Supplementary Information (SI) for Analyst.

Supplementary Information: Molecular Imprinting of Aptamer/Carbamazepine Complexes for the Development of an Optical Nanosensor.

Iqra Salim[a], Ellie Richards [a] and Jon Ashley *[a]

Abstract: Monitoring anti-seizure medications (ASMs) such as carbamazepine is important to ensure that the correct dosage is given to patients which provides the maximum therapeutic effect. However, therapeutic windows for carbamazepine are narrow and need to be highly tailored towards the patients. Therefore, there is a need for more precise analytical methods for the monitoring of ASMs. Here we report a new hybrid aptamer/nanoMIP optical nanosensor utilising fluorescence quenching for the detection of carbamazepine. The nanosensor relies on a co-operative based binding mechanism whereby when the aptamer binds to carbamazepine it undergoes structural switching to change its 3D conformation and binds to the nanoMIP. Using a solid-phase imprinting technique, we synthesized nanoMIPs which recognise and bind to the aptamer/carbamazepine Complexes. The resultant nanoMIPs can then selectively recognise and bind to the aptamer complex resulting in a switch-off signal. The sensor demonstrated a LOD of 12.7 nM, excellent sample recoveries in 50% human serum (around 95%).

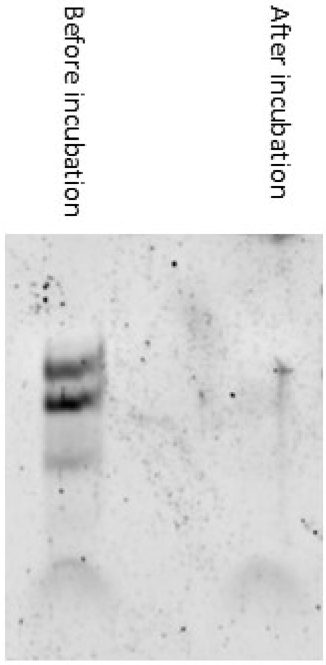

Figure S1 Representative native gel analysis (5% polyacrylamide gel) of carbamazepine aptamers before and after incubation with epoxide functionalised glass beads. Different bands represent different aptamer conformations.

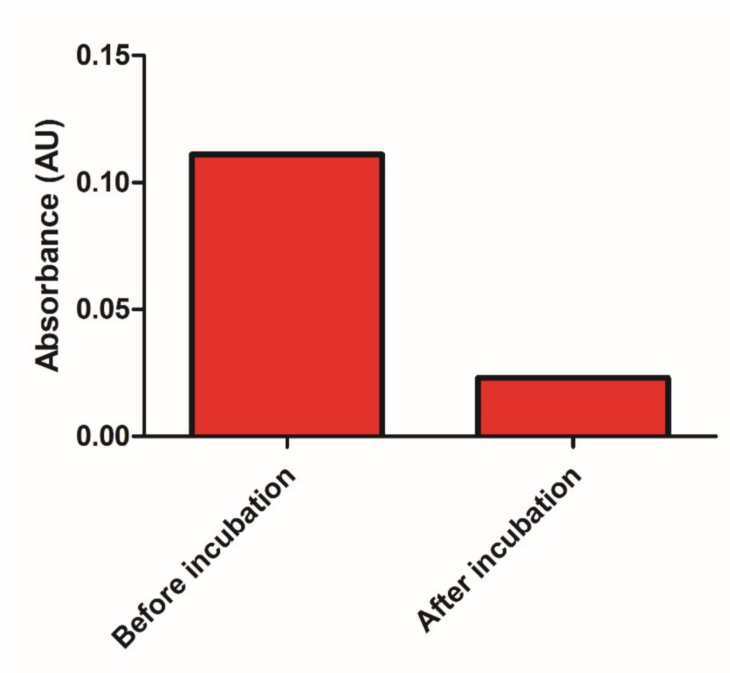

Figure S2 UV absorbance at 285.5 nm for carbamazepine (before and after incubation with aptamer functionalised glass beads.

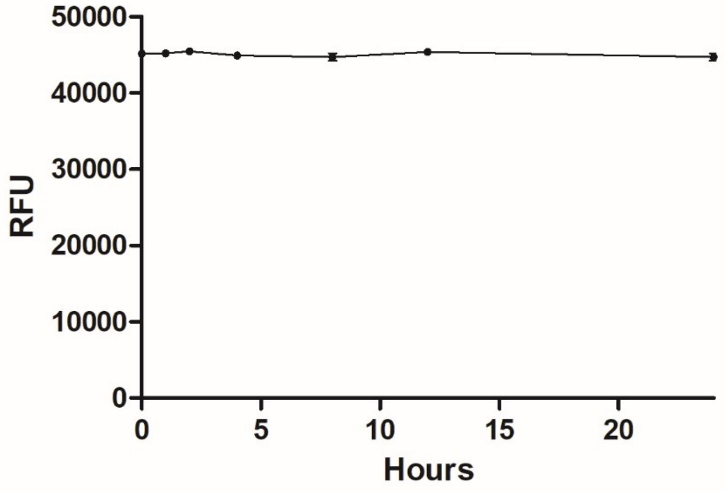

Figure S3 Stability of Fluorescence signal of nanoMIPs over 24 hrs (n = 2, %CV: 0.99%).

Method	Typical matrix	Sensitivity LOD	Recovery / accuracy (reported)	Reference
This work		LOD: 12.7	95 – 96.1 %	This work
Electrochemical aptasensor	Buffer; human serum; diluted finger-prick blood	LOD: 1.25 nM	76.1–125.9% (5 min) and 82.7–114.5% (30 min)	Chung, et al. (2022)[1]
Electrochemical MIP sensor (screen-printed carbon electrode; DPV)	Tap/river/sea water	LOD: 0.58 µM	89.4 – 115.9%	Poza-Nogueiras et al. (2026)[2]
UHPLC–MS/MS (environmental monitoring method)	Surface water / wastewater	LOD: 0.42 nM	77–160%	Koziarska et al. (2025)[3]
LC–MS^3 (therapeutic drug monitoring method)	Human plasma	LOD: 2.12 µM	100.7–110.5%	Ma et al. (2022)[4]
SERS (surface-enhanced Raman)	Saliva (treated)	LOD: 1.26 nM	not reported	Chen et al. (2021)[5]

Table S1 A comparison of different methods used to detect and quantify carbamazepine.

References

1 S. Chung, N. K. Singh, V. K. Gribkoff and D. A. Hall, *ACS Omega*, 2022, **7**, 39097–39106.

2 V. Poza-Nogueiras, A. Puga, J. G. Pacheco and C. Delerue-Matos, *Talanta*, DOI:10.1016/j.talanta.2025.129128.

3 M. Koziarska, M. Strzebońska and E. Szalińska, *Sci. Rep.*, 2025, **15**, 1–17.

4 D. Ma, Z. Ji, H. Cao, J. Huang, L. Zeng and L. Yin, 2022, DOI: 10.3390/molecules27041224.

5 N. Chen, Y. Yuan, P. Lu, L. Wang, X. Zhang, H. Chen and P. Ma, *Biomed. Opt. Express*, 2021, **12**, 7673.