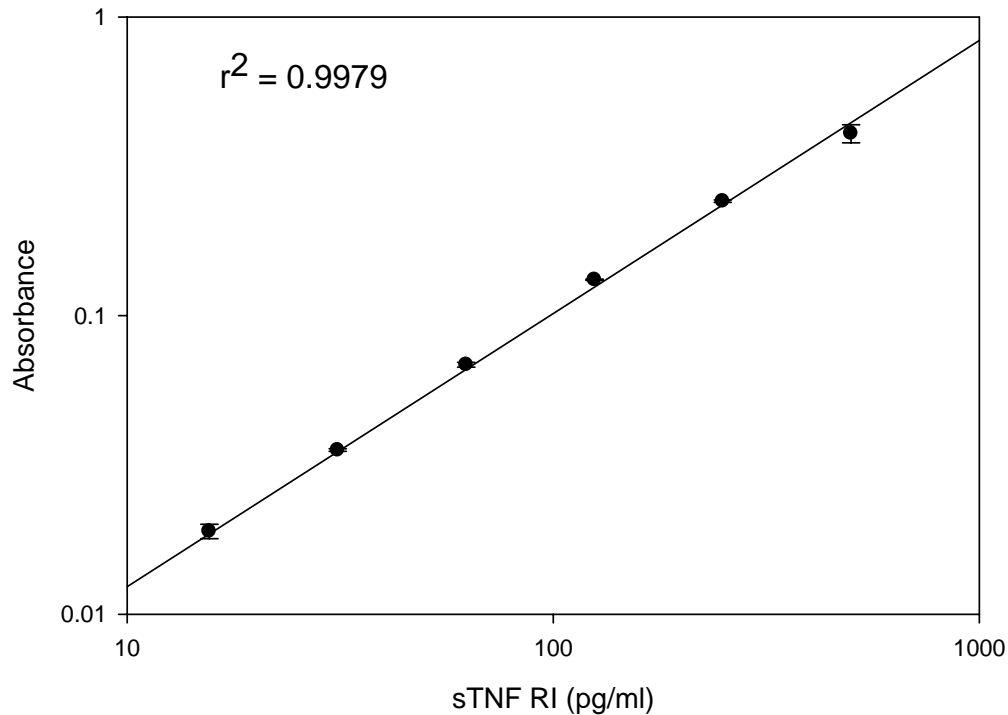


## Human sTNF RI Standard Curve



Plasma required: 50 microliters

Range of values: 0.5 - 1.9 nanograms/ml

Human Tumor Necrosis Factor Receptor I (or Type B) was measured utilizing a human sTNF RI Immunoassay kit purchased from R & D Systems, Minneapolis, MN (Catalogue #DRT100). The assay employs a quantitative sandwich enzyme immunoassay technique utilizing a monoclonal antibody specific for sTNF RI precoated into wells of a 96 well microplate. Standards and samples are added to wells resulting in the binding of sTNF RI to the immobilized antibody. After washing to remove any unbound substances. A mouse monoclonal antibody against sTNF RI which is conjugated to horseradish peroxidase is added. After removal of unbound conjugate a substrate solution is added to each well and color allowed to develop proportionate to the amount of sTNF RI present. The color development was stopped with the addition of 2 N  $H_2SO_4$  and the microplate was then read at 485 nm and also at 590 nm as a background control. Log-Log plots of concentration vs absorbance for standards were prepared and concentrations of unknown samples extrapolated from the standard curve and adjusted for any dilution of plasma.