

Supplementary Materials:

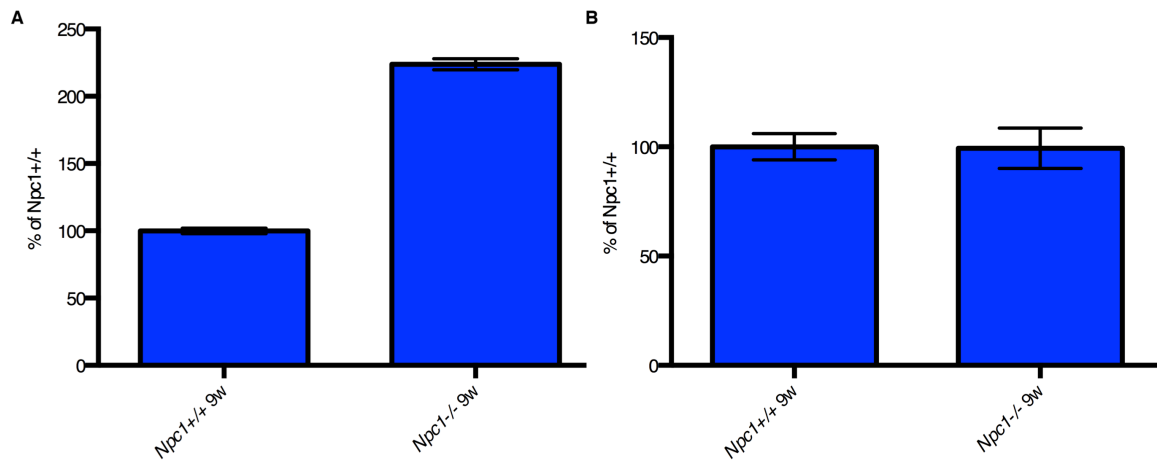


Figure S1: Comparison of Lysotracker (a) and LipidTox (b) staining on splenic B cells from 9 week-old *Npc1*^{+/+} and *Npc1*^{-/-} mice (n=3-5 per group). Elevation in Lysotracker staining was seen in 9-week-old *Npc1*^{-/-} mouse B cells but there was only low level staining with LipidTox and no difference between the two genotypes. These data indicated that Lysotracker is detecting storage in *Npc1*^{-/-} mice and that phospholipidosis is not contributing significantly to the cellular pathology of NPC disease B cells.

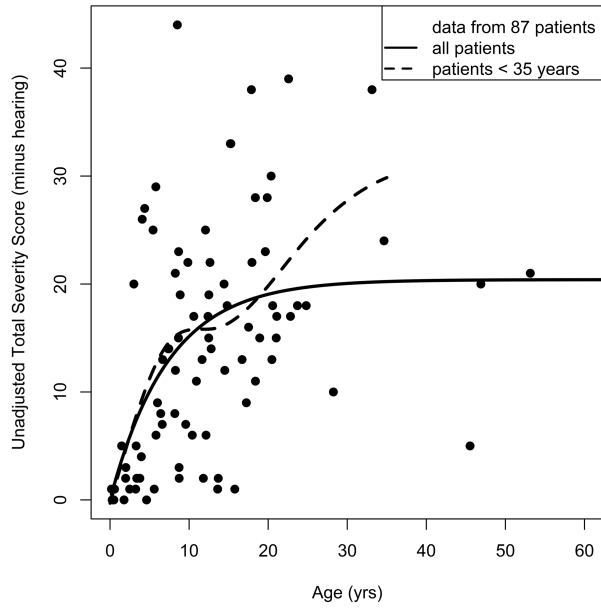


Figure S2: Plot of total severity score (minus hearing) against patient age either including (solid line: nonlinear asymptotic double exponential function) or excluding (dotted line: regression spline with four degrees of freedom) patients over the age of 30. The poor relationship of the fitted model to the data set prompted us to investigate whether latent sub-populations existed within the data set (see Fig. 5).

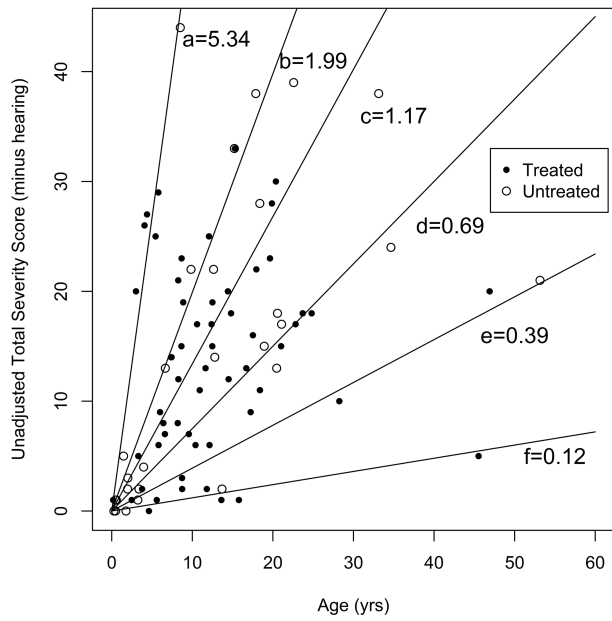


Figure S3: Re-plot of Fig. 2b in the main manuscript indicating miglustat treated patients (filled black circles, termed treated in the key) and untreated patients (open larger circles) to illustrate that all sub-populations (a-f) contained both miglustat treated and untreated patients.

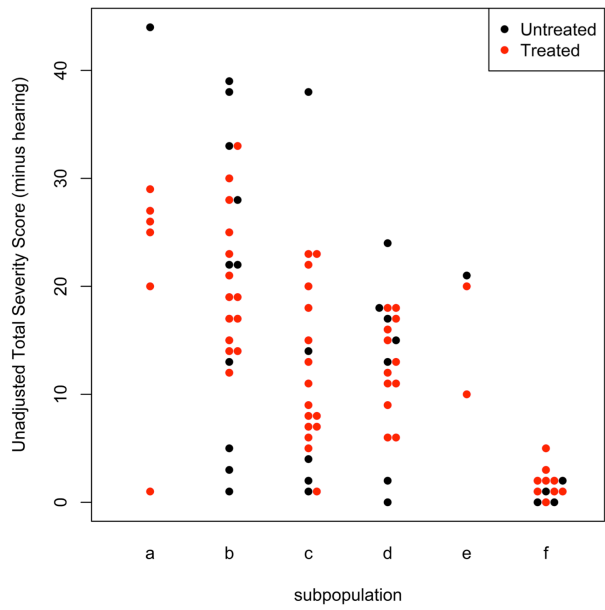


Figure S4: Alternative version of Supp. Fig. 3 presented as a scatter plot (red are miglustat treated and black are untreated NPC1 patients).

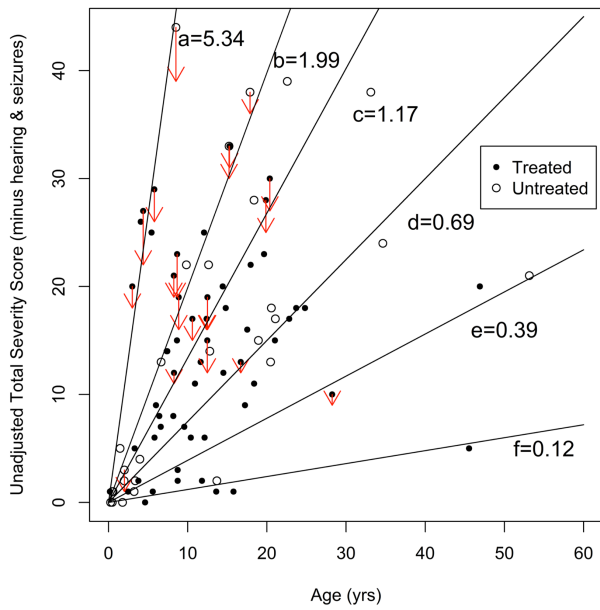


Figure S5: Graph of severity score against age (see Fig. 2b) with patients with seizures indicated in red with the end of the arrow representing severity score minus seizures (hearing is excluded in all patients as not measured in all centers). Seizures were predominantly present in the high severity groups (a-c).

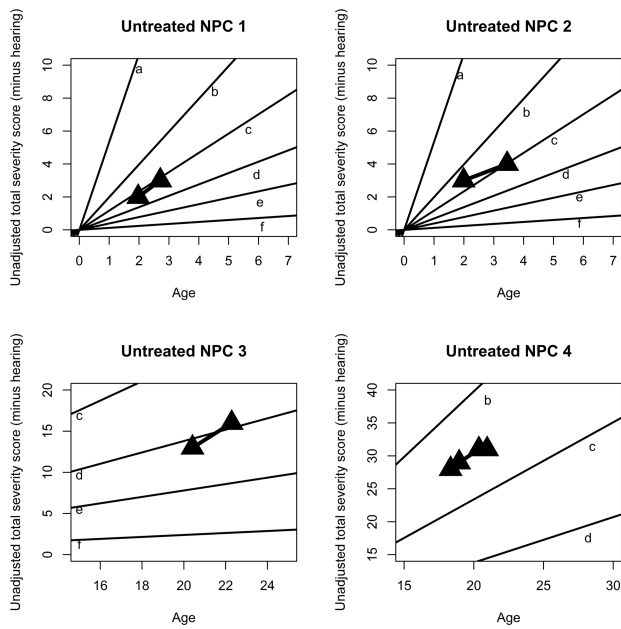


Figure S6: Zoomed in areas of plots from Fig. 2b showing that untreated patients 1-4 track individual severity lines over time, suggesting the plot in Fig. 2b may have potential prognostic utility. This now merits additional historic data analysis to test the validity of this in a larger study cohort.

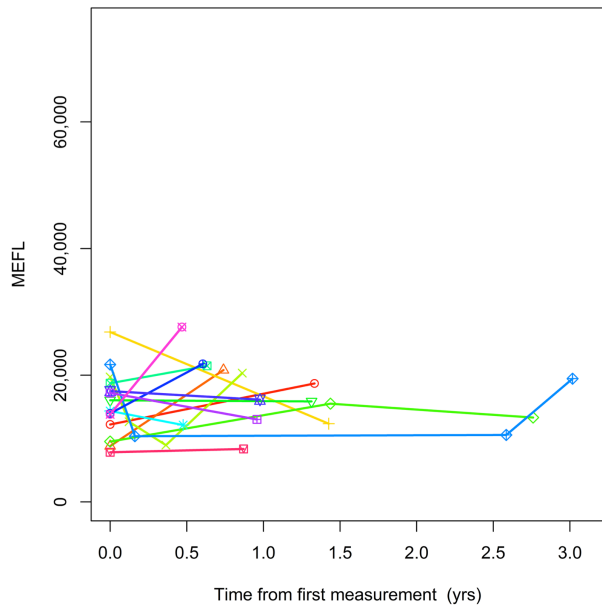


Figure S7: Repeat measures of MEFL in adult controls (heterozygous carriers and healthy volunteers) showing a broad range of individual MEFL values (in agreement with Fig. 2a) but minimal change (no slope) over time for each person when analyzed multiple times over variable periods of time (see also Table 1).

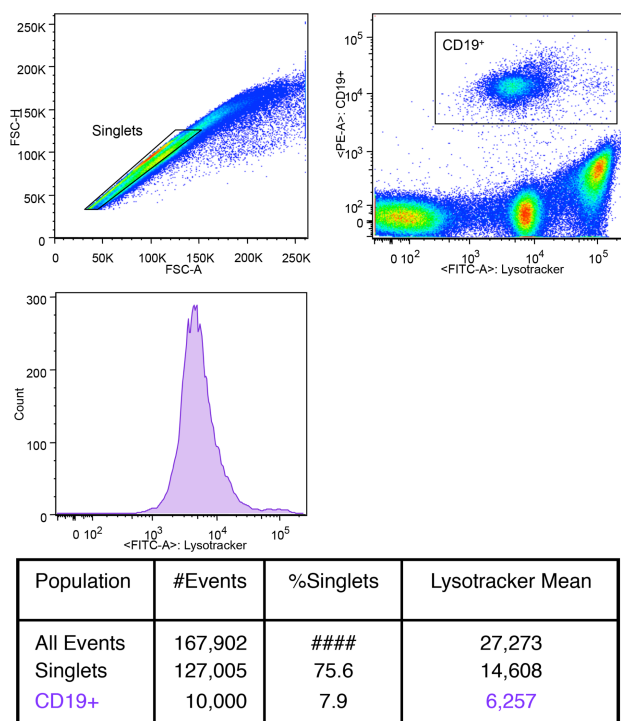


Figure S8: Representative gating and analysis of flow cytometry plots of a human blood sample. Singlet cells were defined and cells double positive for CD19 and Lysotracker[®]-green selected from this population. The 'Lysotracker Mean' values are standardized into mean equivalents of fluorescence (MEFL) values using Rainbow Calibration Particles (Becton Dickinson, UK).