Vaccination remains the most effective way to prevent the infection with Influenza viruses. However, their constant antigenic drift implies that current human Influenza vaccines need to be annually updated with high inherent costs. Virus-like particles (VLPs) have become widely used as vaccine candidates because of their versatility, immunogenicity, and safety profile.

In this iBET project we are attempting to produce a candidate for a universal vaccine for which 35 different VLPs (mono, trivalent and pentavalent) were purified. Here we describe three recent advances on Influenza VLPs bioprocessing: two new analytical tools and the development of an integrated all filtration purification process, inserted in the “anything but chromatography” concept.

The first method is a label-free tool that uses Biolayer interferometry technology applied on an Octet platform to quantify Influenza VLPs at all stages of DSP. Human and avian sialic acid receptors were used, in order to quantify hemagglutinin (HA) content in several mono- and multivalent Influenza VLP strains. The applied method was able to detect and quantify HA from crude sample up to final VLP product with high throughput, real-time results and improved detection limits, when compared to traditional approaches, crucial for in-line monitoring of DSP.

Using a click-chemistry approach that involves Azidohomoalanine incorporation and functionalization, Influenza VLPs were selectively and fluorescently tagged. Taking into account that this chemical tag does not affect particle size, charge and biological activity we report here a valuable tool to online/at-line product monitoring during DSP optimization of virus related biopharmaceuticals. Moreover, using this tool coupled with FACS we were able to discriminate between VLPs and baculovirus, the major impurity of the system.

The proposed all-filtration process will be described, with special focus on the clarification stage, followed by multiple ultrafiltration and diafiltration steps to achieve the needed concentration and purity specifications. Using this all-filtration platform, we are able to speed up the time process, to improve the scale-up and to reduce costs due to the removal of chromatographic steps.