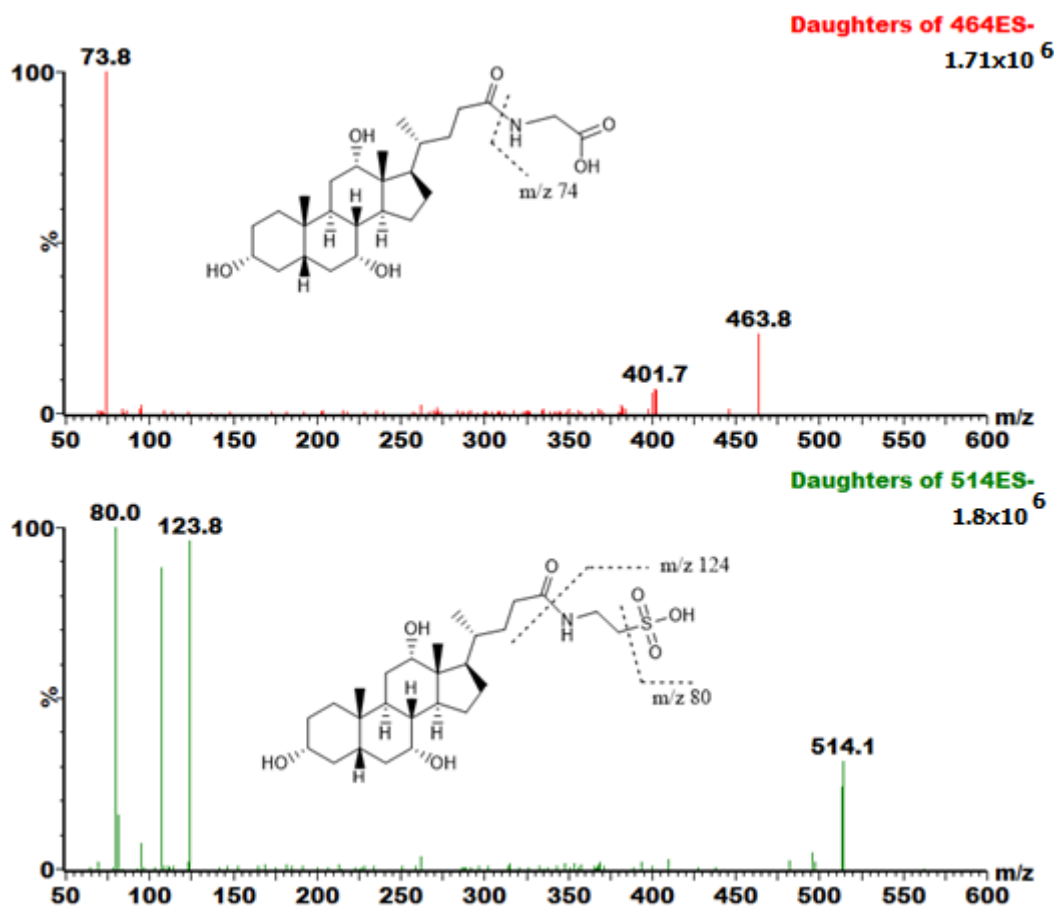


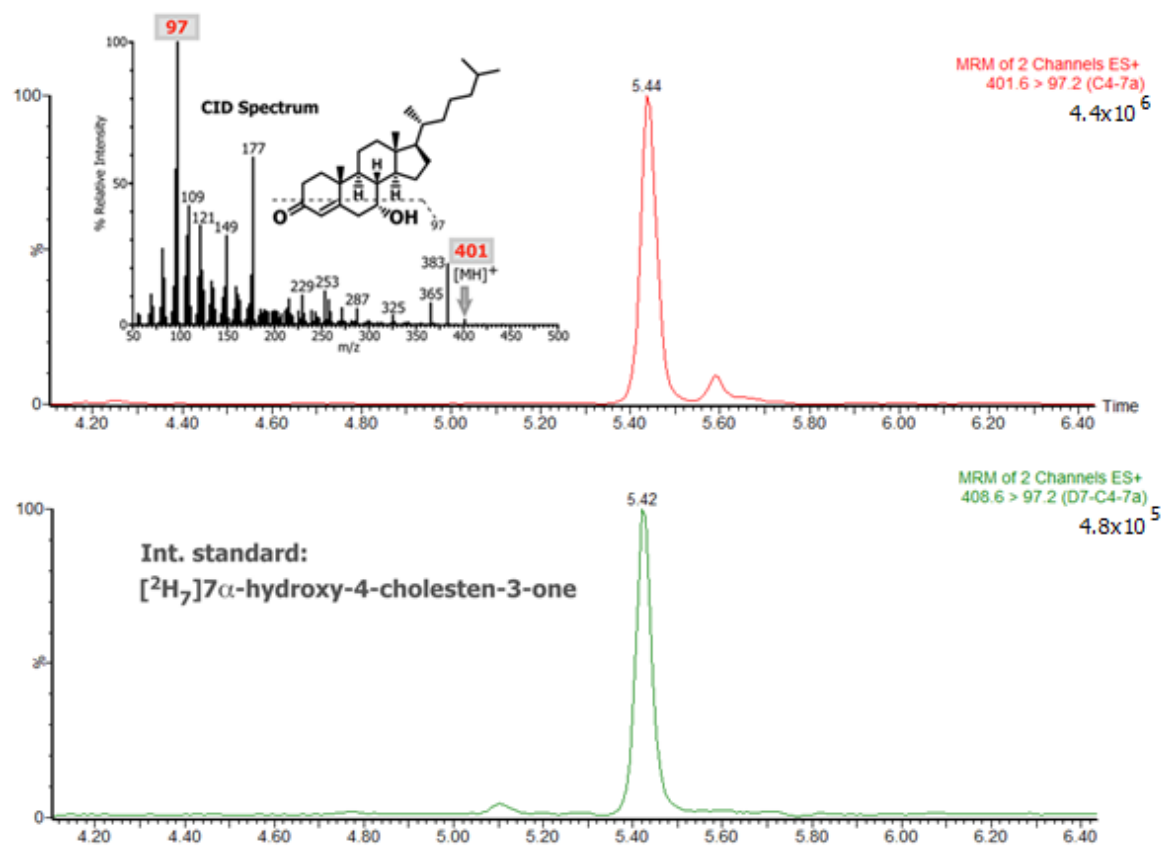
(a)



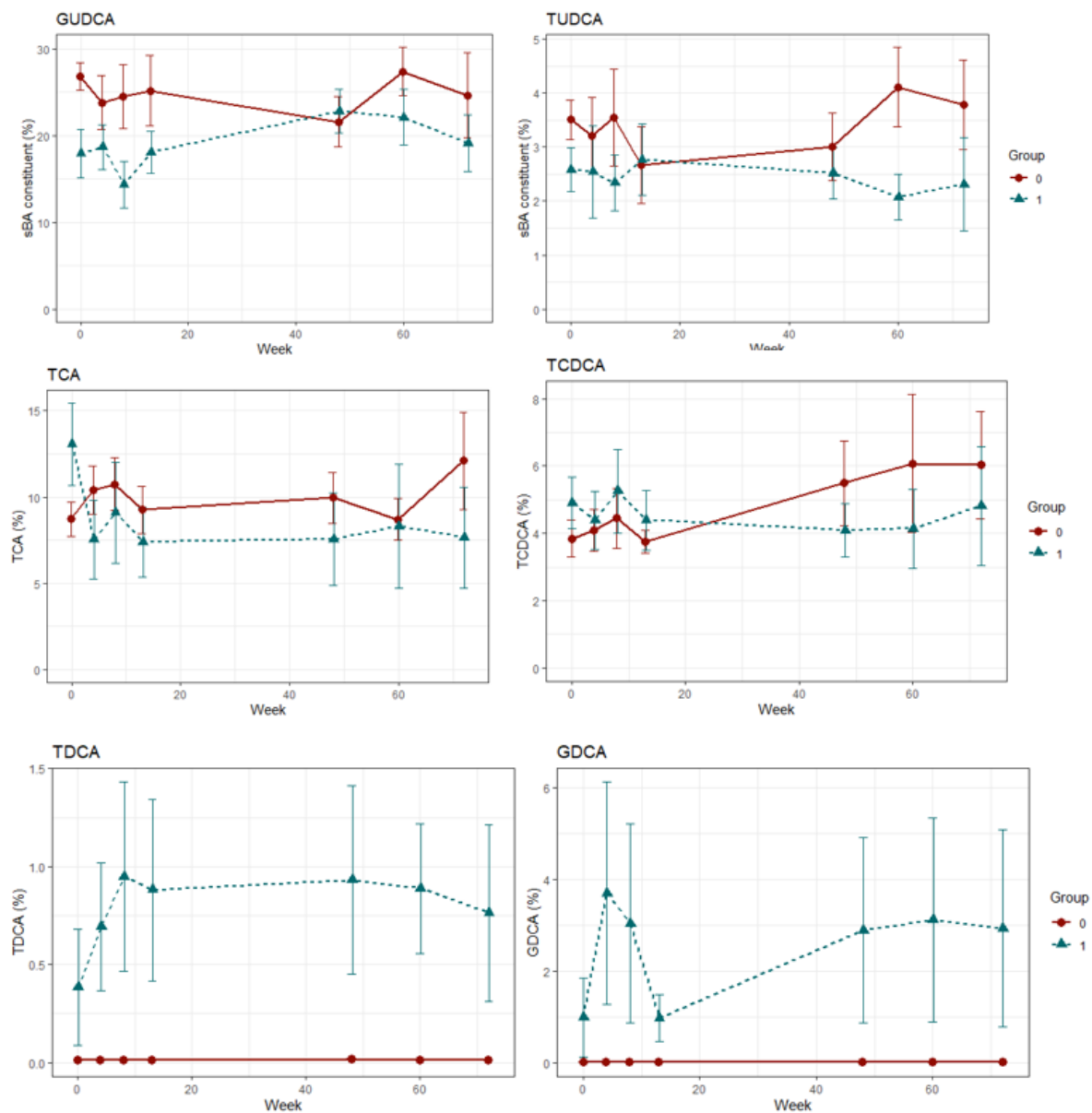
(b)

Supplemental Figure S1. (a) Serum bile acid profiling by UHPLC-MS/MS, total 15 BA subspecies quantified in the established assay. CHA, cholic acid; CDCA, chenodeoxycholic acid; DCA, deoxycholic acid; LCA, lithocholic acid; GCA, glycine-conjugated cholic acid; GCDCA, glycochenodeoxycholic acid; GDCA, glycodeoxycholic acid; GLCA, glycolithocholic acid; GUDCA, glyoursodeoxycholic acid; TCA, taurine-conjugated cholic acid; TCDCA, taurochenoxycholic acid; TDCA, taurodeoxycholic acid; TLCA, tauroolithocholic acid; TUDCA, taoursodeoxycholic acid; UDCA, ursodeoxycholic acid. (b) Representative bile acid tandem MS mass spectra used in quantification in the established assay. Glycine-conjugated cholic acid (GCA) and taurine-conjugated cholic acid (TCA) were shown as examples.

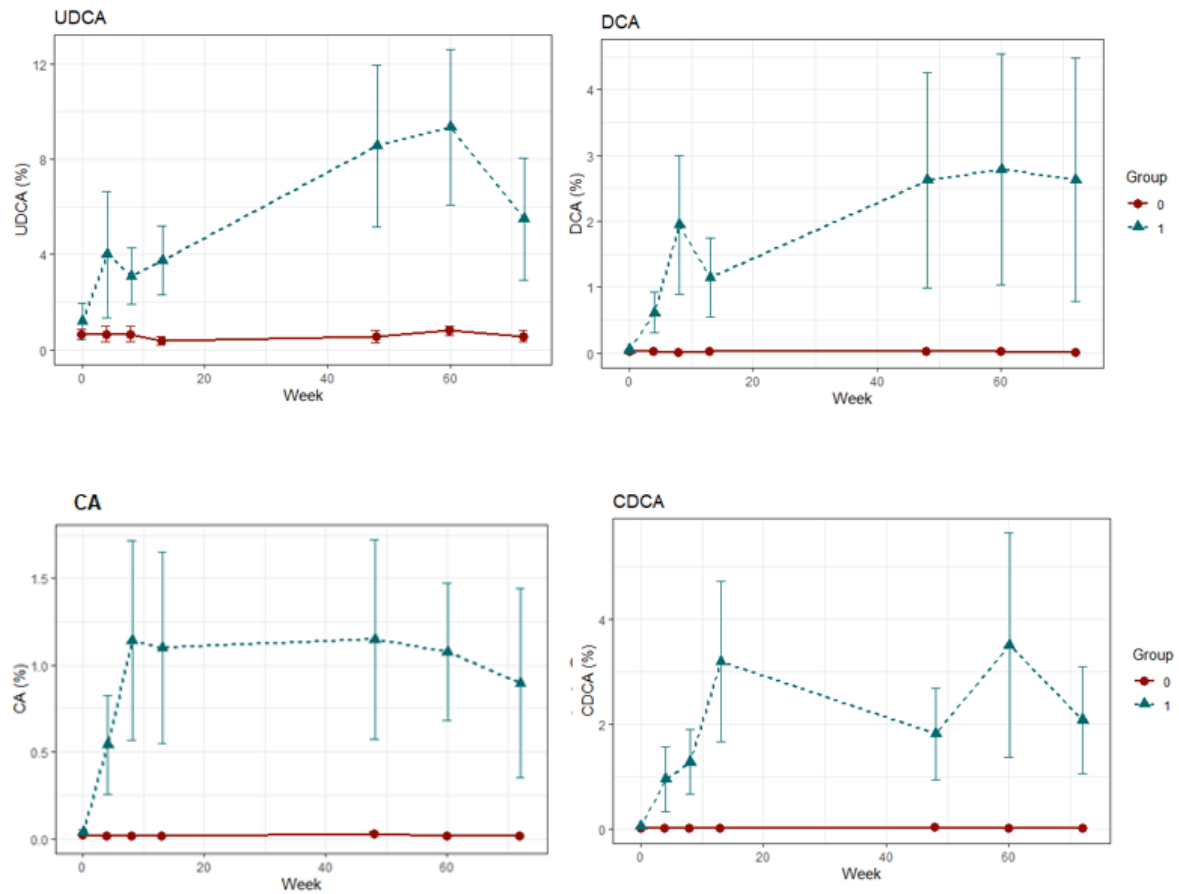
LC-ESI-MS/MS measurement of serum 7 α -Hydroxy-4-cholesten-3-one



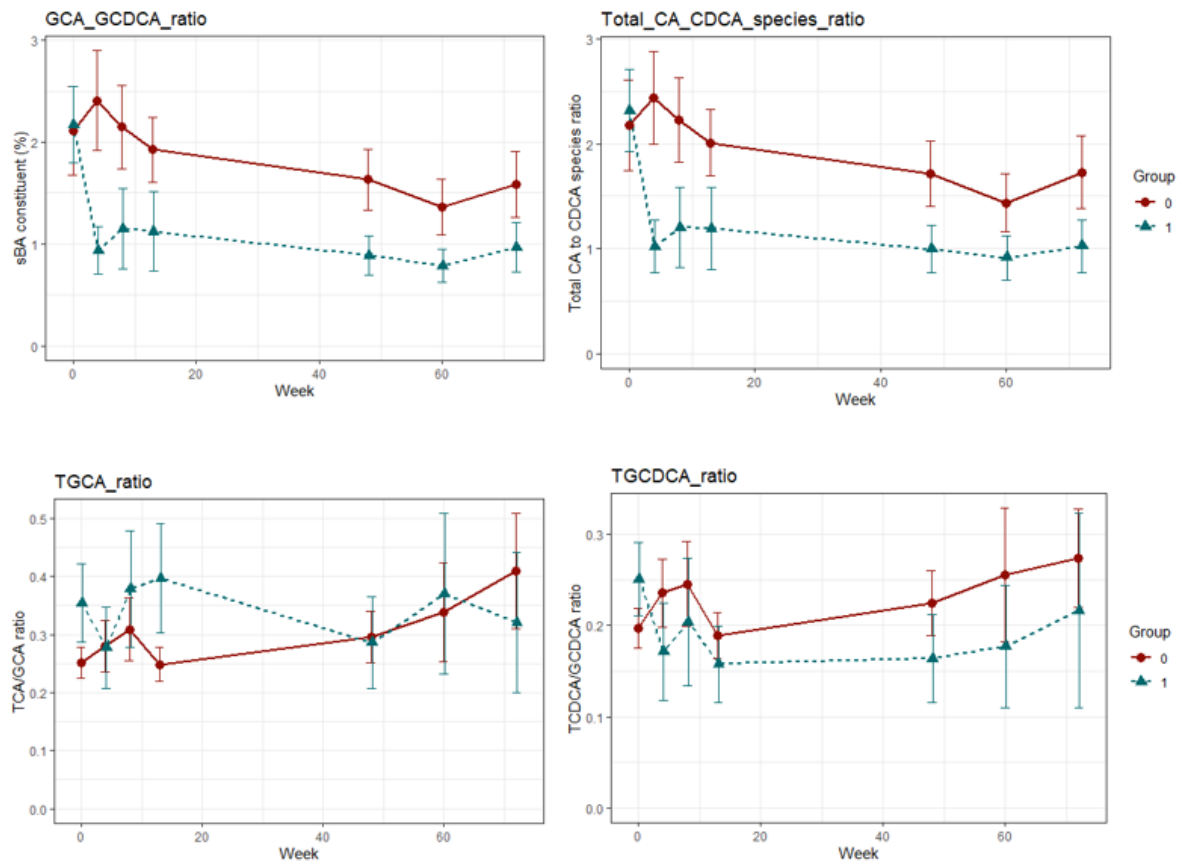
Supplemental Figure S2. Representative chromatogram of serum C4 profiling by UHPLC-MS/MS



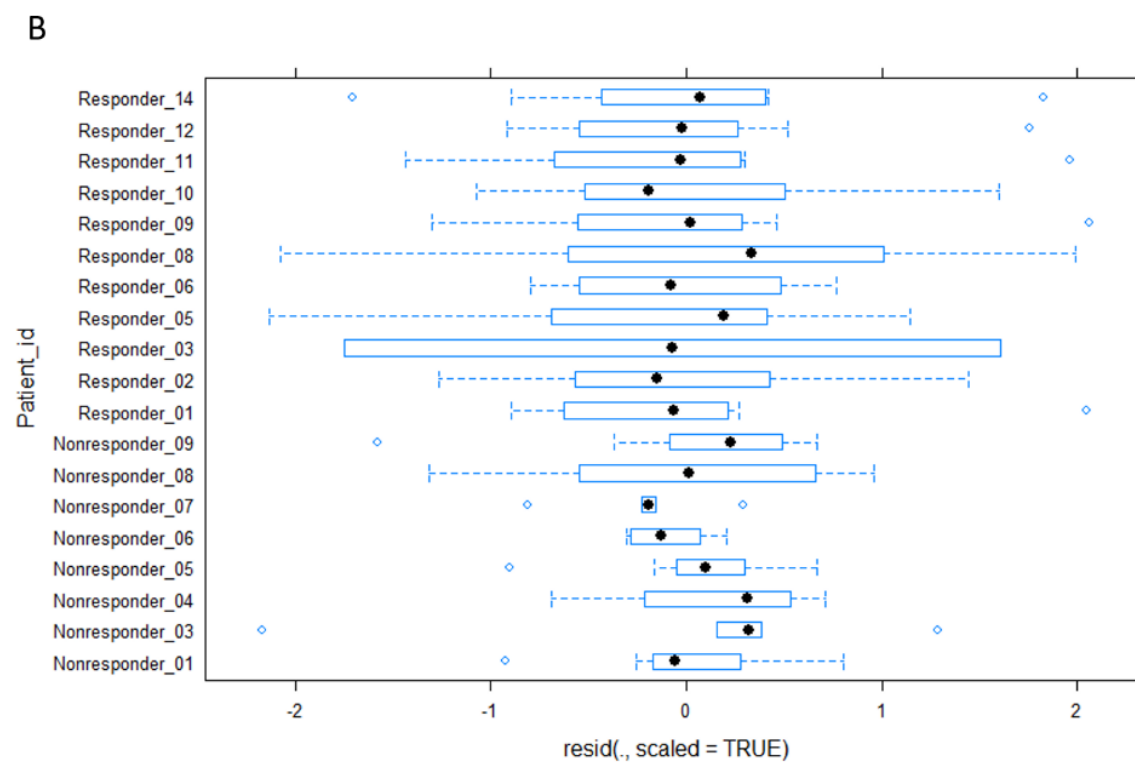
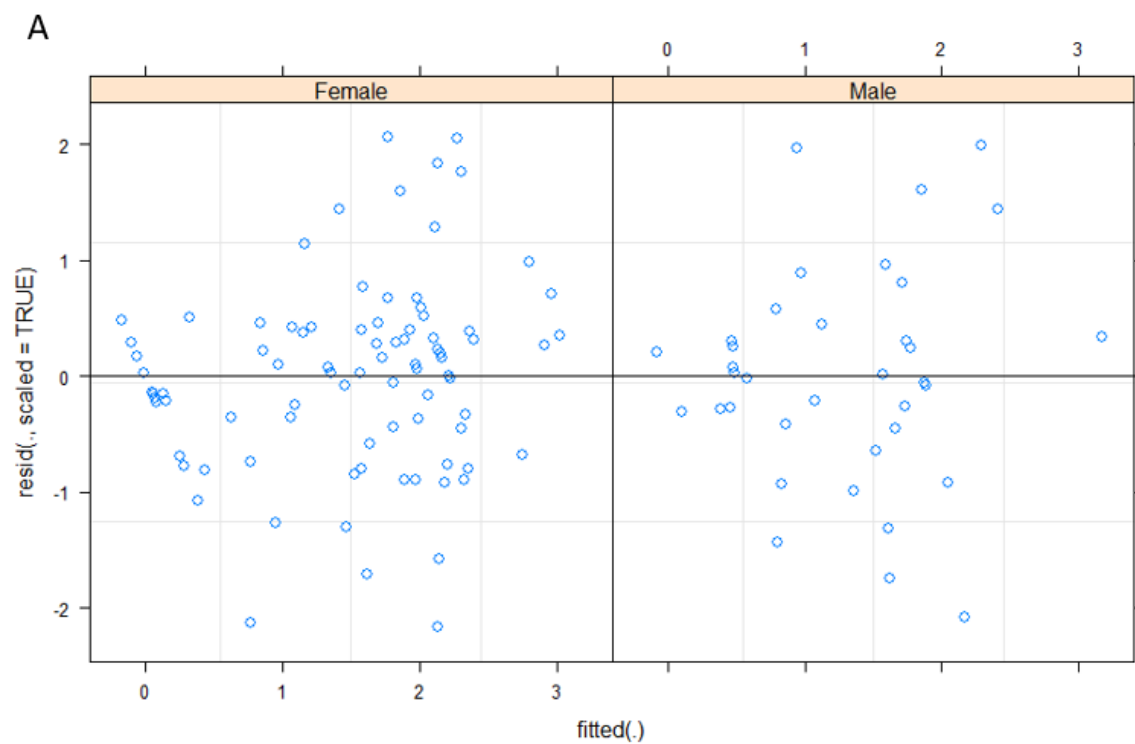
Supplemental Figure S3. Change in conjugated serum bile acid composition (%) over 72 weeks of marlixibat treatment in pruritus responders vs non-responders. Group 0-pruritus nonresponder, 1-pruritus responder

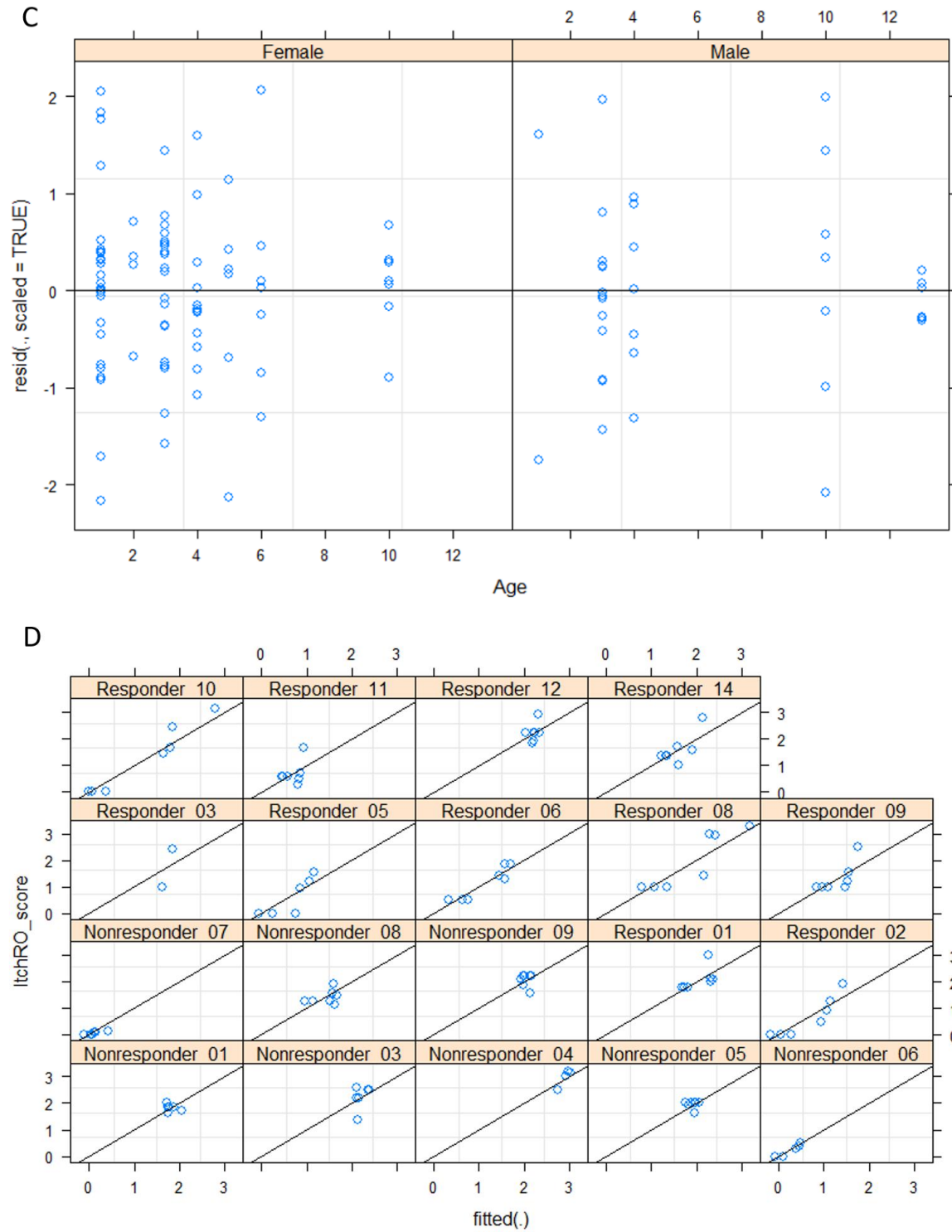


Supplemental Figure S4. Change in unconjugated serum bile acid composition (%) over 72 weeks of maralixibat treatment in pruritus responders vs non-responders. Group 0-pruritus nonresponder, 1-pruritus responder

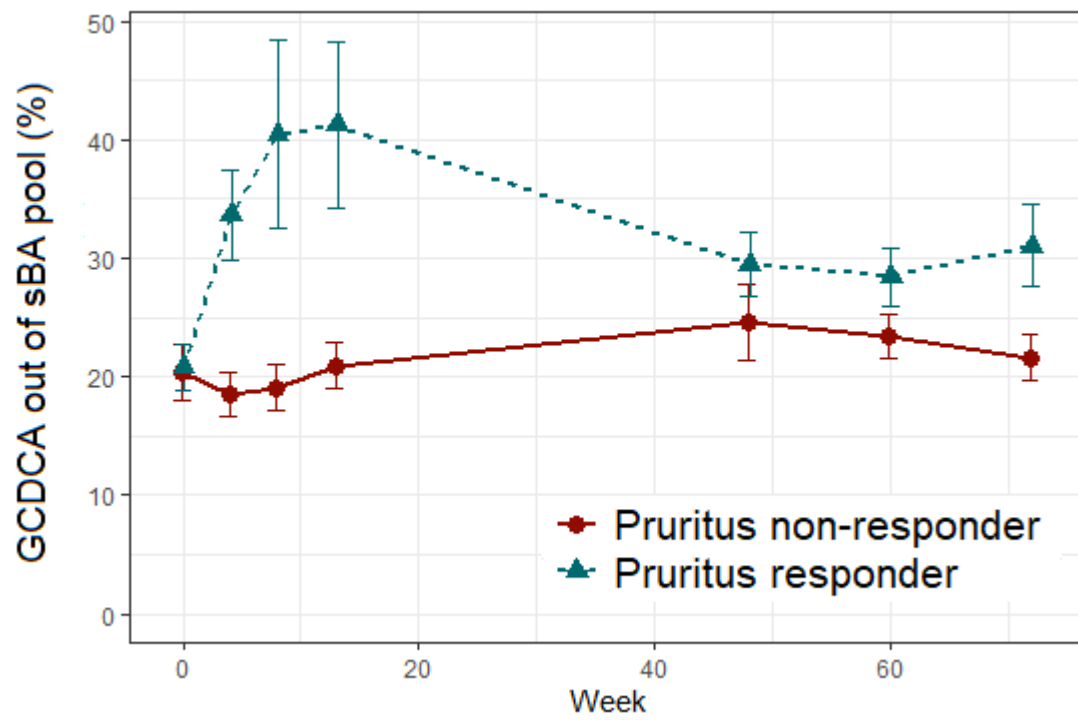


Supplemental Figure S5. Change in serum bile acid composition (%) relationship (ratio) over 72 weeks of maralixibat treatment in pruritus responders vs non-responders. Group 0-pruritus nonresponder, 1-pruritus responder





Supplemental Figure S6. (A) Residuals from LMM model plotted by gender; (B) LMM model residuals from each patient plotted based on pruritus responders vs non-responders; (C) Residuals from LMM model plotted by gender and age; (D) LMM model fitted outcome variable from each patient plotted comparatively to original data



Supplemental Figure S7. Profile of GCDCA composition (%) over 72 weeks in PFIC2 patients under MRX treatment