Virus-like particles (VLPs):

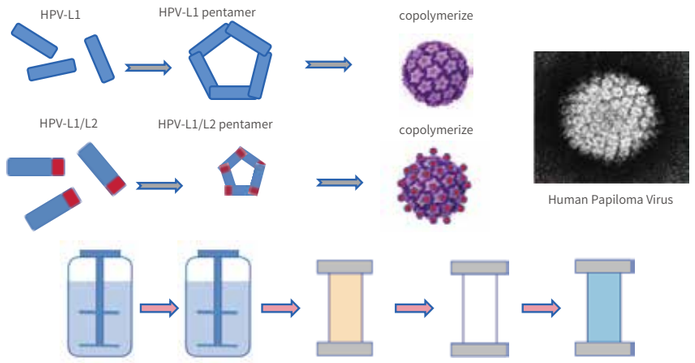
Virus-like particles (VLPs) are hollow particles composed of one or more structural proteins of a virus. They do not contain viral nucleic acids and cannot replicate autonomously, yet they are morphologically identical or similar to authentic viral particles.

Non-Encapsulated VLPs: These relatively simple VLPs are made of one or several capsid proteins and can be expressed in both prokaryotic and eukaryotic cells.

Encapsulated VLPs: These more complex VLPs consist of a matrix wrapped in lipid membranes containing glycoproteins derived from the host cell. They are commonly expressed in eukaryotic systems such as yeast, insect cells, and plant cells, requiring co-expression of several structural proteins for intracellular self-assembly.

VLPs exhibit strong immunogenicity and biological activity, making them crucial in disease prevention. A notable example is the HPV vaccine, which is prepared from recombinant HPV-L1 proteins expressed in E. coli or yeast. L1 proteins spontaneously form pentamers, which then copolymerize into VLPs.

The purification process involves lysis and clarification, followed by enrichment using one-step cation chromatography. Further purification is achieved using Smartarose 6FF, Smac CORE 700, or Butyl Beads 4FF, and finally removed by Smac Q40 or MMC.



HPV-L1 proteins' pentamer formation and copolymerization. We provide three Purification protocols for HPV vaccine

Nucleic acid vaccines:

Nucleic acid vaccines can be categorized into mRNA vaccines and DNA vaccines.

mRNA vaccines involve injecting the mRNA sequence encoding an antigen protein into the human body, where it directly expresses the antigen and induces an immune response. Since mRNA has no risk of infection, the preparation process is highly generic, requiring minimal optimization. This results in a shorter development cycle and strong immunogenicity without the need for adjuvants, making it suitable for emergency immunization.

DNA vaccines involve injecting a recombinant eukaryotic expression vector encoding a specific protein antigen directly into the animal body. The exogenous genes are expressed within the host, and the generated antigen activates the immune system, inducing specific humoral and cellular immune responses. Vaccines under development using this method include those for malaria, influenza, rotavirus, and HIV.

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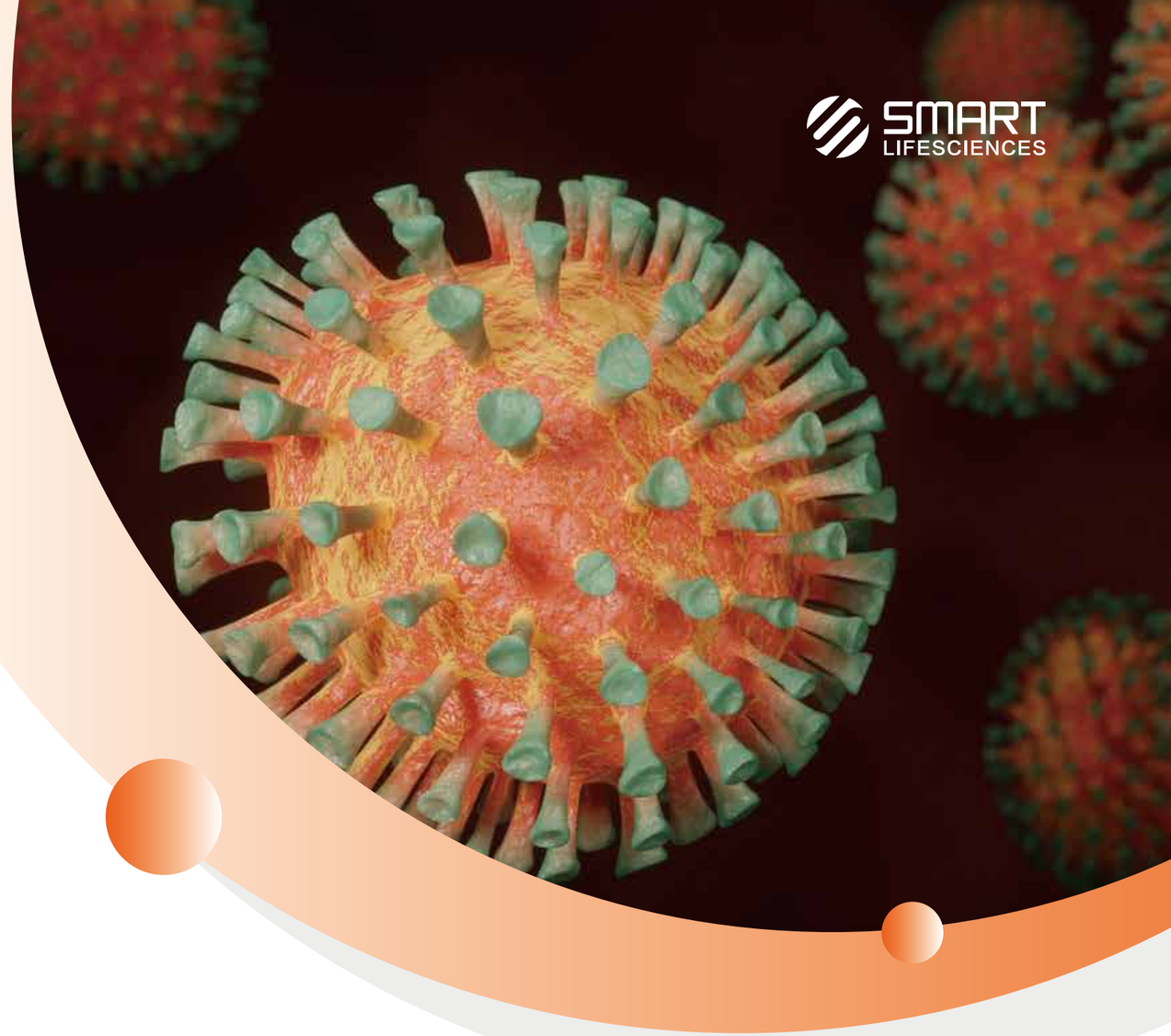


SMART LIFESCIENCES

Guide to Vaccines Purification

Professional Manufacturer
of Chromatography Resin

Vaccines

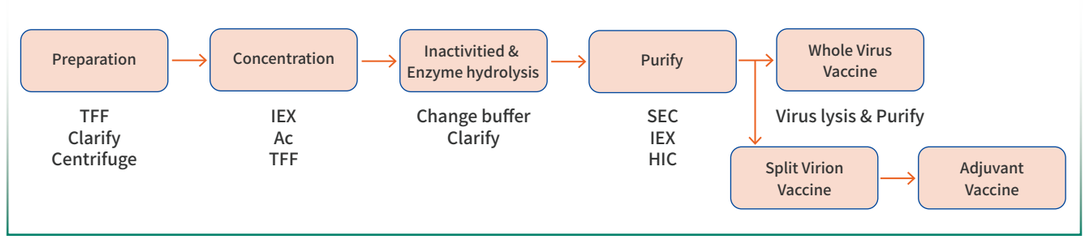
Vaccination is an effective way to protect people from harmful diseases. It utilizes the body's natural defenses to resist specific pathogens and enhance the immune system.

Vaccines stimulate the immune system to produce antibodies, mimicking the body's natural response to an actual disease. Types of vaccines include traditional vaccines based on inactivated and live/attenuated antigens (viruses or pathogens), live vectored vaccines, recombinant protein vaccines (especially VLPs), and nucleic acid vaccines.

Virus vaccine :

For viral antigens, traditional vaccines are obtained from whole viruses, while split vaccines are produced by further processing whole viruses to extract protein antigens. The purification of whole viruses is relatively straightforward, involving TFF concentration, gel filtration, and ion exchange chromatography (IEX). Common gel filtration chromatography includes Smartarose 4FF/6FF and dextrangel. IEX is widely used for virus enrichment.

Most impurities in the starting material originate from the culture medium and cell matrix. These impurities have potential health effects and must be strictly controlled. Q and hydrophobic interaction chromatography are effective in reducing HCP/HCD residues in viral stocks. Additionally, it is recommended to shorten the purification period to prevent the degradation of large cell debris and nucleic acids, as well as the adsorption of viral components, which can complicate the purification process.



Purification of Viral Vaccines

Whole virus vaccine, split virion & adjuvant vaccines:

Based on the whole virus vaccine, split virion and adjuvant vaccines have been developed. The purification process for split virion vaccines involves extracting and inactivating the whole virus, then splitting it into single protein molecules using appropriate methods. Subsequently, the lysate and some impurities are removed through further purification to obtain the monovalent stock.

Adjuvant vaccines are produced by adding the adjuvant to the split vaccine to further increase the immune titer. Taking influenza vaccine as an example, each variant follows a different development process.

step	Influenza vaccine, inactivated,avianized	Influenza vaccine, live,avianized	Influenza vaccine, Split Virion,avianized	Influenza vaccine, Split Virion,MDCK	
1	Harvest	Harvest	Harvest	Harvest	Harvest
2	Clarify/Centrifuge	Clarify/Centrifuge	Inactivitied	Clarify/Centrifuge	Clarify/Centrifuge
3	100KD TFF Capture	100KD TFF Capture	Clarify/Centrifuge	300KD TFF Capture	300KD TFF Capture
4	Inactivitied	—	100KD TFF Capture	Nuclease Hydrolysis	Nuclease Hydrolysis
5/6	DGU / TFF — SEC	DGU / TFF — SEC	DGU / TFF — SEC	DGU / TFF — SEC	Smac DeVirS / Inactivitied
7	Sterile Filtration	Sterile Filtration	Virus lysis	Inactivitied	Clarify/Centrifuge
8	—	—	TFF/Purify	Clarify/Centrifuge	Core700
9	—	—	Sterile Filtration	Core700	Smac Q
10	—	—	—	Smac Q	Virus lysis
11	—	—	—	Virus lysis	TFF/Purify
12	—	—	—	TFF/Purify	Sterile Filtration
13	—	—	—	Sterile Filtration	—

Purification protocols for different types of influenza vaccines

Adenovirus/adenovirus vector:

Adenovirus is a class of unenveloped double-stranded DNA viruses with an icosahedral capsid composed of 240 hexamers, 12 pentamers, and 12 fibrils attached to each pentamer.

Adenoviruses can infect almost all tissues and cell lines and carry gene fragments up to 8kb, making them crucial in immunology, gene therapy, and cell therapy. It is one of the most commonly used vector viruses.

	Preparation	Purify	
Line A	Cell lysis & Clarify	Smac Q 40	CORE700
Line B		AAV Affinity Beads 4FF	CORE700

Two commonly used AAV purification methods

AAV Affinity Beads 4FF is an affinity chromatography medium designed for purifying adeno-associated viruses (AAV), demonstrating high affinity for subtypes 2, 5, and 8.

Matrix	Highly crosslinked 4% agarose
BioProcess resin	Yes
Capacity	＞1×10^{13}Copies/ml resin
Particle size, d50V	45-165 μm
Flow velocity	⩾300cm/h
Pressure	0.3 MPa/3 bar
Storage	2 - 8°C,in 1×PBS contain 20% ethanol