

# Electrochemical Immunosensor Platform for Fast Screening of Human Respiratory Pathogens

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Capability for rapid detection and identification of different types of respiratory virus infections would greatly enhance point-of-care health care delivery.<sup>1</sup> This paper presents a flow-through immunoassay technique coupled with an electrochemical sensor array for a multi-analyte screening of human clinical samples for respiratory viral pathogens. The solid phases of the flow-through system are a porous polymeric membrane on which the immunochemical reaction occurs and porous working electrodes where the product of the enzymatic reaction is detected.

Intermittent Pulse Amperometry is used as a detecting technique for sandwich immunoassay through measurements of the electro-reduction current of TMB<sup>+</sup> formed from TMB in a catalytic cycle involving HRP label, H<sub>2</sub>O<sub>2</sub>, and TMB.

The system is capable simultaneous detection and identification of influenza A (IA H1N1 and H3N2 strains), influenza B (IB) and RSV viruses in liquid nasal swab samples from patients with no sample extraction or clean-up procedures. The format of the device allows separation and concentrating of virus particles and accumulating product of enzymatic reaction close to the surface of a working electrode without it being swept away in the flow of liquid through the device. This method allows detection of viral pathogens in clinical samples within 20 minutes while simultaneously determining 8 different antigens. The method demonstrates also good agreement with a standard direct fluorescent antibody (DFA) clinical method.<sup>2</sup>

An example of the described multichannel/multianalyte flow-through immuno-sensing approach is shown in Fig. 1. There are different combinations of immobilized antibodies in equivalent amounts in each channel: channels 1 and 2 had anti- IA, IB and RSV; channels 3 and 4 had anti- IA and IB; channels 5 and 6 had anti-IB and RSV; and channels 7 and 8 had anti-IA and RSV antibodies. One RSV positive and one negative human clinical samples were used as a model “unknown” samples. Only channels that have RSV specific antibodies (1, 5 and 7) gave positive sensor responses with statistically approved difference to nonspecific channels (2-4, 6 and 8).

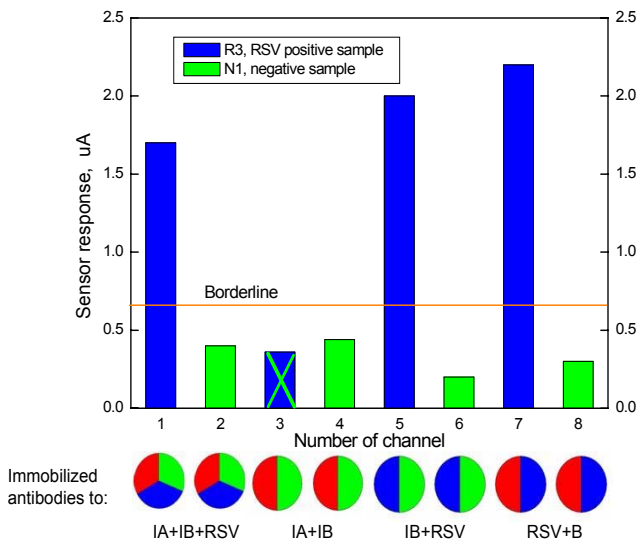
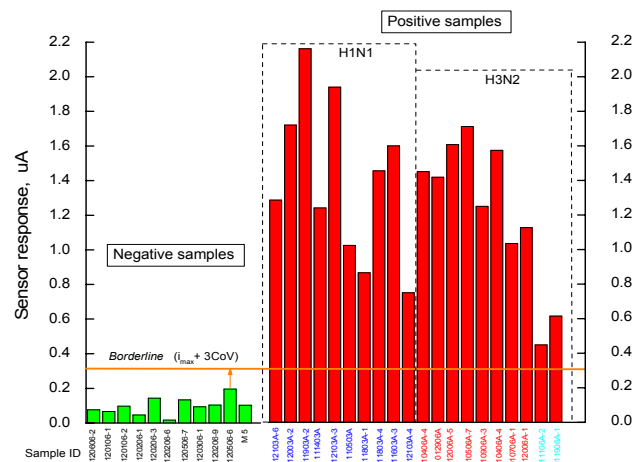


Figure 1. Screening of human clinical samples for RSV

This multi-plexed antibody approach in the 8-channel immunosensor platform will allow extending of number of detected viral respiratory analytes up to 12 in one run.

The multichannel flow-through electrochemical immunosensor was also evaluated using 31 clinical samples: 11 were negative for influenza, 10 were positive for influenza A H1N1 and 10 were positive for H3N2. The data shown on Fig. 2 establishes a influenza A positive discrimination threshold using



clinical samples. No false positive or false negative results were observed.

Figure 2. Detection of Influenza A virus in human clinical samples.

## References:

1. Ellis, J. S., and Zambon, M. C. (2002), *Rev Med Virol.* 12, 375-389.
2. Holland, C. A., and Kiechle, F. L. (2005), *Curr Opin Microbiol* 8, 504–509.

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