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# Polson method for IgY isolation with modification.

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## ABSTRACT

Immunoglobulin Y (IgY) is the major molecule present in the avian egg yolk. This protein functions as antibody and fulfils important functions in the protection of the embryo against challenging stimuli [1]. IgY has been isolated from the egg yolk of many birds [1]. These antibodies are important therapeutically, but also they have been used as immunological tool in many immunodiagnostic assays.

Much time IgY concentration is calculated from the water soluble fraction (WSF), which means that there is not need of treating the preparation with alcohol to separate the protein pellet.

## Reference

1. Justiz Vaillant AA, Ramirez N, Cadiz A, Ferrer B, Akpaka P, et al. (2013) Separation and Reactivity of Avian Immunoglobulin Y. J Chromat Separation Techniq 4: 173. doi:10.4172/2157-7064.1000173

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## GUIDELINES

The modification in the Polson method (1990) is based on the substitution of the protein separation step with PEG by another suitable reagent as ethanol or a substance as trichloroacetic acid [1].

## Reference

1. Asemota H, Curtello S, Vaillant AAJ, Mohammed W, Vuma S, et al. (2013) Purification of Avian IgY with Trichloroacetic Acid (TCA). J Chromatograph Separat Techniq 4: 205. doi:10.4172/2157-7064.1000205

## MATERIALS

NAME	CATALOG #	VENDOR
100 ml Polyethyleneglycol 600	orb65596	biorbyt
Chloroform	366919-1L	Sigma
Trichloroacetic Acid	490-10	Sigma Aldrich
Ethanol	100983	Merck Millipore
PBS		Life Technologies

- 1 Wash the egg with warm water and crack the eggshell carefully.
- 2 Separate the egg yolk from the egg white manually and place the egg content on a tissue paper to help to remove as much egg white as possible.
- 3 Wash four times the egg yolk with 100 ml of PBS, pH 7.4.
- 4 Break the egg yolk membrane and pour it into a 50 ml tube. Then, dilute egg yolk 1:5 in PBS, pH 7.4, and add an equal volume of chloroform (ACS reagent grade, Sigma-Aldrich).
- 5 Shake the mixture, and vortex and roll the tube containing the mixture on rolling mixer for 15 minutes.
- 6 Decant the supernatant, which contains the water-soluble fraction (WSF) that is the diluted egg yolk plasma, rich in IgY (25 mg/ml), which is approximately four times the IgY concentration present in the serum (6 mg/ml).
- 7 Precipitate egg yolk antibodies (IgY) by dropping the WSF with cold ethanol, trichloroacetic acid (TCA) or the classic polyethylene glycol. (PEG).
- 8 Estimate the protein concentration.
- 9 Run a protein gel electrophoresis (PAGE) of the IgY to assess the molecular weight of the chicken egg yolk antibody, protocol for this can be found elsewhere.
- 10 Then use Phosphate-buffered saline (PBS) pH=7.4 to dialyze the preparations for 8 hrs at 4°C.
- 11 The dialysate is stored at -20°C until further use.

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