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Chicken immunization with KLH-gp120 fragment (254-274) conjugate.

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1 Works for me dx.doi.org/10.17504/protocols.io.bjnjkmcn

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ABSTRACT

Chicken immunization with peptides is inefective if only just the peptides are being inoculated. However, to make the immune response effective the fragment 254-274 of HIV-1 was conjugated with a carrier protein (KLH) that produced a critical immune response, assessed by ELISA, Immunoblot analysis and dot blot [1-4]. The Polson method (1990) can be used effectively to separate the IgY antibody from the egg yolk of immunized chickens [5].

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MATERIALS

NAME	CATALOG #	VENDOR
5 x 2ml No-Waste™ Freund's Incomplete Adjuvant (FIA)	786-099	G-Biosciences
2ml No-Waste™ Freund's Complete Adjuvant (FCA)	786-709	G-Biosciences
Fragment 254-274 of Gp120 of HIV (peptide)		

1 Two healthy layer chickens (brown Leghorn), aged approximately 6 months, are injected intramuscularly at multiple sites on the breast with the peptide-keyhole limpet hemocyanin **(**KLH) conjugate.

2 The chickens are immunized on day 0, with 0.2 µmol/ml of the fragment 254-274 of HIV gp120-conjugated to KLH (immunogen) in 0.5 ml complete Freund’s adjuvant (Sigma-Aldrich Co, St. Louis Missouri).

3 On days 15, 60, and 90 chickens are immunized with 0.2 µmol/ml of the immunogen in 0.5 ml incomplete Freund’s adjuvant (Sigma-Aldrich Co, St. Louis Missouri).

4 The eggs are collected post-immunization.The immunoglobulin Y is separated using the Polson method (1990).

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