

## Assessment of Analyte Trapping in Paper Matrices and its Effect on Sensor Performance

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Paper based sensors have found application in the clinical, environmental, and veterinary monitoring sectors. Popular examples of paper sensors include lateral flow tests (LFTs) and paper based microfluidic devices. The popularity of these sensors lay in their ability to provide healthcare at remote testing sites in a simple and low cost manner. To improve the performance of this sensor, a better understanding of its operational mechanism is required. Fluids travel through unsized paper *via* a fiber-swelling and pore-filling mechanism [1], carrying along with it any analyte it may have contained. During this movement the analyte can become intertwined within the complex paper matrix, and as a result, is prevented from reaching the detection zone of the sensor. To date, little published work has reported on the extent of analyte trapping in paper sensors, and the effect this has on the sensor performance. In this work, the movement of *Escherichia coli* (*E.coli*) through a lateral flow test is monitored by labelling the bacteria both colorimetrically and fluorescently. For colorimetric analysis, bacteria were stained red. RGB colour profiling (Fig. 1) was then used to identify bacteria entrapment along the LFT. Fluorescent imaging (see image insert in Fig. 1) was used to assess *E.coli* trapping in the membrane section of the LFT. Results indicate little to no trapping along the membrane. Scanning electronic microscopy analysis of the sample pad, conjugate pad, test line and control line were performed (image inserts in Fig. 1). The analysis indicates that significant *E.coli* entrapment occurs in the lower region of the LFT, and hence bacteria are prevented from reaching the test line and undergoing detection. The constituent sections of a LFT were separated and gently vortexed in buffer in order to elute and count the number of trapped bacteria in each section. The results are displayed in Fig. 1. The results of all these studies serve to confirm that the amount of analyte reaching the test line for detection is lower than that originally contained in the sample. A flow through test was performed to assess how the decrease in analyte concentration reaching the test line impacts the detection limit of LFTs. Flow-through tests have no sample or conjugate pads, and sample can be loaded directly onto the test line. This prevents any analyte losses prior to detection. The results indicate that entrapment does impact the sensor performance, especially when the detection limit of the sensor is lower than the amount bacteria that can become trapped. This can have serious consequences on the design of the sensor, especially for those sensors with detection limits lying in low concentration ranges.