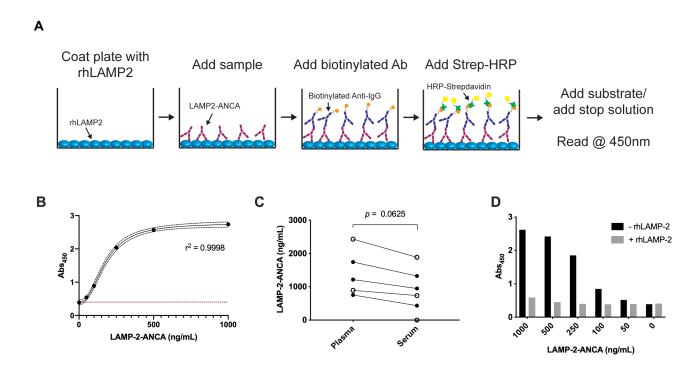


Supplementary Material

1 Supplementary Figures



Supplementary Figure S1. Custom indirect ELISA for detection of LAMP-2-ANCA in biosamples (serum or plasma). (A) Schematic and (B) representative standard curve with serial dilutions of mouse anti-human LAMP-2 mAb (x-axis: 0, 50, 100, 250, 500, 1000 ng/mL) plotted against Absorbance at 450 nm (Abs₄₅₀; y-axis). Data points were fit to a sigmoidal, 4-parametric curve ($r^2 = 0.9998$). Dotted red line denotes lower-limit of detection (Abs₄₅₀ = 0.402). (C) Concentration of LAMP-2-ANCA (y-axis; ng/mL) in paired plasma and serum (x-axis) from pediatric patients with vasculitis (n = 3; closed circles) and inflammation due to an autoinflammatory disease (n = 2; open circles). Paired plasma and sera from pediatric patients show slightly elevated LAMP-2-ANCA concentration in plasma (n = 5 p = 0.0625) (D) To validate the assay, standards (50 - 1000 ng/mL anti-human LAMP-2 mAb; x-axis) were added to the plate with (grey bars) and without (black bars) 25 µg rhLAMP-2. Absorbance (Abs₄₅₀; y-axis) of paired standards was measured. The rhLAMP-2 protein effectively blocks >97% of binding of 500 ng/mL mAb or less.