

## Supplemental information

### **A newborn screening method for cerebrotendinous xanthomatosis using bile alcohol glucuronides and metabolite ratios**

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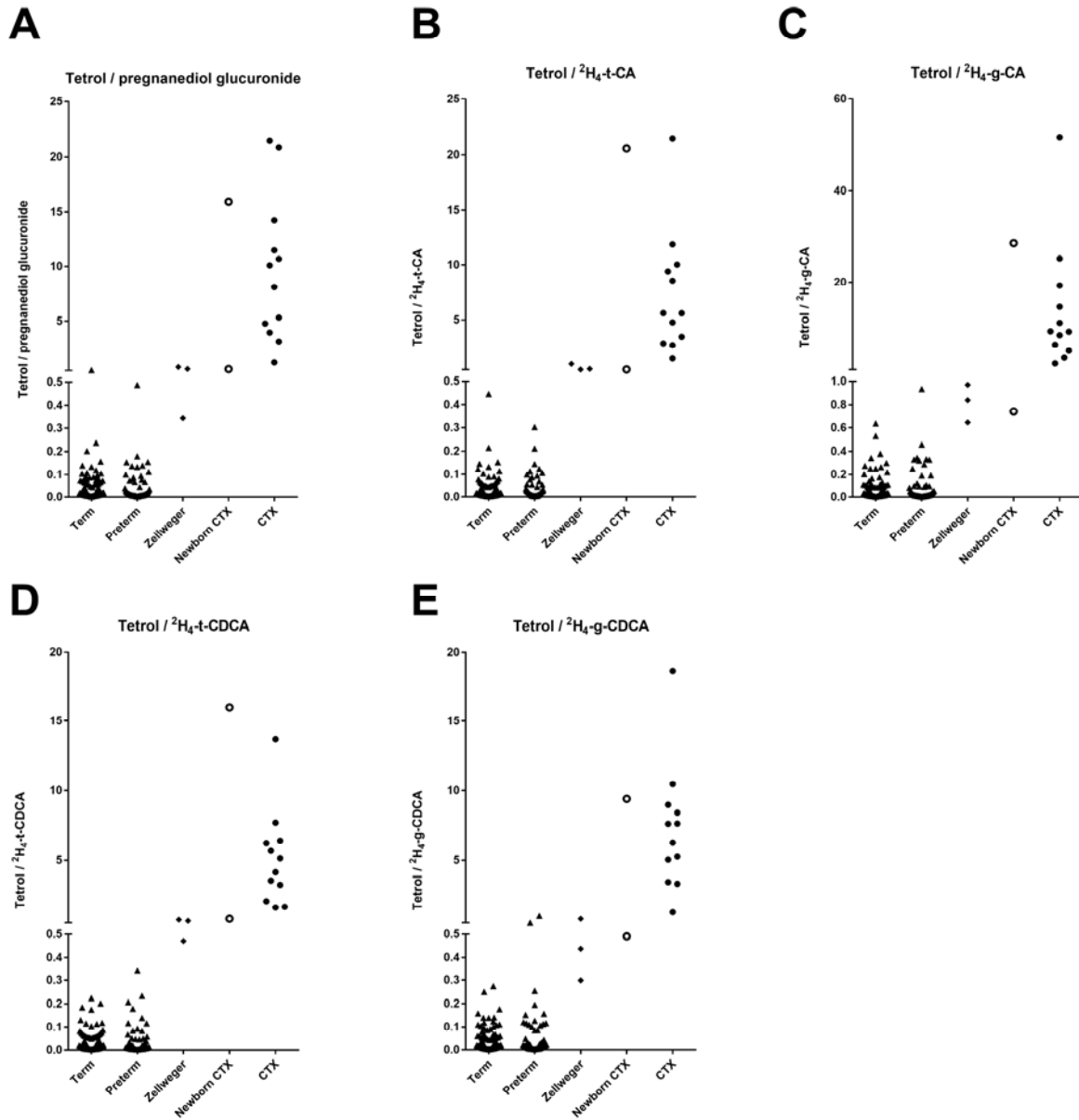
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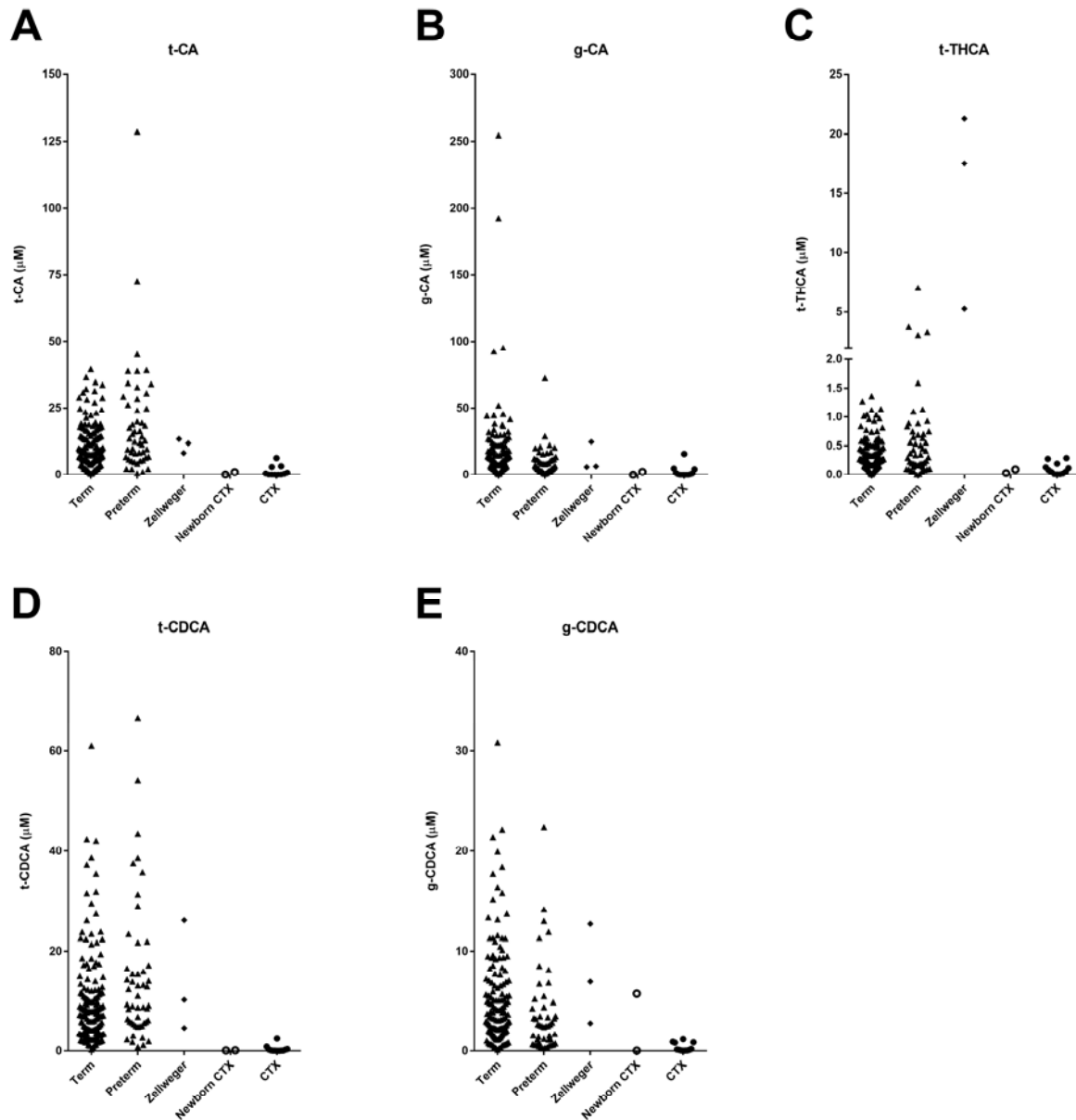
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Supplemental figure S1



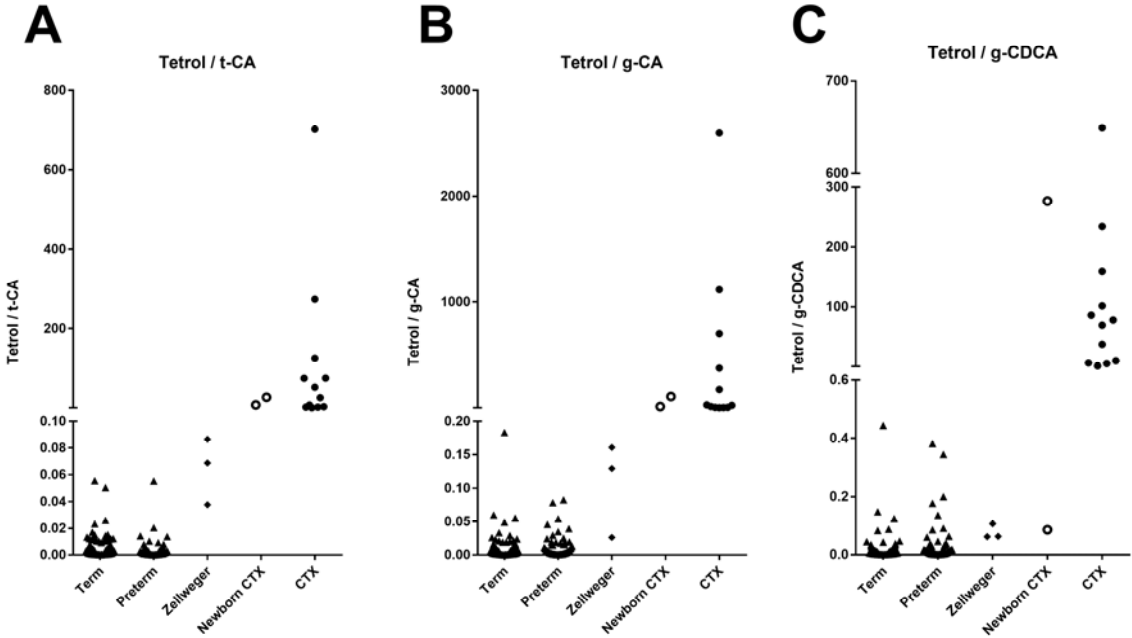
Tetrol response in term/preterm controls (triangle), Zellweger (diamond), newborn CTX and untreated CTX patients (open/closed circle) calculated using different internal standards, (A) pregnanediol glucuronide (B)  $^2\text{H}_4\text{-t-CA}$  (C)  $^2\text{H}_4\text{-g-CA}$  (D)  $^2\text{H}_4\text{-t-CDCA}$  and (E)  $^2\text{H}_4\text{-g-CDCA}$ . Despite a clear elevation of the tetrol response for all used internal standards, the separation of controls and CTX patients was not satisfactory. Zellwegers showed a considerably increased tetrol response which overlapped with the CTX range.

## Supplemental figure S2



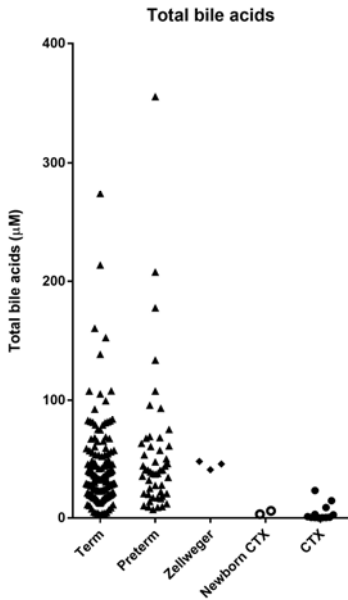
Concentrations of bile acid and the bile acid intermediate t-THCA in term/preterm controls (triangle), Zellweger (diamond), newborn CTX and untreated CTX patients (open/closed circle) calculated different internal standards, (A) t-CA, (B) g-CA, (C) t-THCA, (D) t-CDCA and (E) g-CDCA. Bile acid and t-THCA concentrations in CTX patients were at the low end of the control range. As expected, t-THCA clearly accumulated in Zellweger patients.

Supplemental figure S3



Other investigated metabolite ratios in term/preterm controls (triangle), Zellweger (diamond), newborn CTX and untreated CTX patients (open/closed circle): (A) tetrol/t-CA, (B) tetrol/g-CA and (C) tetrol/g-CDCA. All were less discriminative than the tetrol/t-CDCA ratio.

Supplemental figure S4



Total bile acid levels in term/preterm controls (triangle), Zellweger (diamond), newborn CTX and untreated CTX patients (open/closed circle).