CLAIMS

What is claimed is:

1. A method for conjugating a protein and a virus, comprising: activating a virus by exposing the virus to an environment at a pH of 4.50 to 5.50; and mixing the virus and a protein after the activating step in a conjugation reaction to form a conjugate mixture.
2. The method of claim 1, wherein the virus activating step further comprises chemically modifying surface-exposed lysine residues on the virus to facilitate amine-targeted conjugation of the protein during the conjugation reaction, and wherein the virus activating step is conducted in a buffer for a period of time between 18 hours and 72 hours.
3. The method of claim 1 or claim 2, wherein the protein is an antigen, and further comprising preparing the antigen by sub-steps of purifying the antigen and exposing the antigen to a pH of 6.5 or less prior to the mixing step.
4. The method of claim 3, further comprising harvesting the antigen from a source antigen, wherein purifying the antigen comprises removing cellular debris from the antigen, concentrating the antigen, performing at least one chemical separation to separate host cell contaminants from the antigen, subjecting the antigen to affinity chromatography to elute the antigen, and subjecting the antigen to multi-modal chromatography to separate residual impurities from the antigen based on size differences between the impurities as retentate and the antigen as permeate.
5. The method of any one of claims 1-4, further comprising passing the virus through a tangential flow filtration apparatus comprising a porous filter to purify the virus and passing the virus through a membranous filtration device comprising a porous membrane to concentrate the virus.

6. The method of claim 1 or claim 3, wherein the protein is an antigen comprising an influenza hemagglutinin antigen (HA) and the virus is a tobacco mosaic virus (TMV).
7. The method of claim 6, wherein the TMV and HA are mixed at a ratio of at least 4:1.
8. The method of claim 6, wherein the TMV and HA are mixed at a ratio between 1:1 and 4:1.
9. The method of any of claims 1-3, further comprising quenching the conjugation reaction using at least one liquid containing free amines.
10. The method of claim 9, further comprising, following the quenching step, separating residual impurities from the conjugate mixture based on size differences between the impurities as retentate and the conjugate mixture as permeate.
11. The method of any of claims 1-3, wherein the conjugation reaction achieves at least 50% reduction in non-conjugated, native molecular weight, virus coat protein.
12. The method of any of claims 1, 7, or 8, wherein the virus is TMV and the TMV has a radius, and the step of mixing the virus and protein is performed after the TMV radius has increased by at least a factor of 2.75 during the activation step.
13. The method of any of claims 1, 7, or 8, wherein the virus is TMV and the TMV has a radius, and the step of mixing the virus and protein is performed after the TMV radius has increased to at least 195 nm during the activation step.