ADVANCED PROCESS CONTROL STRATEGIES FOR CONTINUOUS INFLUENZA VIRAL PARTICLE PRODUCTION

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Key Words: Continuous Manufacturing, Continuous Viral Vaccine Production, Advanced Control Strategies

Influenza presents a significant public health burden globally with an estimated 3 to 5 million severe cases and 300,000 to 650,000 fatalities annually [1]. Vaccines are currently one of the most important tools available to healthcare professionals to address this problem. Most influenza vaccines are currently manufactured as virus particles generated using embryonated chicken egg-based processes. This established process has multiple limitations such as complex logistics and supply chains arising from the need to procure large numbers of pathogen-free eggs that need to be simultaneously inoculated and maintained, lack of compatibility with certain influenza strains, and potentially lower vaccine efficacy due to higher frequency of mutations, for example [2]. These issues have motivated the development of cell culture-based manufacturing platforms for the influenza vaccine and this effort has seen commercial success with the launch of Flucelvax®. The shift to cell-based manufacturing platforms has also encouraged the development of continuous manufacturing which potentially offers improved product quality control and manufacturability.

Influenza, like many other viruses, can generate defective interfering particles (DIPs) in-vivo and in cell cultures. These DIPs interfere with standard virus particle (STV) reproduction and lower viral titers during production. DIPs also introduce complex and undesirable process dynamics such as oscillatory behavior which can impact product quality and steady-state operation [3]. DIPs can also be seen as a contaminant that necessitates further downstream purification to eliminate from the final product. Correspondingly, successful operation of a continuous viral particle production process requires strategies to maximize productivity while mitigating undesirable process dynamics and minimizing DIP content. While DIPs are largely seen as a nuisance, they are gaining attention as potential antiviral/antitumor therapies [4]. It is therefore of interest to see if the same bioreactor configuration can be adapted to effectively produce high purity DIPs as well. Currently known strategies to achieve successful operation such as optimizing cell lines and virus strains to minimize *de-novo* generation of DIPs and/or adopting novel bioreactor configurations are complex and costly. In contrast, it would be of significant interest if successful operation can instead be achieved through the use of process control on a well-established continuous bioreactor platform such as a two-stage continuously stirred tank bioreactor.
To explore the process dynamics and control of a two-stage continuously-stirred tank bioreactor, a dynamical model adapted from [3] is considered. The first stage serves as the cell bioreactor to provide the second stage virus bioreactor with a supply of fresh target cells. The dynamical model has six states which track four types of cells – target, infected with STVs, infected with DIPs, and coinfected – and two types of viral particles: STVs and DIPs. Structural analysis of the model is first carried out to characterize qualitative features of the system which demonstrate the presence of a Hopf bifurcation at low reactor dilutions which is associated with the oscillatory behavior. Subsequently, a preliminary control study was carried out by implementing a state feedback control law which demonstrates that it is possible to suppress undesirable oscillatory behavior by controlling the inlet target cell concentration of the feed stream to the second stage viral bioreactor. Since most of the states are not measurable, advanced process control strategies including nonlinear model predictive control are designed and compared. The simulation results predict that the two-stage bioreactor with such controls can produce high yields of either STVs or DIPs.

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