Use of fluorescently-labeled nanoparticles to study the sterile filtration behavior of therapeutic viruses – the critical effect of surfactant concentration

Mohammadreza Pazouki, Noelle Wilton, David R. Latulippe

Department of Chemical Engineering, McMaster University, Hamilton, Ontario, Canada

Viruses engineered for gene therapy and the destruction of tumor cells (i.e. oncolytic viruses) are a growing market with profound implications for human health. For example, in 2015 the FDA approved the first ever oncolytic virus-based therapy for the local treatment of melanoma lesions. One of the main challenges in therapeutic virus manufacturing processes is the dead-end sterile filtration step that is highly desirable from a safety and regulatory perspective. While numerous studies have explored the use of virus-filtration membranes to selectively retain viruses and allow smaller biotherapeutic molecules (e.g. monoclonal antibodies) to pass through the membrane, few have focused on the transmission of viruses through sterile filtration membranes. A recent study led by our group evaluated the effects of microfiltration membrane morphology on the filtration behavior of Rhabdovirus Maraba [1], a bullet-shaped, enveloped virus that has been shown to be a promising oncolytic virus [2].

Due to the numerous challenges associated with running filtration studies with actual therapeutic viruses, the use of nanoparticles as virus surrogates has been widely reported in virus retention studies [3–5]. Although the effect of adding surfactants for stabilization of nanoparticle suspension is often reported in different applications, there is a large variation in the amount and type of surfactants that are used. Hence, it was determined that a detailed study on the role of different surfactants and surfactant concentration would benefit this field. In this study, fluorescent polystyrene nanoparticles (PNPs) were used as virus surrogates to investigate the performance of two sterile filtration membranes (Durapore PVDF and MiniSart PES) with 0.22 micron pore size ratings. Three different sizes of fluorescent PNPs (from Spherotech) with average sizes of 58, 182, and 571 nm (determined via dynamic light scattering) were compared in a dead-end constant flux filtration process. These nanoparticles were selected to represent the width, length and relative aggregate dimensions of the Rhabdovirus Maraba, respectively. The concentration of Tween 20 in the feed buffer solution (0.1 M carbonate with 0.1% (w/v) sodium azide) was varied from 0% to 0.1% (v/v) for each nanoparticle-membrane combination. We discovered that there exists a critical surfactant concentration (CSC) at which the transmission of the 58 and 182 nm fluorescent PNPs was dramatically altered from near complete transmission to complete retention. To the best of our knowledge, this effect has not been previously reported in the membrane literature. For the MiniSart PES membrane, the CSC for Tween 20 was found to be approximately 0.002% (v/v). Scanning electron microscopy images were obtained using different post-filtration membranes to demonstrate the dramatic shift in the transmission behavior of the sterile filtration membranes.

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