

1. Fluorescence Detection with Different Core-Size Collecting Optical Fibers

Figure S1 shows measurements of fluorescence intensity collected with two different core-size optical fibers: 200 and 400 μm (black squares and red circles, respectively). Excitation power is adjusted in the two cases so as to have similar fluorescence signal intensity. Using a larger core size optical fiber presents the advantage of allowing larger sampling, which results in a reduced uncertainty.

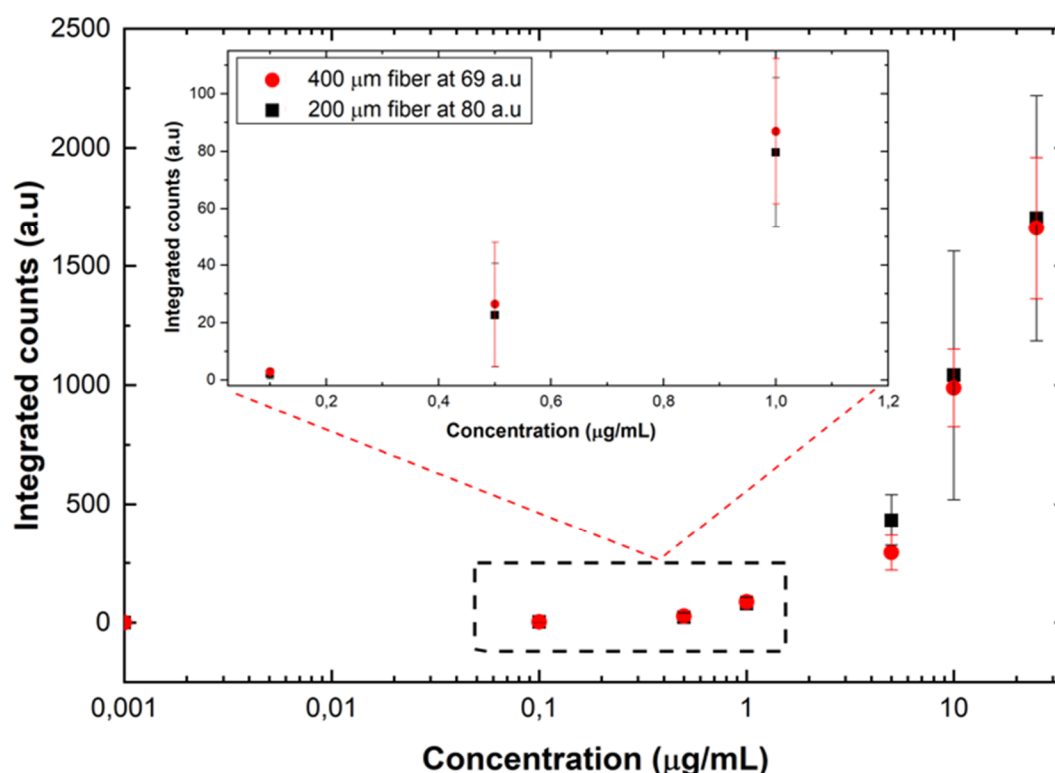


Figure S1. Calibration curve of fluorescence as a function of concentration of anti-IgG in a sandwich immunoassay with IgG on a designed planar antenna (as in Figure 4b in the manuscript) for a collecting optical fiber size-core of 400 μm (black squares) and 200 μm (red circles). In order to reach similar values of collected integrated fluorescence intensity, the excitation power (expressed in arbitrary units) must be higher for the smallest core size.

2. Photograph of the Entire Installation

The prototype version used for the measurements reported in the paper shows a total footprint of $100 \times 50 \times 60 \text{ cm}^3$, which can be easily reduced to $60 \times 30 \times 25 \text{ cm}^3$ considering realistic minimized dimensions for the translation stage and the terminal.

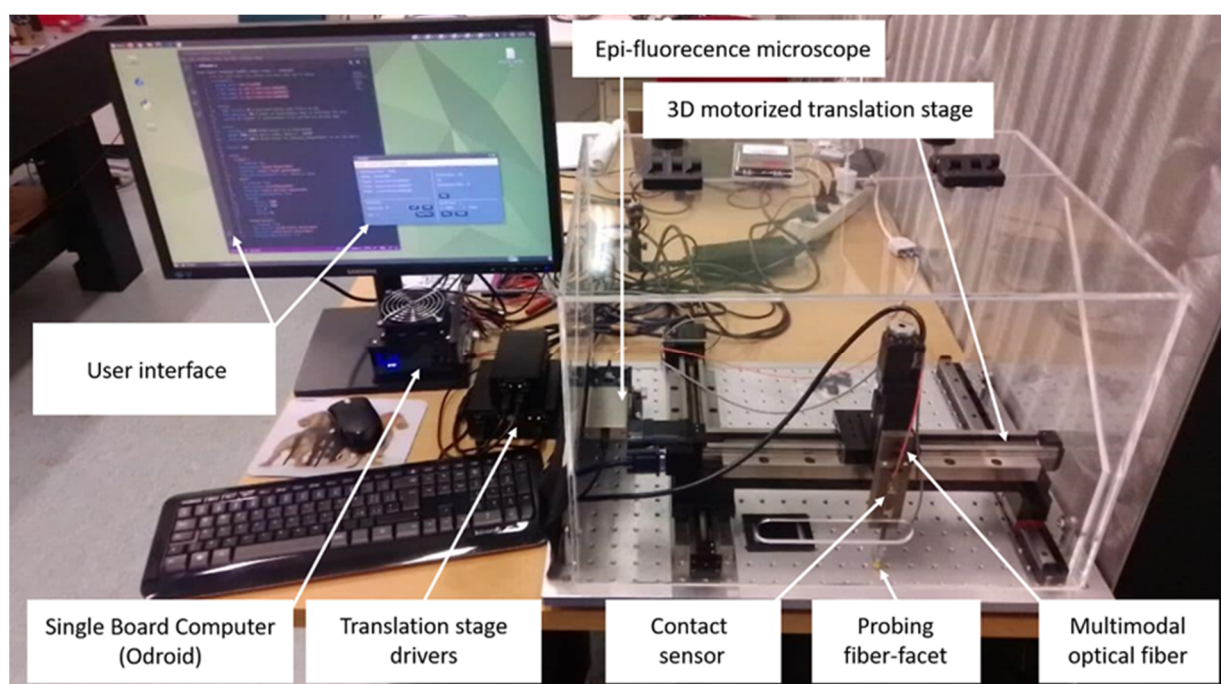


Figure S2. Image of the homemade prototype consisting of a compact epi-fluorescence microscope system, a 3D motorized translation stage, and a multimode optical fiber (everything inside the plexiglass box) and connected to the computer showing the user interface.

3. Reproducibility of the Measurements Performed with the Prototype

In order to demonstrate the reproducibility of the measurements, an arbitrary sample with a large spatial gradient of the CRP concentration, was measured 30 times. Figure S3 shows the number of counts and the standard deviation for each pixel in a 3×3 matrix of the measured sample (in particular, the centers of two adjacent pixels are separated by $500 \mu\text{m}$).

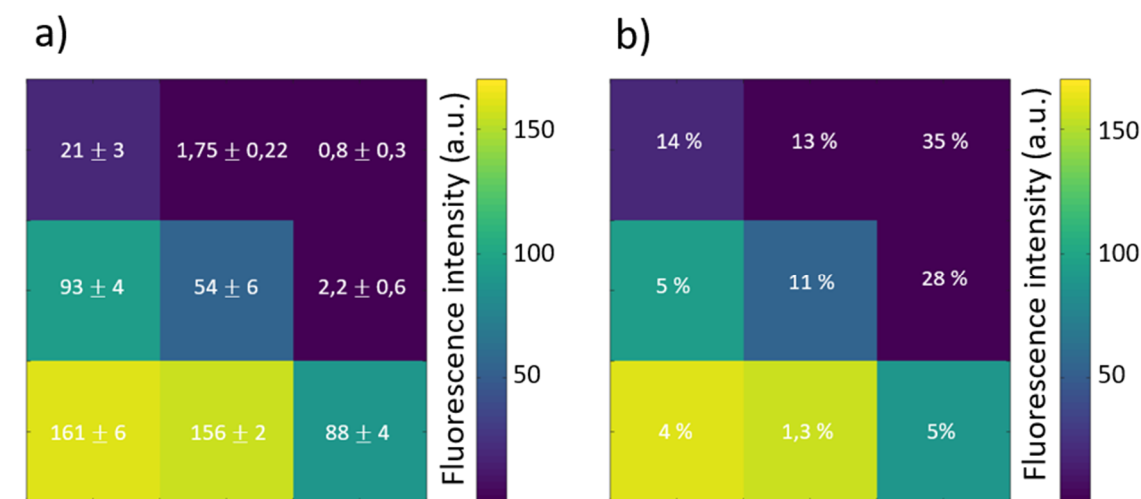


Figure S3. Fluorescence signal of an arbitrary sample in a 3×3 matrix comprising high and low fluorescence signal. (a) absolute values, defined as mean and standard deviation over 30 measurements, (b) percentage errors.

The analysis demonstrates the high reproducibility of the measurements. Indeed, both fluctuations in the illumination power and/or repeatability of the positioning would

result in a high variability of the collected fluorescence intensity. Even in the case of mapping a sharp edge (see Figure S3), we report a small uncertainty also for next neighbor pixels with a very different average signal.

4. Selection of the Area for Analysis: Comparison between Procedures

The selection of the central region of 6×6 pixels can be afforded manually or automatically. In the first case, it is cropped from a 15×15 pixel maps avoiding the so-called coffee ring shape of the dried drop (solid white square in Figure S4(a)). In the second case, the central area is given by a fix position inside the 15×15 pixels (dashed white square in Figure S4(a)). Figure S4(b) shows in black and red scatter points the average fluorescence intensity as a function of the concentration following the manual or the automatic procedure, correspondingly. For most of the concentrations, the automatic procedure gives higher values of fluorescence intensity, probably due to the fact that some pixels from the coffee ring shape are considered in a 6×6 pixel cropped area, increasing the total fluorescence. However, the trend with the concentration is maintained, indicating that the selecting procedure does not introduce a significant variation in the overall behavior.

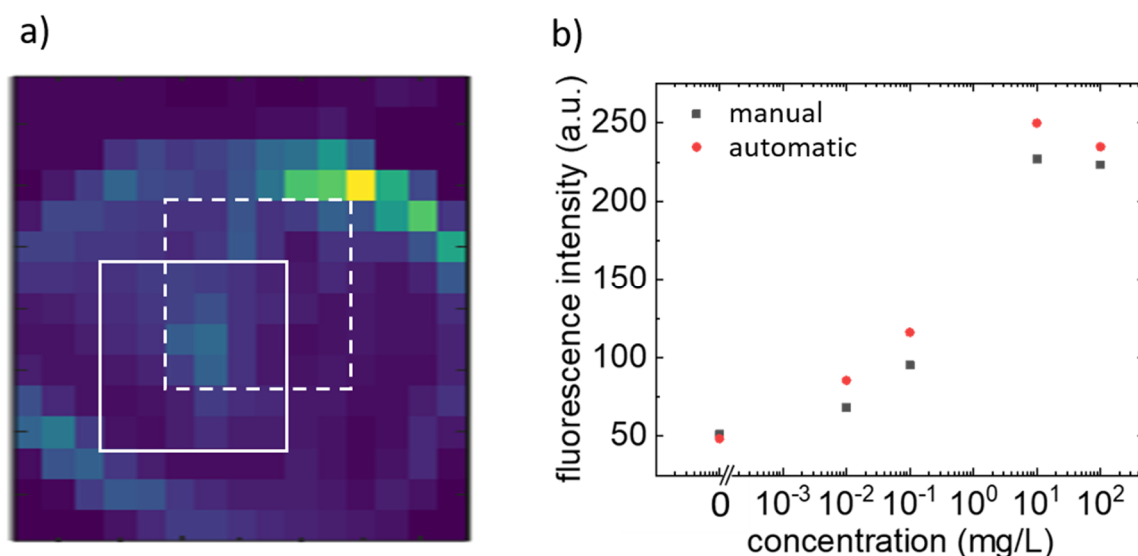


Figure S4. (a) Map of fluorescence intensity of a drop (15×15 pixels) showing the coffee ring shape. Solid and dashed white squares stand for the 6×6 pixel central regions selected manually or automatically, respectively. (b) Fluorescence intensity as a function of concentration using the manual (black squares) or the automatic (red circles) procedure to select the 6×6 central area.

5. Illustrative Fluorescence Intensity Maps of Inhomogeneous Assays

Figure S5 shows the colormap of the fluorescence signal for two different drops (a) low and (b) high concentration of CRP assay. The maps cover an area of 15×15 pixels (each pixel corresponds to $0.5 \mu\text{m}$) and central regions of 6×6 pixels are highlighted with a white square.

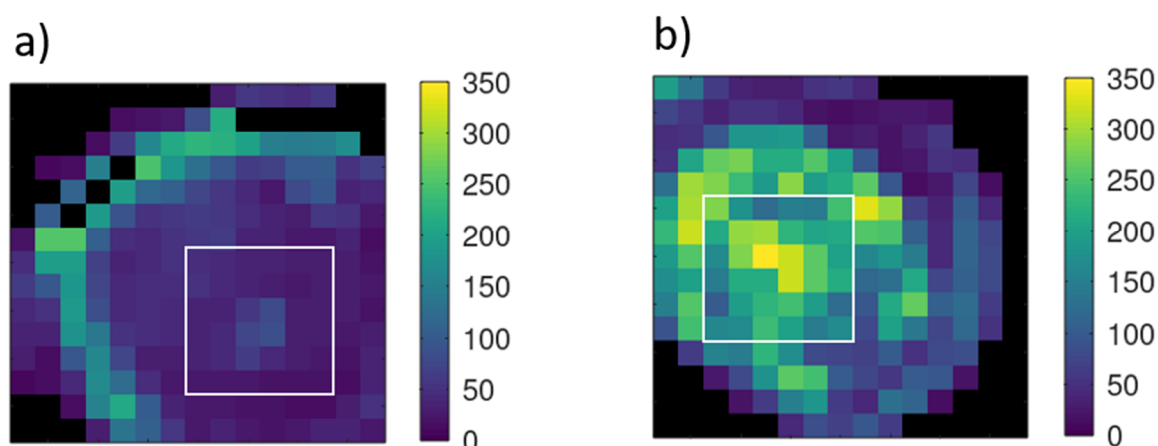


Figure S5. Illustrative fluorescence intensity maps for a drop of (a) low and (b) high concentration of CRP assay. Both maps show the coffee ring shape typical for dried drops. The difference in fluorescence intensity in the central areas between the two maps demonstrates the difference in concentration of the CRP assay.

In these fluorescence intensity maps, a preliminary filtering of the data is shown, in which pixels below a threshold intensity and outlier values were deleted (represented by black color in Figure S5). This kind of data processing can be useful for characterization and manufacturing quality control, for example to check the homogeneous distribution of the labelled antibody molecules on the spot, or to identify possible systematic non-specific binding that could lead to an unwanted systematic background.

6. Video of the Prototype Operation

Video S1 shows the prototype at work, in a typical operational sequence.