AUG 17, 2020

An easy chromatographic method for purification of Immunoglobulin Y (IgY) using HiTrap™ Columns.

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OPEN ACCESS**DOI:**dx.doi.org/10.17504/protocols.io.bju7knzn

Protocol Citation: Angel A Justiz-Vaillant 2020. An easy chromatographic method for purification of Immunoglobulin Y (IgY) using HiTrap™ Columns.. **protocols.io** <https://dx.doi.org/10.17504/protocols.io.bju7knzn>

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Protocol status: Working
We use this protocol and it's working

Created: Aug 17, 2020**Last Modified:** Aug 17, 2020**PROTOCOL integer ID:**
40575

- 1 Fill the syringe or pump tubing with de-ionized water. Remove the stopper and connect HiTrap™ column to syringe (use the connector supplied).

- 2 Snap off tab on the column outlet.
- 3 Wash out the ethanol with 26 ml of de-ionized water.
- 4 Equilibrate column with 26 ml of binding buffer. The recommended flow rate is 5ml/min.
- 5 Apply the IgY sample using a syringe fitted to Luer connector or by pumping it onto the column.
- 6 For better results, use a flow rate of 0.5 to 5.1 ml/min during sample application.
- 7 Wash with at least 51 ml of binding buffer or no material remains in the effluent.
- 8 Maintain a flow rate of 5 to 11 ml/min for washing.
- 9 Elute with 51 ml of elution buffer using a one-step or using a linear gradient though larger volumes are often required to break the interaction.

- 10 After elution, regenerate the column by washing with 36 ml of wash buffer.
- 11 Re-equilibrate the column with 26 ml of binding buffer.
- 12 The column is now prepared for a new purification.