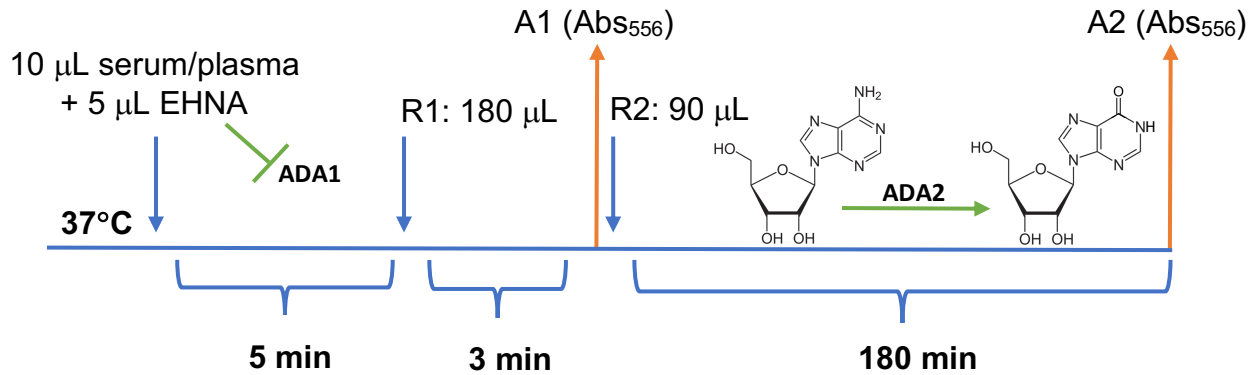
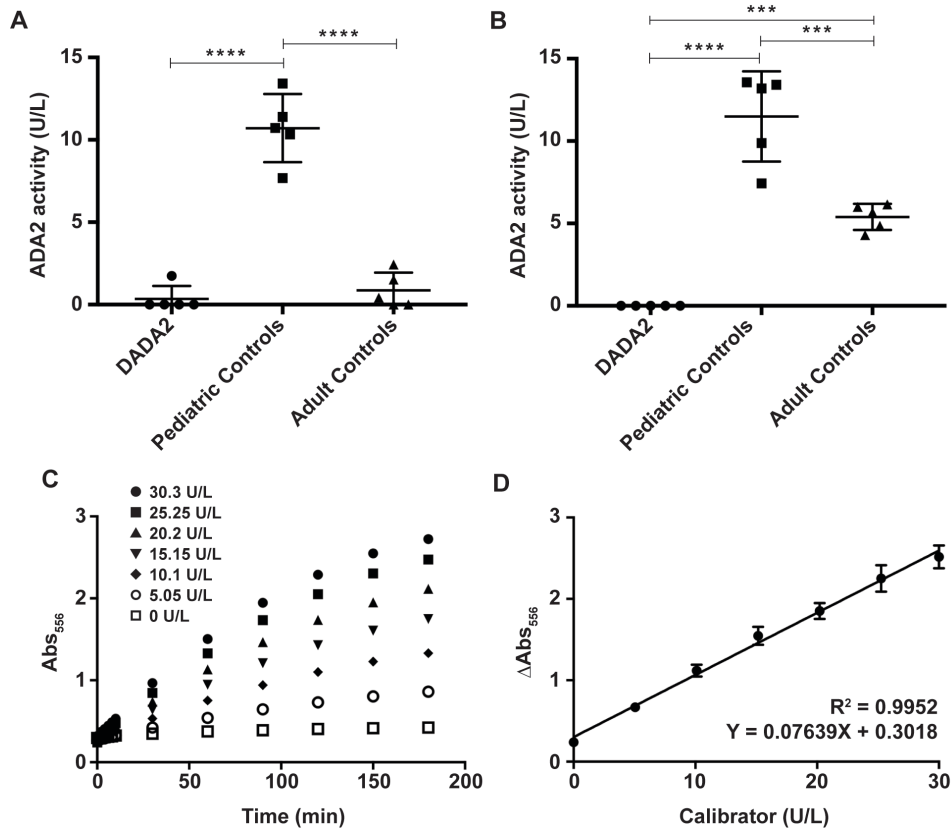


Supplementary Figure 1. Schematic overview of the ADA2 activity assay

One unit of adenosine deaminase (ADA) activity (U/L) was defined as the amount of ADA (inclusive of ADA1 and ADA2) that generates one μmole of inosine from adenosine per minute at 37°C . Specific ADA2 activity was taken to be the ADA activity of serum/plasma measured in the presence of $5\ \mu\text{L}$ of $1000\ \mu\text{M}$ EHNA, an ADA1 specific inhibitor (final concentration of $17.54\ \mu\text{M}$ EHNA in the total assay volume of $285\ \mu\text{L}$).



Supplementary Figure 2. Time optimization of ADA2 Assay (A) ADA2 activity (y-axis: ADA2 activity (U/L)) in blood sera or plasma obtained from DADA2 patients (x-axis: DADA2, n = 5), healthy children (x-axis: Pediatric Controls, n = 5), and healthy adults (x-axis: Adult Controls, n = 5) calculated after 10 minutes and (B) 3 hours. Horizontal bars show mean ADA2 activity +/- standard deviation (SD). (C) Absorbance (y-axis: Abs_{556nm}) measured in serial dilutions of calibrator over 3 hours (x-axis: Time (min)) (D) Change in absorbance (y-axis; Δ Abs₅₅₆) of serial dilutions of calibrator (x-axis: Calibrator (U/L), n = 3) after 3 hours. Points show mean change in absorbance +/- SD and trendline shows linear regression of data. * p < 0.05, ** p < 0.01, *** p < 0.001, **** p < 0.0001.



Supplementary Figure 3. Plasma CRP concentration in the healthy pediatric cohort. C-reactive protein (y-axis: CRP $\mu\text{g/ml}$) quantitated by ELISA in plasma (diluted 1:2000) from otherwise healthy children ages 5 months – 18 yrs (x-axis, n = 94). Dotted line denotes upper limit of normal CRP concentration in healthy children (5 $\mu\text{g/mL}$).

