



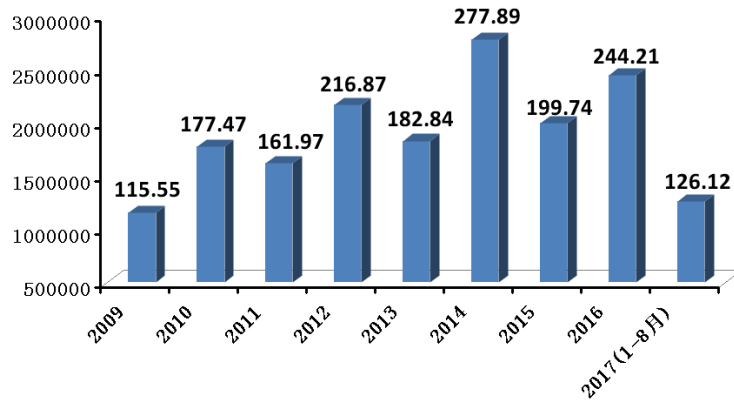
Inactivated Enterovirus 71 vaccine(Human diploid cell)

-Development, purification and clinical evaluation

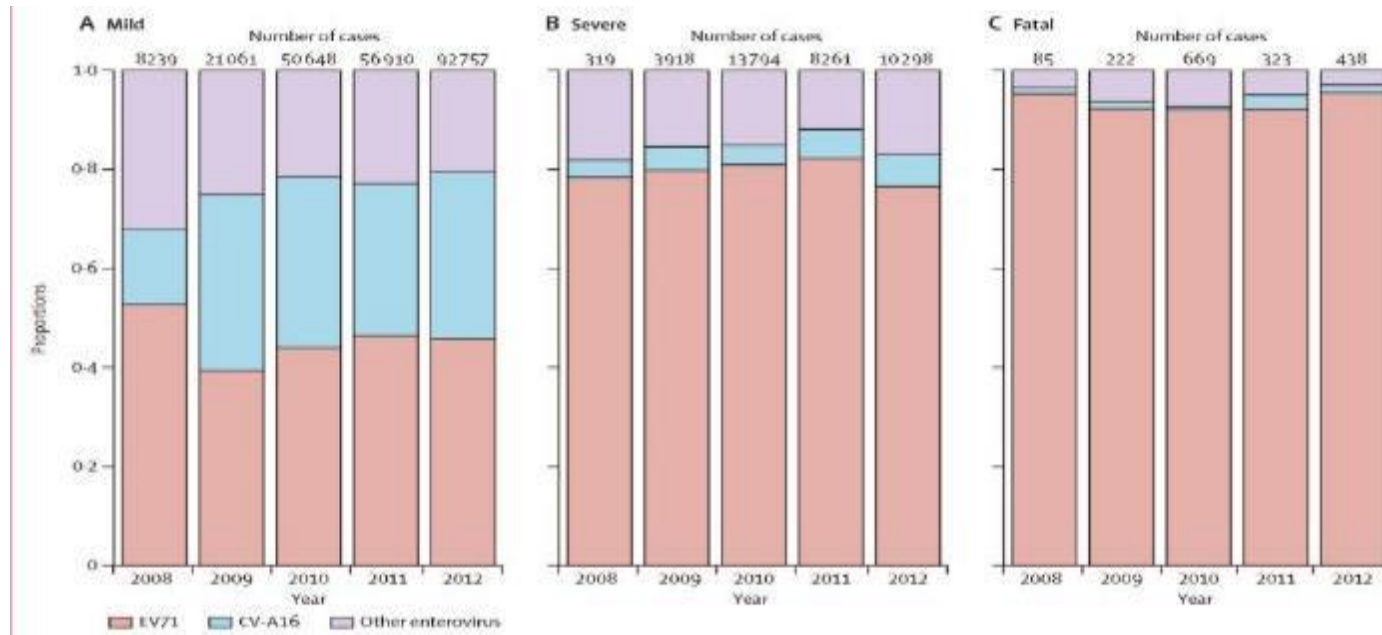
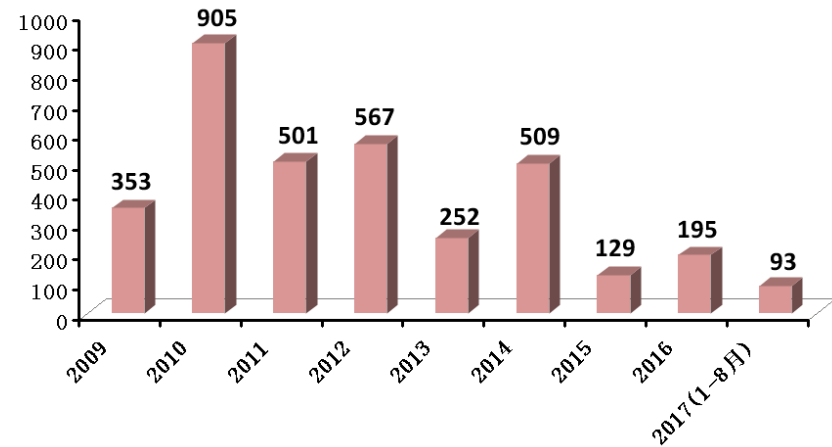
Longding Liu, PhD, Professor
Institute of medical Biology, CAMS, PUMC.

EV71 infection and its risk in China

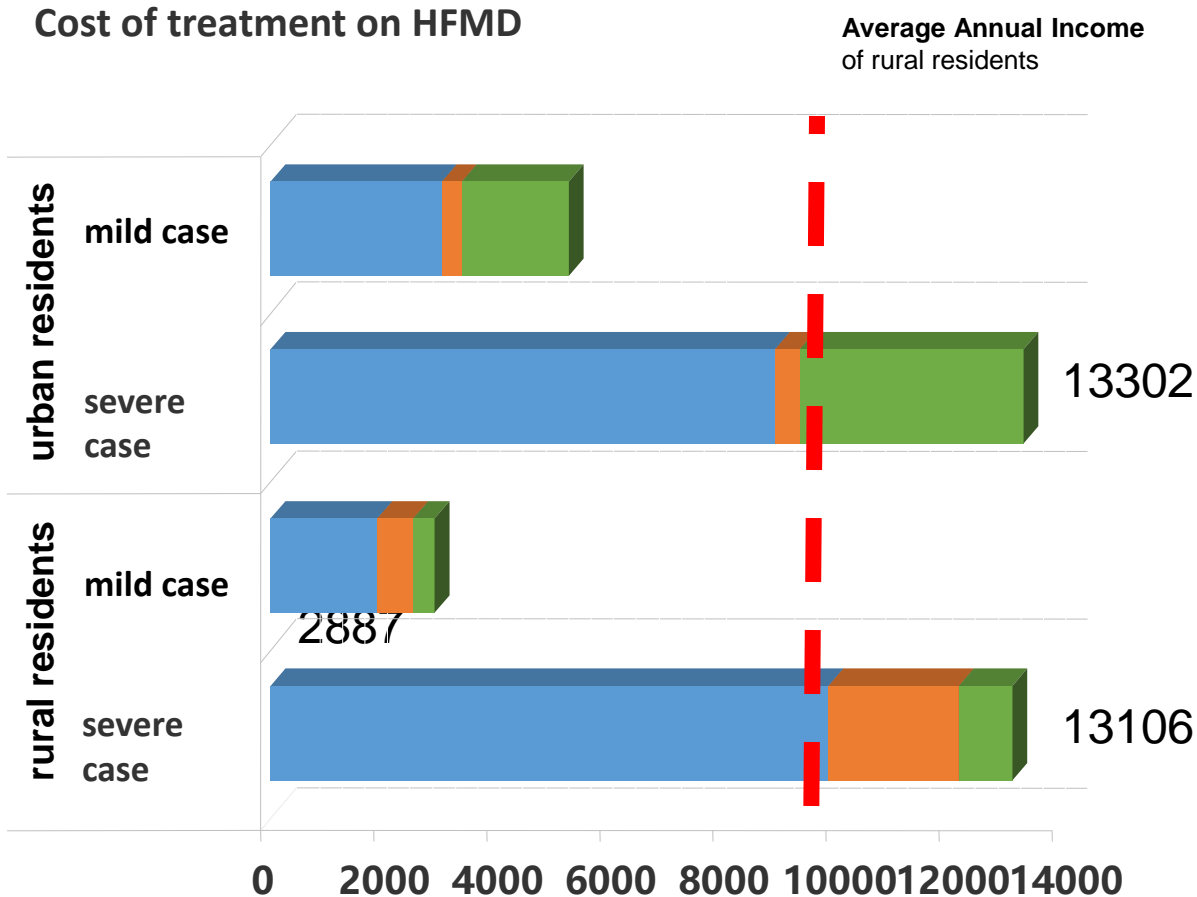
The number of cases of HFMD in China,



The deaths cases of HFMD in China,

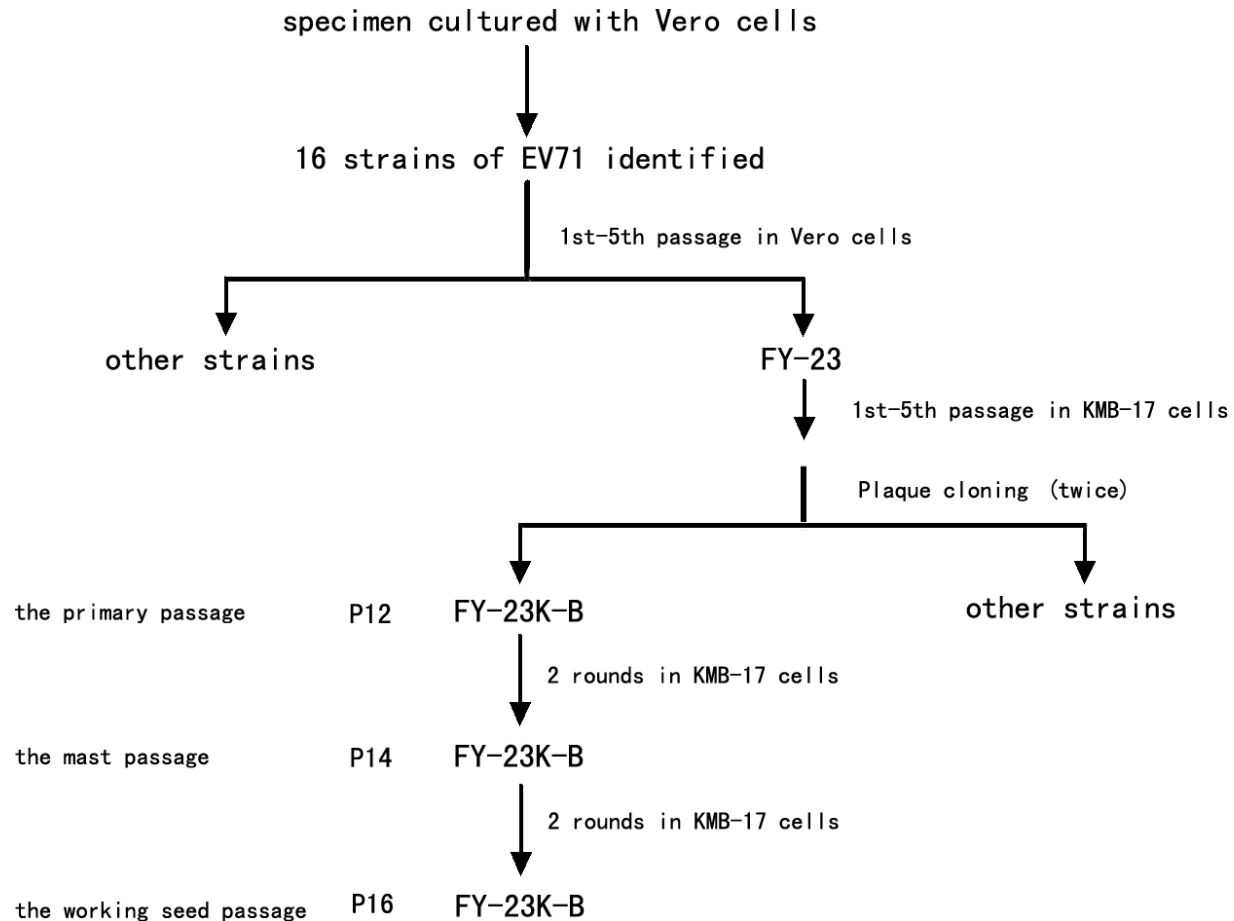


Economic analysis of HFMD



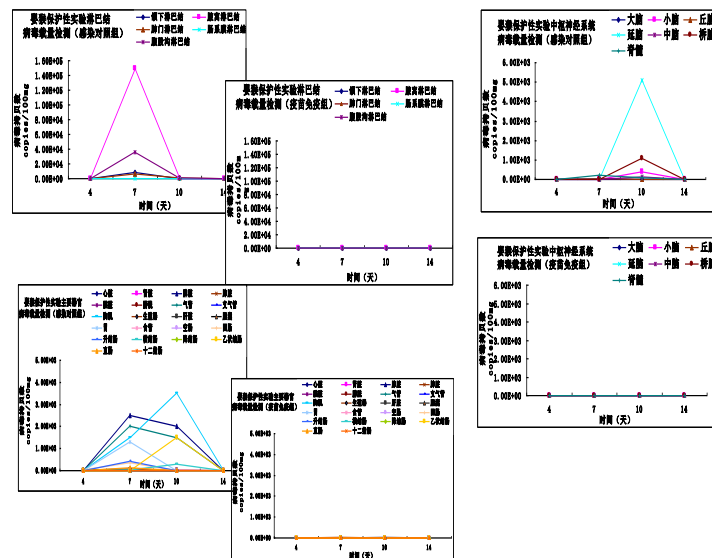
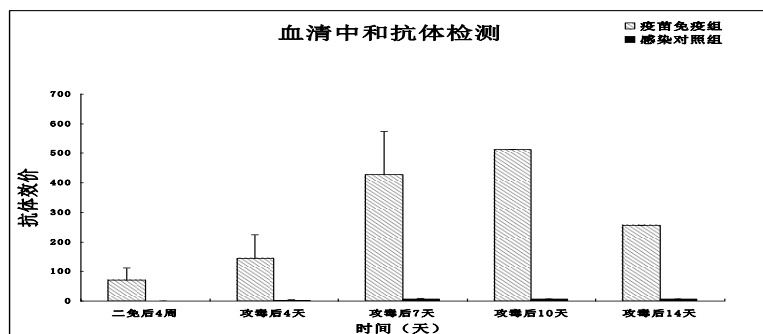
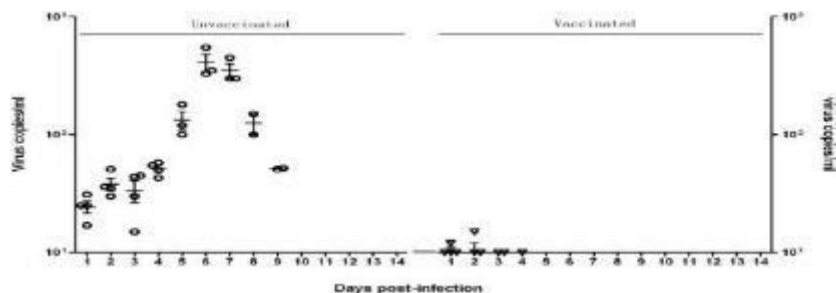
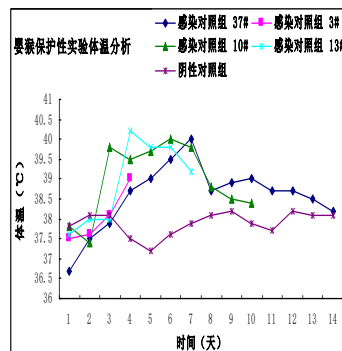
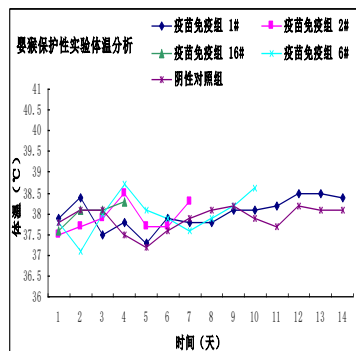
Good news: 20,000 doses EV71 vaccine had been used in areas with population of 12.43 million, incidences of HFMD and its severe case decreased 41% and 68% respectively .



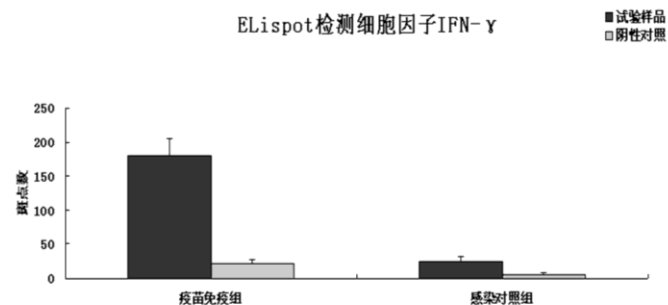


By adaptation in KMB-17 cell substance, the primary, mast and working seed-lot were set up at 12th, 14th, and 16th passage, respectively.

Protective effect of EV inactivated vaccine in monkey



Viral loads detection of various organs of immunized animal at viral challenge



Clinical trial of EV71 inactivated vaccine (human diploid cell)

Approved by China FDA
Registered number NCT01569581 in Clinicaltrials.gov,

Phase I: safety;

Phase II: safety; immunogenicity;

effective dose and immunization program

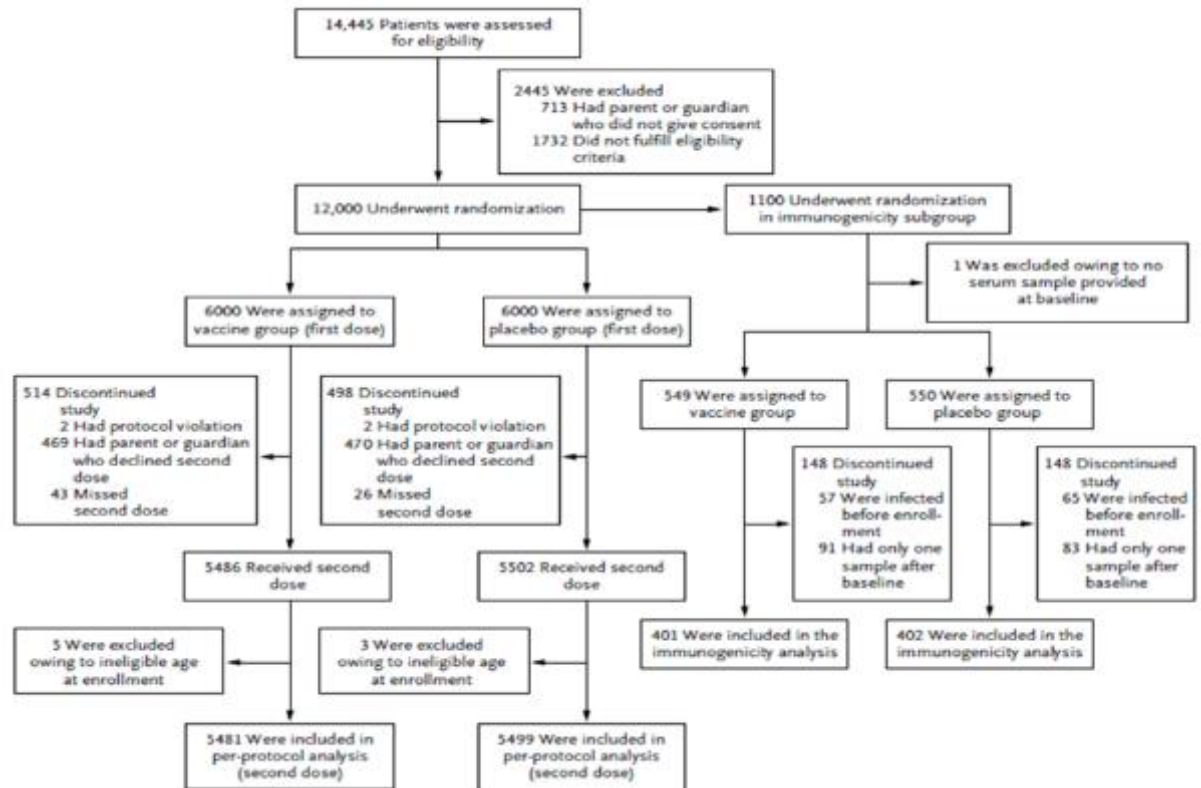
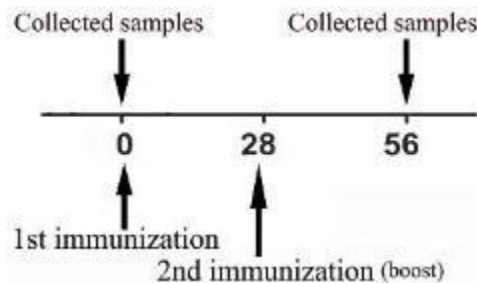
Phase III: safety; effect and clinical

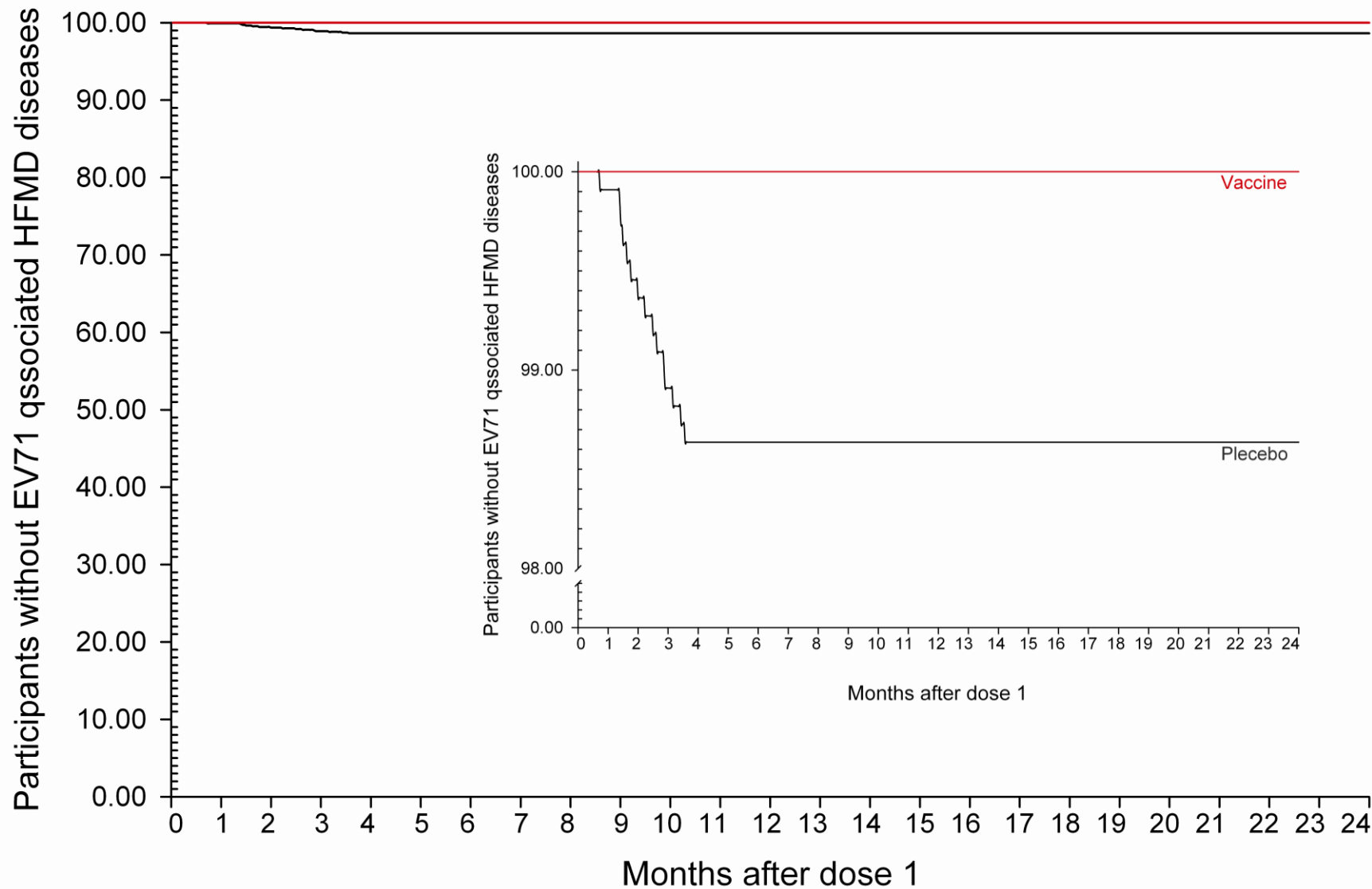
protective efficacy.

[illegible]

Phase III

- Subjects: 12000 Children (6 – 71 month old)
- Principle: random, double-blind and placebo control
- Dose: 100U (320EU)
- Program: 0, 28 days

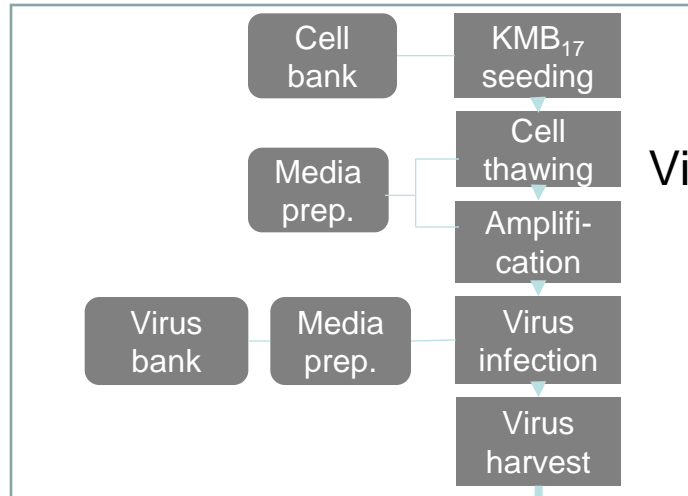




Production process

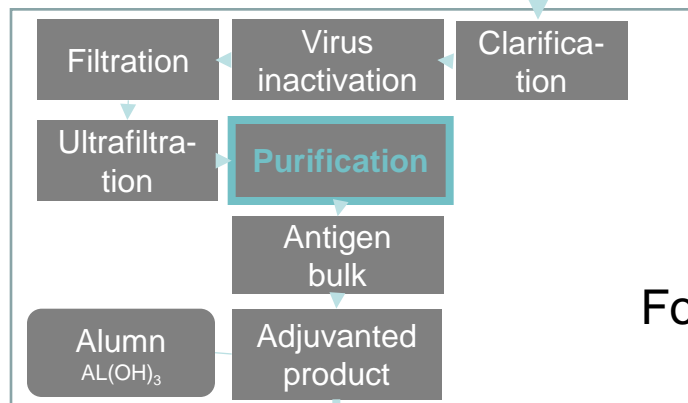
Production process

Virus cultivation and harvest

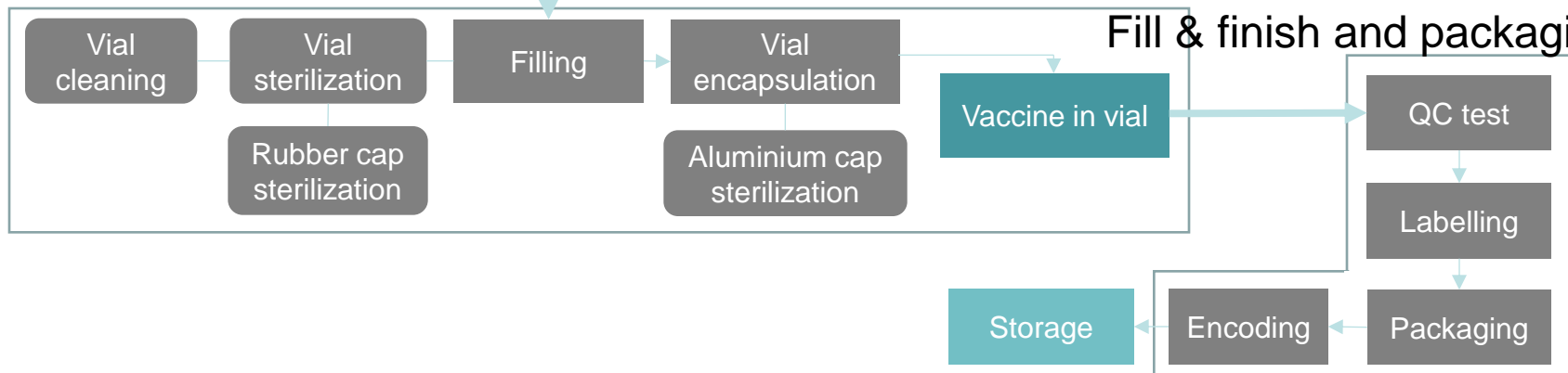


Purification chromatography ÄKTA™ pilot
Sephacrose™ 6 Fast Flow size exclusion
chromatography

Formulation, vaccine bulk

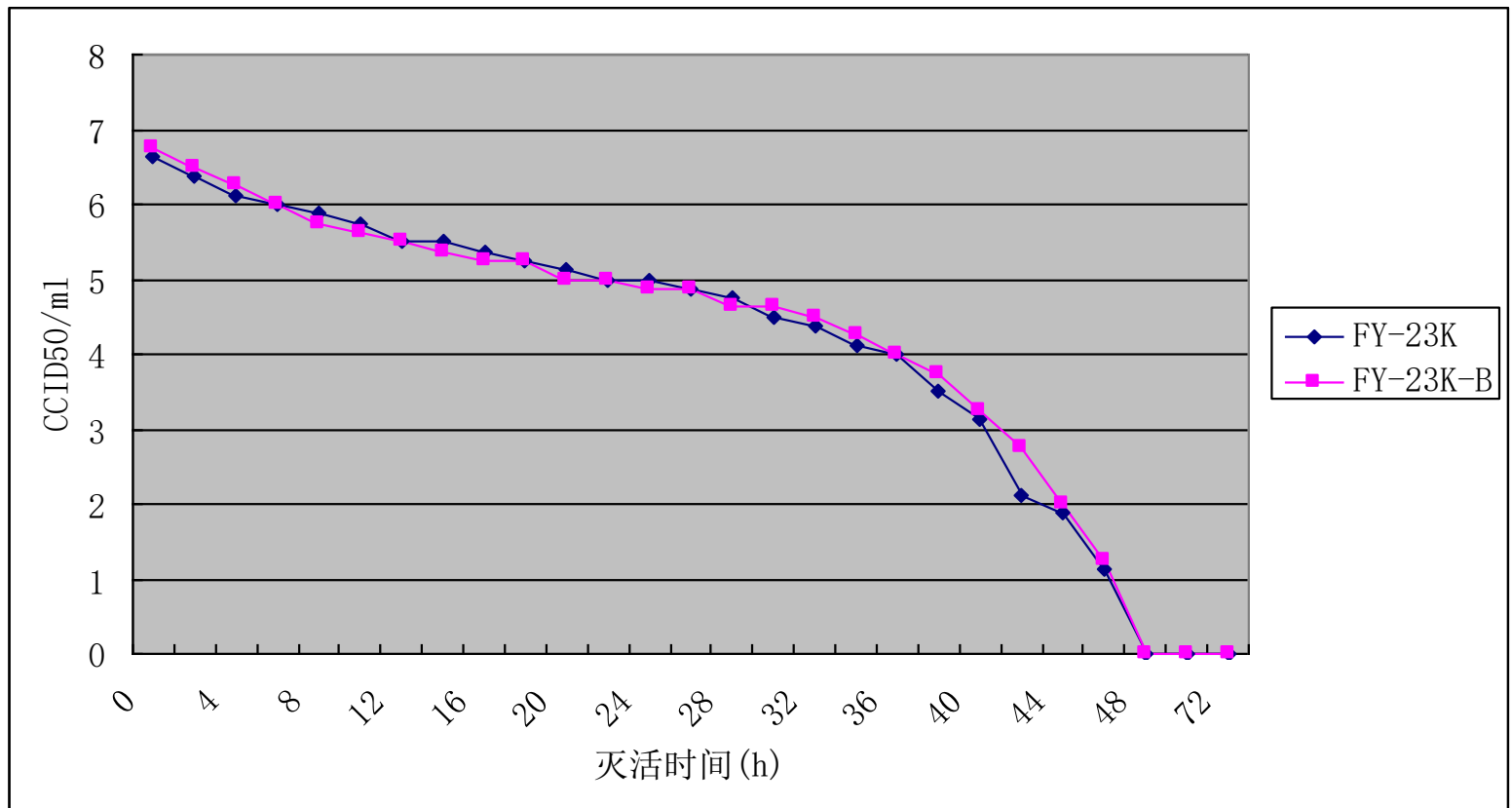


Fill & finish and packaging



Inactivation Validation

- evaluation of the efficiency of viral inactivation and vaccine antigenicity



VALIDATION OF THE PROCESS

- Purification of the bulk
 - exclusion of extraneous agents from the product
 - adequate removal of culture medium and reagent-derived contaminants from the product
 - maintenance within stated limits of the yield of product from the culture

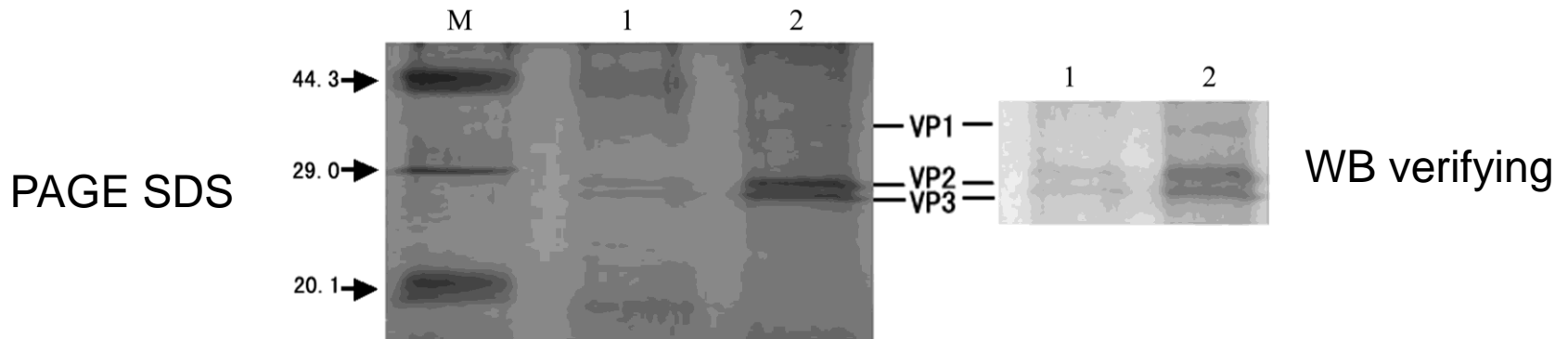
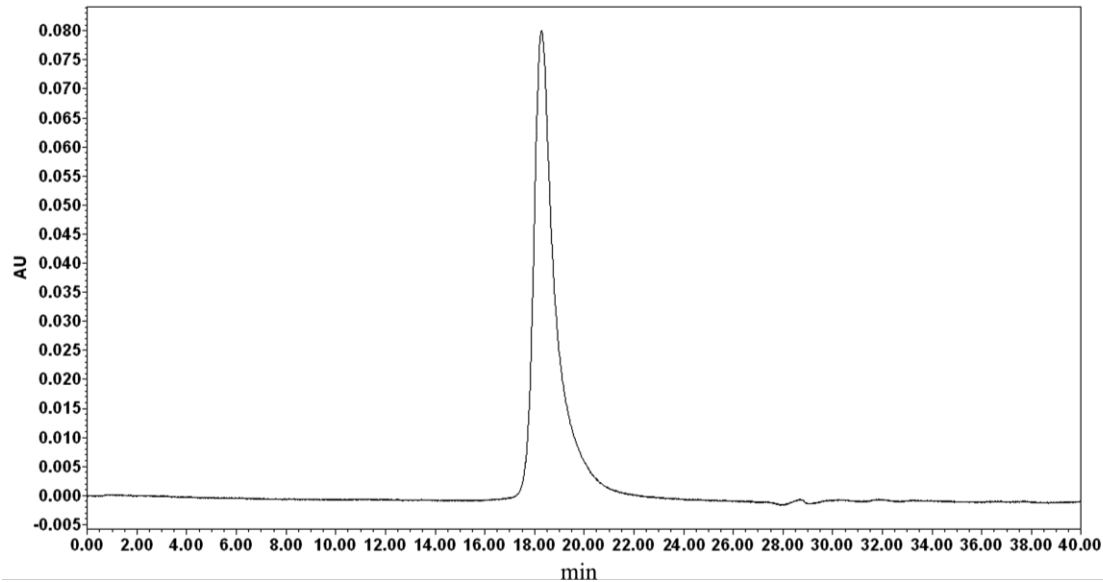


ÄKTA™ pilot



Identity Confirmation

- Purity detection by HPLC



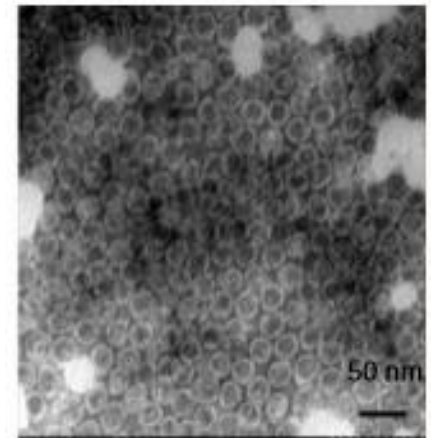


Purification of EV71 VLP

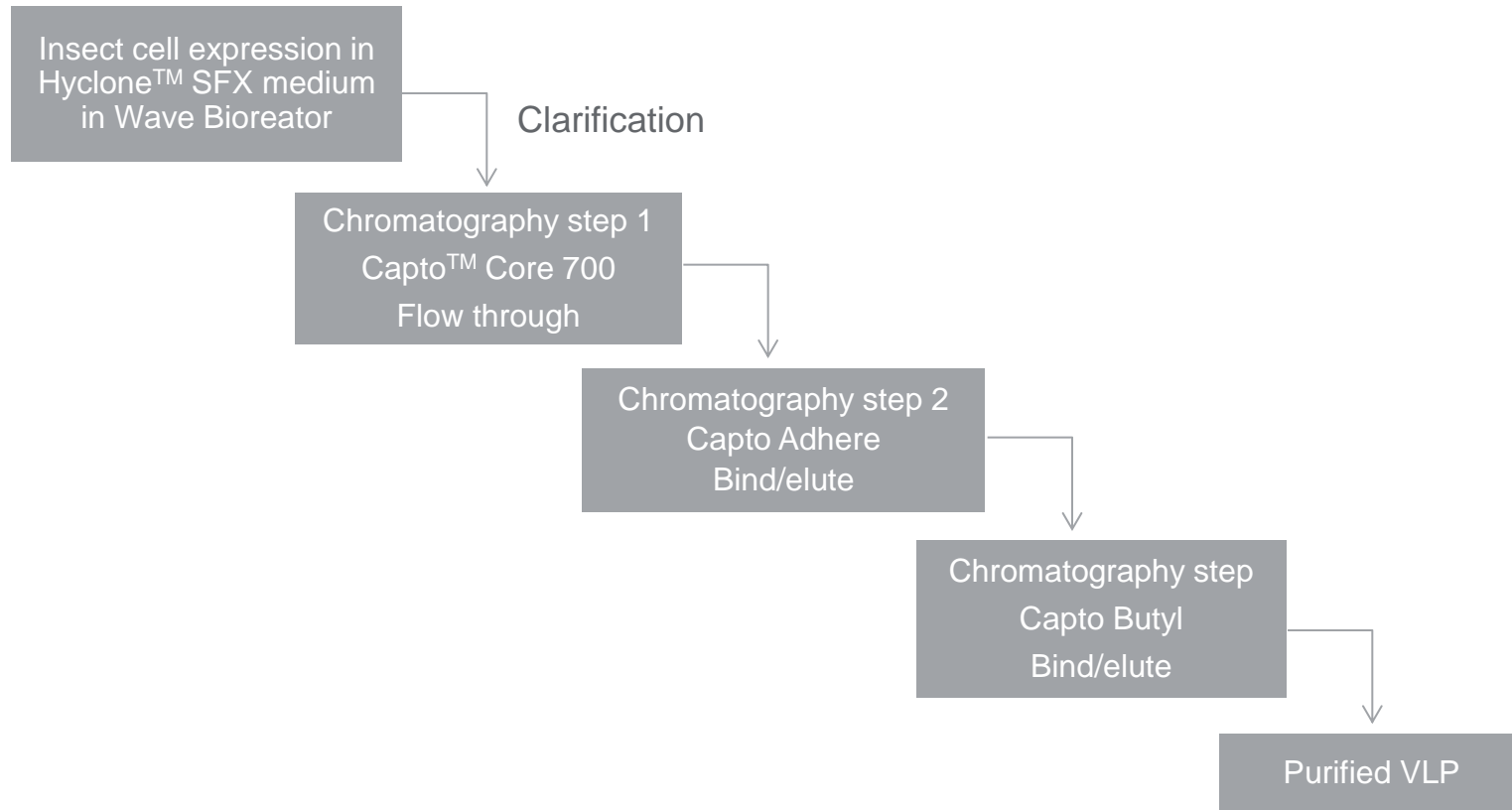
Dr. Mats Lundgren
October 10, 2017

Key facts of EV71 VLP

- VLP as an alternative to inactivated virus based vaccines
- ~30nm size VLP particle
- Baculovirus based expression in insect cells (Sf9) grown in WAVE bioreactor
- Serum-free bioreactor process using Hyclone (GE) SFX medium
- Chromatography based purification as an alternative to density ultracentrifugation
- 3 step chromatography process
- Over 95% purity



Purification process – EV71 VLP

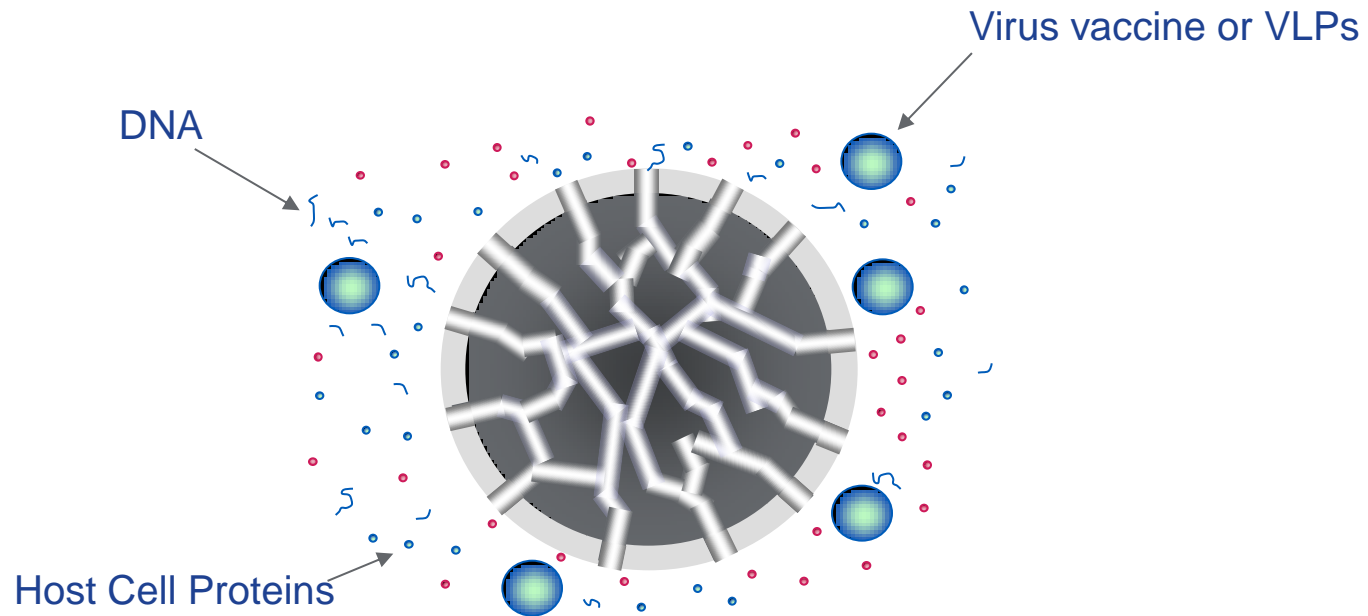


VLP = virus-like particle

ALEX = anion exchange chromatography

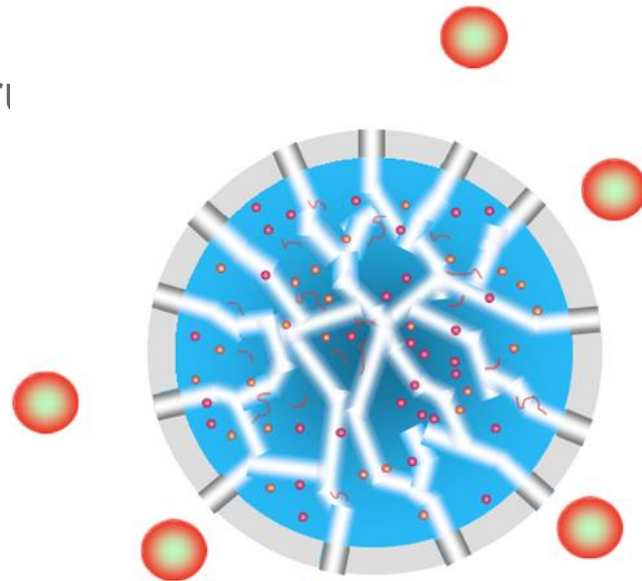


Core beads – the application principle



Application examples Core beads

- Influenza
 - Egg based
 - Cell based
- Dengue, Zika and other Flavivir
- Lentivirus
- Adenovirus
- CMV
- RSV
- Poxvirus vectors
- Polysaccharide conjugates
- VLPs etc.





***Thanks for your
attention!***