg	Roasted coffee beans & black pepper	[2]
2	LC-MS/MS method using an immunoaffinity column	0.02 µg/kg	wheat flour, Job's tears products, rice etc	[3]
3	Competitive enzyme-linked immuno- sorbent assay (ELISA)	3 µg/kg	Wheat, rice & maize	[4]
4	Gas chromatography-mass spectrometry	2.4 µg/kg	Wheat, maize & rice	[5]
5	HPLC-UV	0.26 µg/L	Beer	[6]
6	Fluorescence sensor based on carbon dots-embedded molecularly imprinted polymer	0.019 mg/L	Millet, Rice and Maize	[7]
7	Fluorescence sensor based on Molecularly Imprinted Fluorescent Polymers	0.013 mg/L	Rice, Maize and Soybeans	[8]
8	GO-FAM-FRET fluorescence turn-ON aptasensor	23.56 ng/mL	Chilli & Pepper	Present study

1



2

Figure S3: Characterisation of GO and GO coated surface by SEM. (a-d) SEM images of GO at different resolutions and **(e-h)** SEM images of GO coated surface at a different resolution.

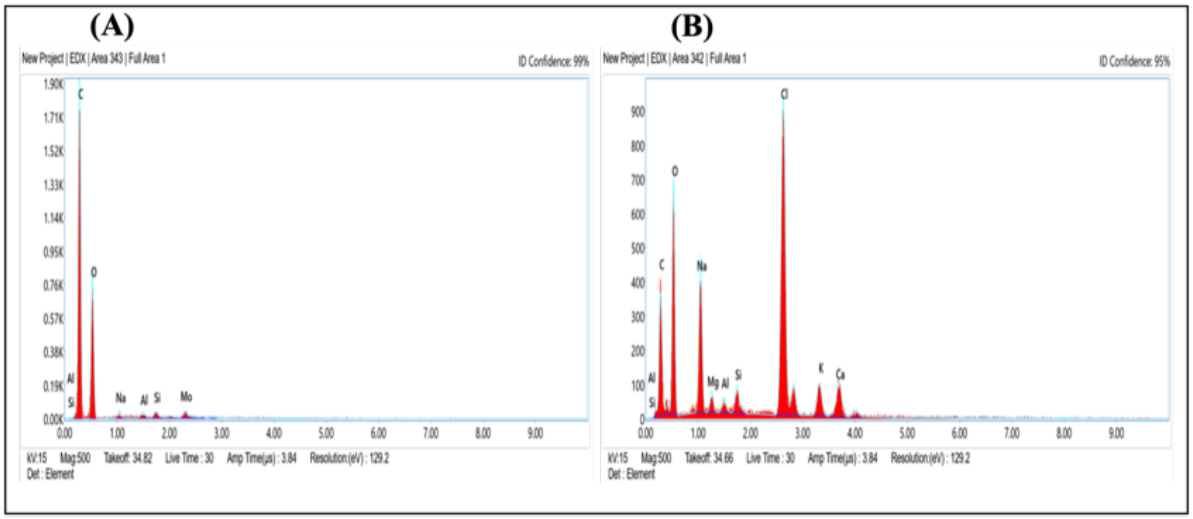


Figure S4: Characterisation of GO and GO-coated surface by EDAX. (A) EDAX analysis of GO and **(B)** EDAX analysis of GO-coated surface.

GO coating concentration optimization for F-MSA-C6

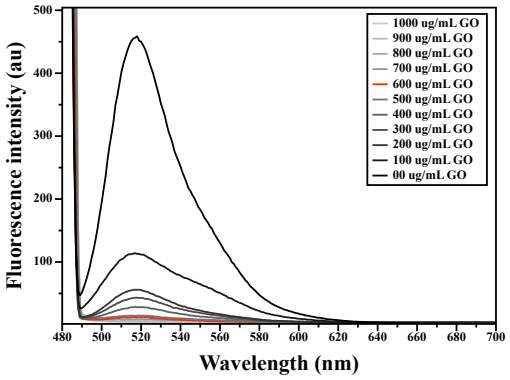


Figure S5: The GO concentration optimization to coat the PP surface to efficiently adsorb F-MSA-C6.

Assay Validation and cross-validation:

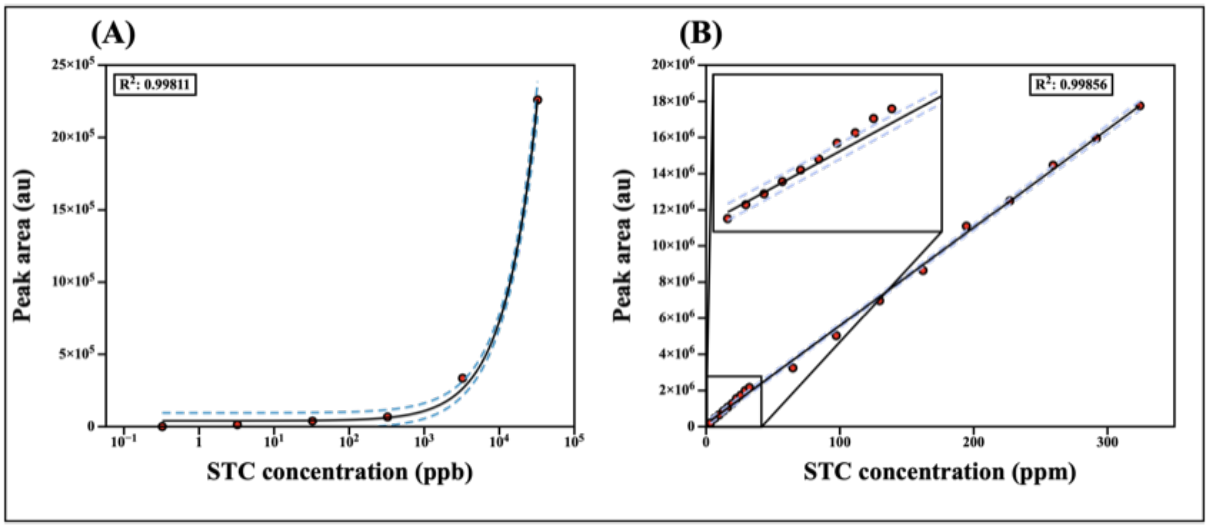


Figure S6: Assay validation and cross-validation. (a and b) calibration curve of STC HPLC analysis (n=2).

Table S3: Precision studies in simulated contaminated samples using developed GO-FAM-FRET aptasensor.

STC concentration (ppb)		Intraday			Interday		
		Mean *	SD	CV (%)	Mean *	SD	CV (%)
Pepper	32.42	34.1	1.41	1.99	32.6	1.78	3.15
	324.3	323	2.83	7.98	329	4.51	20.3
Chili	32.42	34.3	1.36	1.84	35.4	2.01	4.03
	324.3	329	2.06	4.23	333	2.08	9.49

* Mean of three replicates

References

1 K. Setlem, B. Mondal, S. Ramlal and J. Kingston, *Frontiers in Microbiology*, 2016, **7**, 1–14.

2 S. W. C. Chung and A. H. T. Wu, *Food Additives and Contaminants - Part A Chemistry, Analysis, Control, Exposure and Risk Assessment*, 2020, **37**, 355–362.

3 T. Yoshinari, H. Takeuchi, M. Kosugi, M. Taniguchi, M. Waki, S. Hashiguchi, T. Fujiyoshi, Y. Shichinohe, M. Nakajima, T. Ohnishi, Y. Hara-Kudo and Y. Sugita-Konishi, *Food Additives and Contaminants - Part A Chemistry, Analysis, Control, Exposure and Risk Assessment*, 2019, **36**, 1404–1410.

4 M. Oplatowska-Stachowiak, C. Reiring, N. Sajic, W. Haasnoot, C. Brabet, K. Campbell, C. T. Elliott and M. Salden, *Analytical and Bioanalytical Chemistry*, 2018, **410**, 3017–3023.

5 M. Z. Hossain and T. Goto, *Mycotoxin Research*, 2015, **31**, 17–22.

6 A. Veršilovskis, S. De Saeger and V. Mikelsone, *World Mycotoxin Journal*, 2008, **1**, 161–166.

7 L. Xu, G. Fang, M. Pan, X. Wang and S. Wang, *Biosensors and Bioelectronics*, 2016, **77**, 950–956.

8 J. M. Liu, F. Z. Cao, G. Z. Fang and S. Wang, *Polymers (Basel)*, , DOI:10.3390/polym9070299.