# Paper Microfluidic Platform for Detection of Viral Gastroenteritis

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**Introduction:** Noroviruses, belonging to the family *Caliciviridae*, are highly infectious agents that cause acute gastroenteritis in both children and adults worldwide; yet, access to routine diagnostic tests remains limited. The long-term goal of this project is to develop a rapid diagnostic system for detection of noroviruses that can be employed at the point-of-need. Our approach capitalizes upon recent advances in the fabrication of low-cost microfluidic devices using patterned paper as an instrument-free platform for sophisticated biological assays. These devices are known as microfluidic paper analytical devices (μPADs) or two-dimensional paper networks (2DPNs).[1, 2] In these devices, capillary action draws fluids through the channels eliminating the need for expensive pumps and fluid handling equipment found in traditional microfluidic devices.

**Materials and Methods:** In this study, the μPAD was fabricated in inexpensive cellulose fiber paper using a wax printing technique[3, 4] to pattern three fluidic inlets arranged in-line for sequential delivery of sample, wash buffer, and amplification reagents. Sandwich-type immunoassays were implemented within the μPAD channels for detection of clinically-relevant norovirus strains, such as GII.4 which is responsible for the majority of outbreaks in the US. Assay performance was characterized using recombinant viral antigens with nanoparticles detection reagents containing gold and/or magnetic functionality.

**Results and Discussion:** In the presence of ≥ 1 μg/mL viral protein, a positive test result was apparent through the formation of two red/pink spots at the target-specific capture location and control within the μPAD channel (Figure 1). While a binary yes/no test is sufficient for detection, the μPAD immunoassay spot intensity distinctly increased with increasing pathogen concentration and could be used with an analytical reader or scanner to generate a standard curve for more quantitative analysis (Figure 1). Subsequent silver/gold enhancement reactions have been used to lower the detection limit by roughly 5-fold (data not shown), while integration of custom multi-functional magnetic detection particles will enable concentration of pathogen prior to analysis, which is anticipated to push the limit of detection even further.

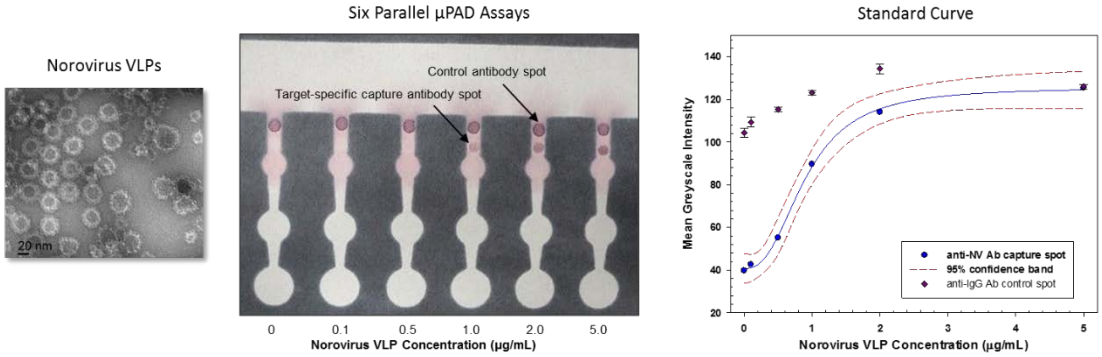


**Figure 1.** Paper-based microfluidic assay for detection of Norovirus GII.4 virus-like particles

**Conclusions:** With additional optimization and multi-plexing to other diarrhea-related pathogens (viral/bacterial/protozoan), it is expected that this research will establish a new platform for point-if-need testing that could positively impact clinical diagnosis and monitoring of diarrheal illness.

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**References:**

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