

Volume: 25 ml

Lot # _____

Sheep Anti-Rat Fx1A Serum (PTX-002S)

For the Induction of Passive Heymann Nephritis (PHN)

Materials supplied:

Anti-Fx1A Serum, 25 ml, in 0.02 M phosphate buffered saline PBS, pH 7.3. Store at -20 C or below. Avoid repeated freeze-thaw. *Centrifuge 3,000 rpm prior to use.* This product has not been validated in mice.

Directions for use:

This package contains sufficient antibody to induce passive Heymann nephritis in 25 rats* (175-200 gm) when used according to the instructions below. Please read carefully before starting the procedure.

Injection of anti-Fx1A Serum: Anesthetize rats and inject 0.4 ml/ 100 gm body weight of anti-Fx1A serum into a tail vein over a 15-30 second period. Production of disease is dose dependent, it is important that the complete dose of antibody is delivered. The recommended doses are tested to produce disease as outlined in this data sheet. Due to variability in antisera lots, source of rats, and investigator preferences, we recommend that dose-response studies be performed to establish the desired severity of disease from each lot of antisera.

Description of Renal Disease¹

Heterologous disease Faint-moderate immunofluorescence deposits of heterologous sheep IgG are noticeable in glomerular capillaries within minutes increasing in intensity and granularity by 3-5 days after injection of a single bolus of anti-Fx1A. Rat C₃ also localizes in a granular pattern and proteinuria develops after 5 days.

Autologous disease becomes noticeable after 7-10 days following administration of anti-Fx1A antibody evidenced by increased proteinuria and glomerular localization of rat (autologous) IgG in a "string of pearls" pattern by immunohistochemistry (Figure 1A) and subepithelial immune deposits by electron microscopy (Figure 1B). By 3 wks

proteinuria can be expected to reach 100-200 mg/24 hours (Bradford assay). Within the first few weeks glomeruli are unremarkable by routine light microscopy, progressing to thickened basement membranes, increased mesangial matrix, and possibly sclerosis after about 3 months^{1#}.

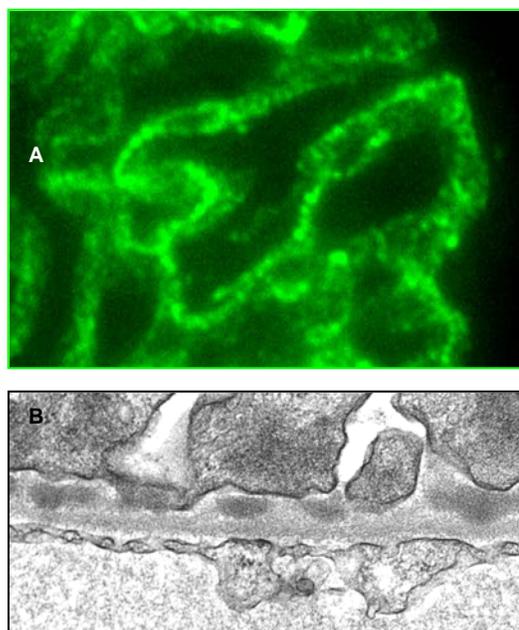


Figure 1A. Immunofluorescence localization of immune deposits (sheep IgG) in a "string of pearls" pattern in the glomerular capillary wall 2-weeks after initiation of Heymann nephritis. (B): Electron dense deposits localize in the subepithelial space.

1. Salant DJ, Cybulsky AV: Experimental glomerulonephritis. *Meth Enzymol* 162:421-461, 1988.

* Male Sprague-Dawley strain recommended. Other strains not tested.

Chronic disease not verified using Probetex anti-Fx1A antibody