VIRUS INFECTIVITY DETECTION BY EFFECTIVE REFRACTIVE INDEX USING OPTOFLUIDIC IMAGING
P. Y. Liu\textsuperscript{1,2}, L. K. Chin\textsuperscript{2,†}, W. Ser\textsuperscript{2}, T. C. Ayi\textsuperscript{3}, P. H. Yap\textsuperscript{3}, T. Bourouina\textsuperscript{4} and Y. Leprince-Wang\textsuperscript{1†}
\textsuperscript{1}Université Paris-Est, UPEM, F-77454 Marne-la-Vallée, France
\textsuperscript{2}School of Electrical and Electronic Engineering, Nanyang Technological University
Singapore 639798
\textsuperscript{3}Defence Medical & Environmental Institute, DSO National Laboratories, Singapore 117510
\textsuperscript{4}Université Paris-Est, ESYCOM, ESIEE Paris, F-93162 Marne-la-Vallée, France

ABSTRACT
This paper presents an optofluidic imaging system to detect influenza virus infection via co-culture of Madin Darby Canine Kidney (MDCK) cells. Influenza flu virus is a serious threat that can cause contagious infections in people in epidemic proportions. Hence, it is crucial to accurately detect and understand the morphological changes that occur in the cells when infected by the influenza virus. Recently, researchers are investigating the biophysical properties of cells and correlating them to biomedical conditions. For example, a decrease in refractive index (RI) is observed in bacterial infected cells \cite{1}. In this paper, an optofluidic imaging system is developed to observe the change of RI in virus infected cells based on scattering signature.

KEYWORDS: Optofluidics, Virus infectivity, Scattering signature

INTRODUCTION
Figure 1 shows the biological process of influenza virus infection model. The influenza virus enters the surface membrane cells of the lung and throat by endocytosis. The virus RNA, accessory proteins and RNA polymerase are released into the cytoplasm of the cells. A virus complex is formed and carried into the cell nucleus. The virus RNA replicates itself in the cell nucleus and creates new influenza virus particles. When the RNA particles are increased in the infected cells as compared to uninfected cells, this leads to the change in the
refractive index in the nucleus and can be used as an indicator for detection. Optofluidic refractometers have been developed extensively [2-4], and in this paper, the change of refractive index is determined by observing the scattering signature of infected and normal cells.

**WORKING PRINCIPLES**

Scattering signature has been exploited to determine the state of biological samples, for example the infection of bacteriophage on *Escherichia coli* [5]. When MDCK cells are infected by influenza virus, the multiplication of RNA particles increases the effective refractive index of the cells. This change can be reflected by measuring the scattering image of the MDCK cells at different time points after infection. In the experiments, different plates of MDCK cells are cultured and infected by influenza virus. At each time point of 1 hr, a plate of MDCK cells are trypsinized and injected to the optofluidic chip to capture the bright field and scattering images of the MDCK cells. Microfluidics facilitate the alignment of the MDCK cells with the incident laser for capturing of the scattering image as shown in Figure 2. The images are subsequently analyzed to determine the size of the MDCK cells (bright field) and the distance of 1st scattering peak from the center (scattering).

**EXPERIMENTAL RESULTS AND DISCUSSIONS**

Figure 3 shows an example of the scattering image of normal and infected MDCK cells. MDCK cells with the incident laser for capturing of the scattering image as shown in Figure 2. The images are subsequently analyzed to determine the size of the MDCK cells (bright field) and the distance of 1st scattering peak from the center (scattering). Figure 4 shows the statistical measurements of normal and infected MDCK cells at 0 hr and 5 hrs after infection. For normal MDCK cells, the positions of 1st peak are relatively similar. However, the positions of 1st peak are significantly decreased in infected cells after 5-hr infection. This is
correlated with the fact that infected cells have increased refractive index, and subsequently narrower scattering bands.

CONCLUSIONS
In conclusion, a different approach to detecting virus infection has been presented. The results show that refractive index changes due to the changes in the nucleus of the cells when MDCK cells are infected by the influenza virus. It is imperative that a direct detection method can be conceived to monitor the changes in the lung and throat to allow early detection and treatment of the influenza flu virus.

ACKNOWLEDGEMENT
The authors would like to acknowledge the financial support from Environmental and Water Industry (EWI) Development Council of Singapore (Grant No. 1102-IRIS-05-02).

REFERENCES

CONTACT
† L. K. Chin; phone: +65-6790 6532; lkchin@ntu.edu.sg
Y. Leprince-Wang; yamin.Leprince@u-pem.fr

Figure 4: Significant shift in the position of 1” peak is observed after 5 hrs of virus infection.