Improving Detection Limits in Lateral Flow Assays (LFAs) Using Photothermal Heating From Magnetic Nanoparticles and Thermochromic Sheets

Sean MacBride, Ivan Popov, Jennifer Lin, Grace Hymas, Amritpal Basi, Abigail Goh, Anja Rellstab, Wenhao Wu

Introduction

Lateral flow assays (LFAs) are diagnostic devices that show the presence or absence of a desired specimen and are often used in detecting pregnancy, specific diseases and the presence of toxic compounds [1]. They are especially designed for easy, quick & single use at point of need.

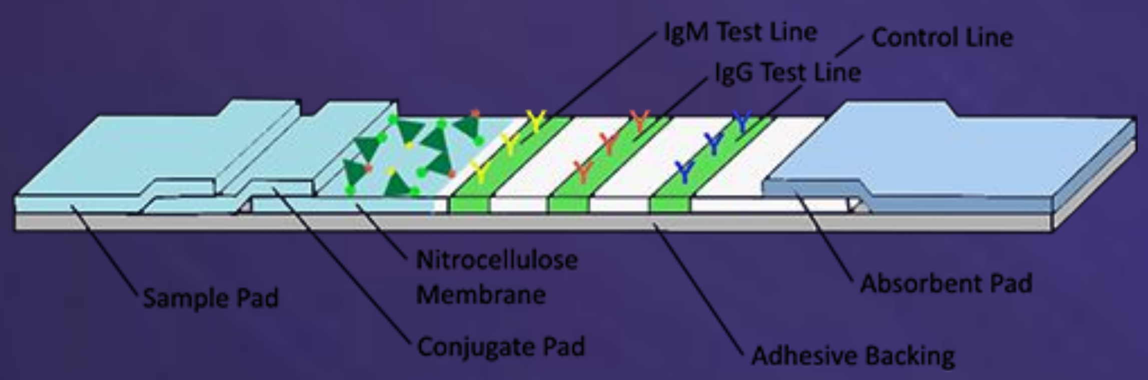


Fig. 1. The components of conventional LFAs.

The sensitivity of LFAs is not usually good, and this is what we hope to improve in our experiment. We simplified this complex LFA to one simply comprising of a nitrocellulose membrane and some deposited nanoparticles.

<u>What Are NANOPARTICLES?</u>

Nanoparticles are microscopic particles with high surface area to volume ratios. They provide a strong driving force for the diffusion process at high temperatures. Magnetic nanoparticles have high magnetic susceptibility and interact with external magnetic fields. Metallic nanoparticles absorb light efficiently (e.g. from a laser) which promotes electrons to excited states. When returned to the ground state, heat is released.

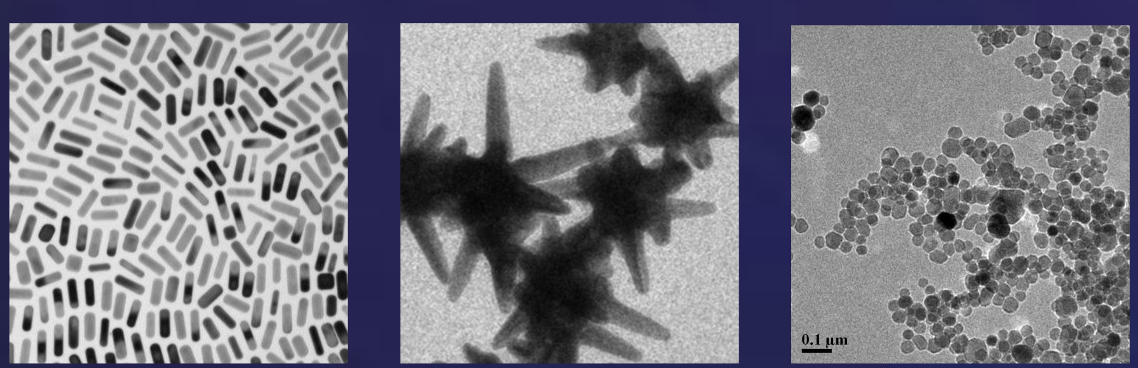


Fig. 2. Images of various types of metallic nanoparticles a. (left) NanoRods, b. (middle) NanoStars and c. (right) spherical nanoparticles.

<u>Our Experiment</u>

We aim to find the lower limit of detection of Iron Oxide and Zinc Ferrite magnetic nanoparticles deposited onto nitrocellulose membranes of different porosities.

This is possible by detecting the temperature change of the membrane when irradiated with a laser both qualitatively and quantitatively.

Methods

The experiment used BIO-RAD nitrocellulose membranes with pore sizes of 0.45 μm and 0.2 μm, and two types of magnetic nanoparticles (Iron Oxide & Zinc Ferrite) in large and small sizes. The nanoparticles were diluted with water to produce solutions of various concentrations, then loaded to the membranes using a pipette. The experiment was then set up as shown below:

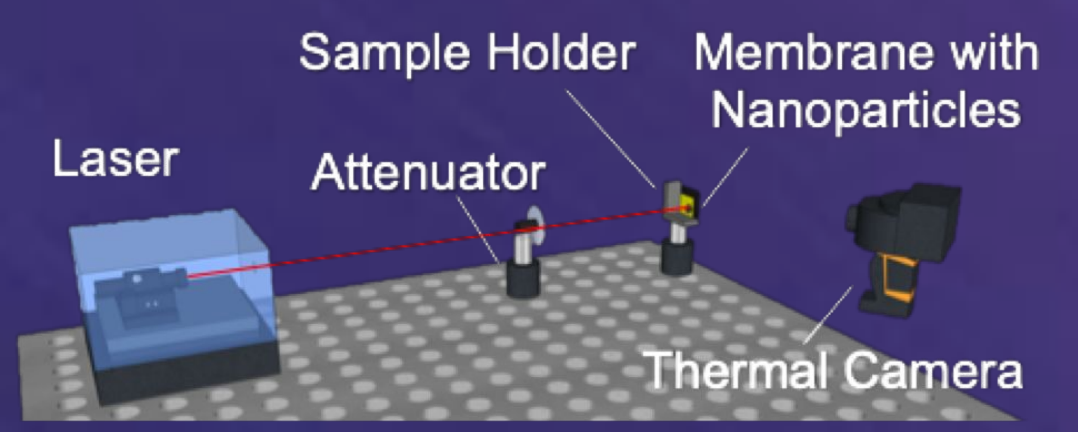


Fig. 2. Experimental set up for measurement of increase in temperature of magnetic nanoparticles deposited onto a membrane when exposed to a laser.

Two ways of monitoring temperature were developed. The first attached thermochromic sheets to the back of the membrane, which changed colour according to temperature, yielding a qualitative temperature change. The other used the Testo 875-1i infrared thermal camera which outputted a quantitative temperature. Readings from this method were taken every 30 seconds so the temperature trend could be seen over time. The process was repeated for membranes both with varying nanoparticles and concentrations.

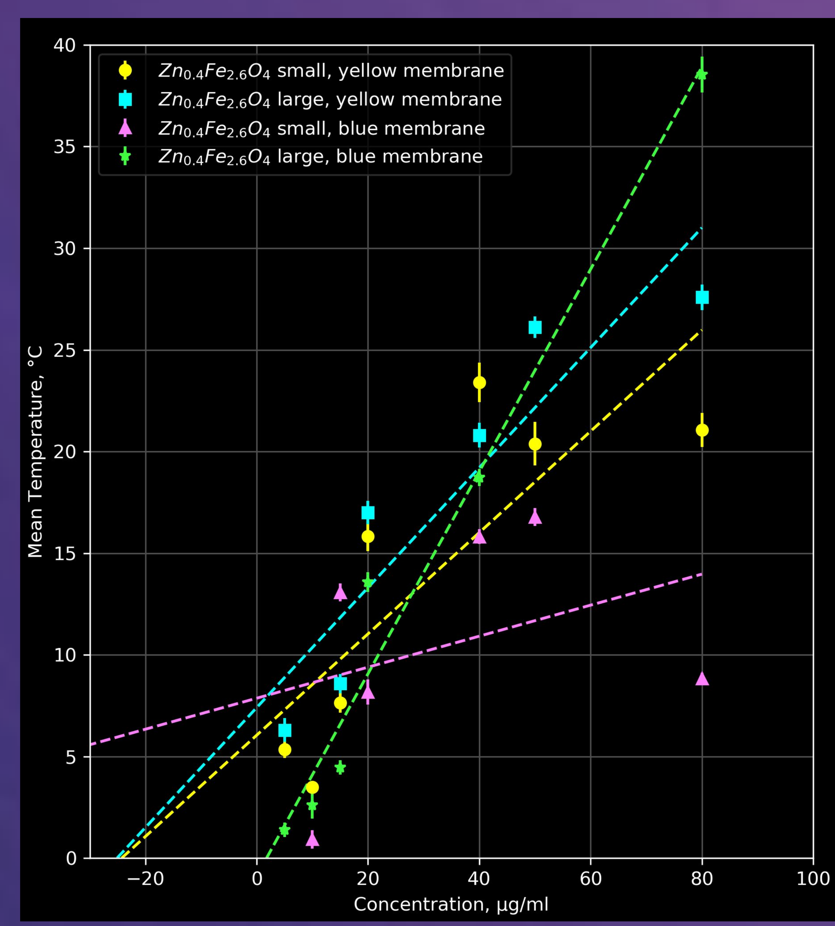
The test results were then analysed and compared with the control group to see if and how the concentration of nanoparticle solutions were correlated to the membrane's temperature changing rates.

<u>Result Quantification and Future Applications</u>

Colorimeter Software is already widely available on iOS and Android and returns very accurate colour values. This allows a remote user to quantify the colour of an LFA test, identify the concentration of analyte in the sample and therefore diagnose the severity of, for example, their disease.

Results

Our results were outputted and plotted using Python. We aimed to see the concentration at which mean temperature was zero - the limit of concentration. The trend between nanoparticle concentration and temperature change initially was unclear, so we attempted to plot a least squares line of best fit:



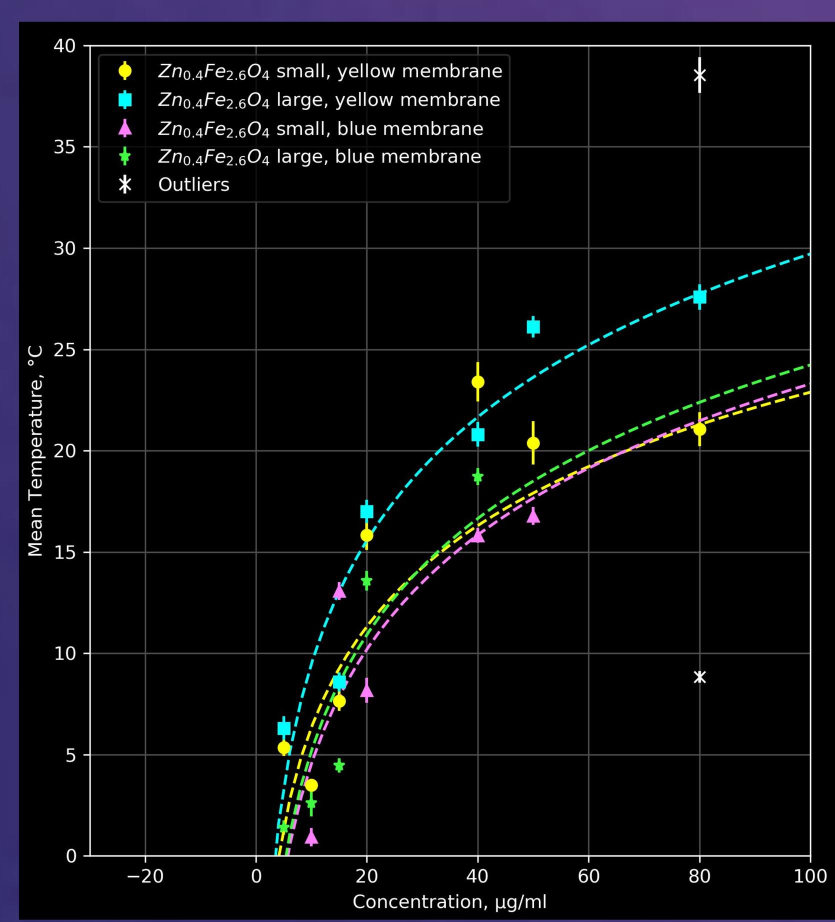


Fig 3. Variation in nanoparticle mean temperature with concentration for membranes of varying porosity with a. (top) a least squares line of best fit plotted and b. (bottom) an optimised logarithmic fit plotted.

We calculated chi-square values for our lines to see how well they fitted our data. Unfortunately - it was not well! The limit of concentration was also impossibly below zero. Therefore we tried some other trends, including a logarithmic fit. This gave us slightly more optimal chi-squared values, although still not perfect.

Conclusion

Thermal imaging was able to investigate the temperature change of the sample tested after laser exposure however it proved largely inconclusive. A logarithmic fit to the experimental data does show a potential correlation between temperature change and either size of nanoparticles or concentration of nanoparticle solution.

Solution depositing should be standardised for further experiments, and larger sample size should be. As the sample size was too small, a statistical conclusion of the detection limit could not be drawn and further experimentation is needed. Despite this, thermally contrasted LFAs are a potential avenue to research further and these results provide promising preliminary data.

Data taken using the infrared camera results used in conjunction with an application which runs a colour calibration curve is the most promising future prospect in quantifying LFA results and should be investigated as the next step in providing users with comprehensive and quantitative information from a quick and simple LFA test.

[1] G. Posthuma-Trumpie, J. Korf and A. van Amerongen, "Lateral flow (immuno)assay: its strengths, weaknesses, opportunities and threats. A literature survey", Analytical and Bioanalytical Chemistry, vol. 393, no. 2, pp. 569-582, 2008. Available: 10.1007/s00216-008-2287-2 [Accessed 11 March 2019].