Table 1. MRM transition energies to quantitate metabolites

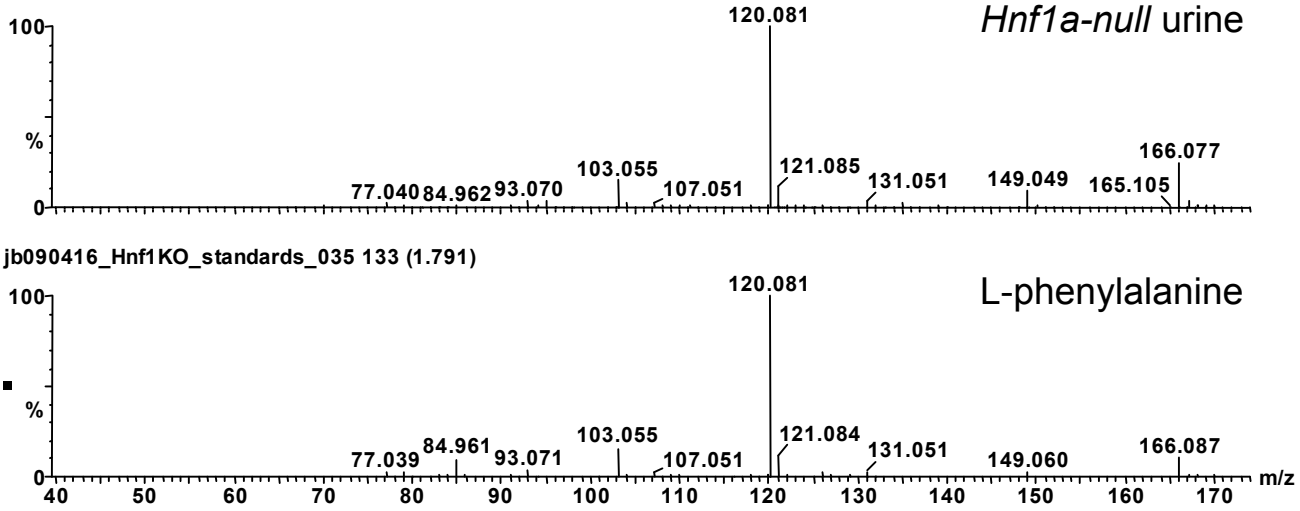
Compound	MRM Transition (*m/z*) E	SI Mode
N-Acetyl-L-phenylalanine	208.1→120.0	Positive
Creatinine	114.0→86.0	Positive
DL-3-Indolelactate	204.0→158.0	Negative
N-Phenylacetylglycine	192.0→73.9	Negative
L-Phenylalanine	166.0→103.0	Positive
DL-3-Phenyllactic acid	164.9→103.0	Negative
Riboflavin	377.2→172.1	Positive
Xanthurenic Acid	206.0→132.0	Positive

Figure 1. Authentication of metabolites identified in urine of wild-type and *H nf1a-null* mice. MSMS fragmentation patterns (A-J) were compared against the indicated authentic standard of the highest quality available. For each panel, the upper MSMS spectrum is from the indicated urine sample and the lower spectrum is of the standard. Panels A-H are spectrums for ions increased in *Hnf1a-null* mice and panels I and J are ions decreased in *Hnf1a-null* mice. All spectrums were acquired in positive mode with the exception of DL-3-phenyllactic acid. Retention time and TOFMS is indicated for each urine and standard comparison.

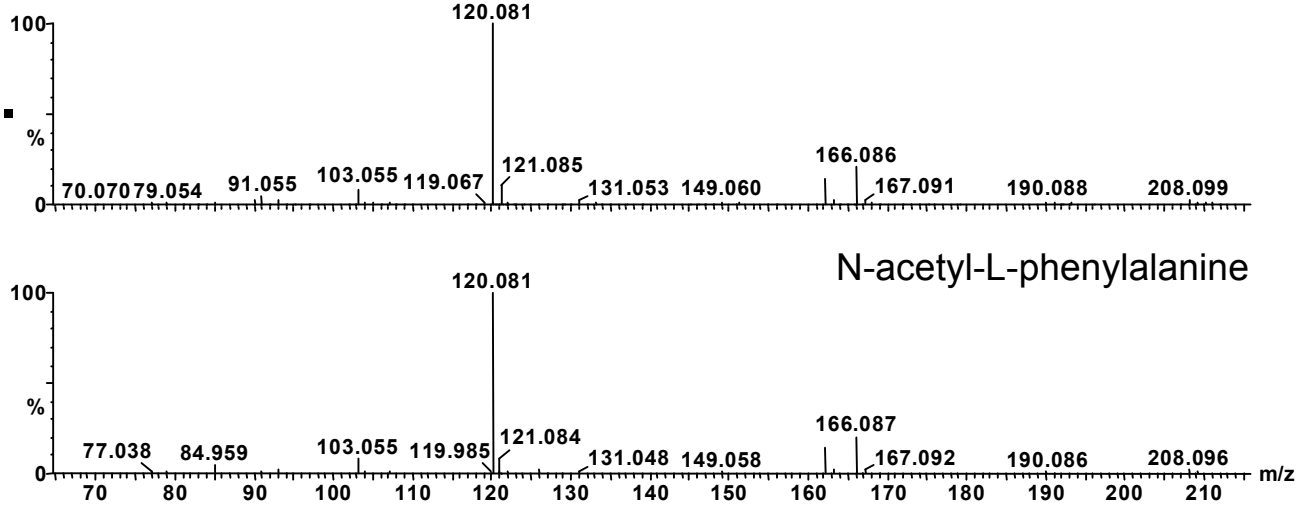
Figure 2. Plasma ACTH concentration. Plasma was collected by retroorbital bleeding from *Hnf1a-null* mice within 3 hours of the initiation of the light cycle. A highly sensitive and specific ELISA assay for the ACTH peptide (Cat#AC562T-100; Calbiotech Inc., Spring Valley, CA) was used to quantify ACTH. Horizontal bars are the average concentration and whiskers represent the 5-95% confidence interval. One sample in each genotype (indicated by red dot) was determined to be an outlier based on the confidence interval as well as the Grubbs' Test. The average ACTH concentration in wild-type mice was 158 pg/mL compared to 48.02 pg/mL in *Hnf1a-null* mice when all samples are included ($p=0.199$). When outliers are removed, the average wild-type ACTH concentration is 73.3 pg/mL compared to 31.58 pg/mL in *Hnf1a-null* mice ($p=0.027$).

Supplemental Figure 1.

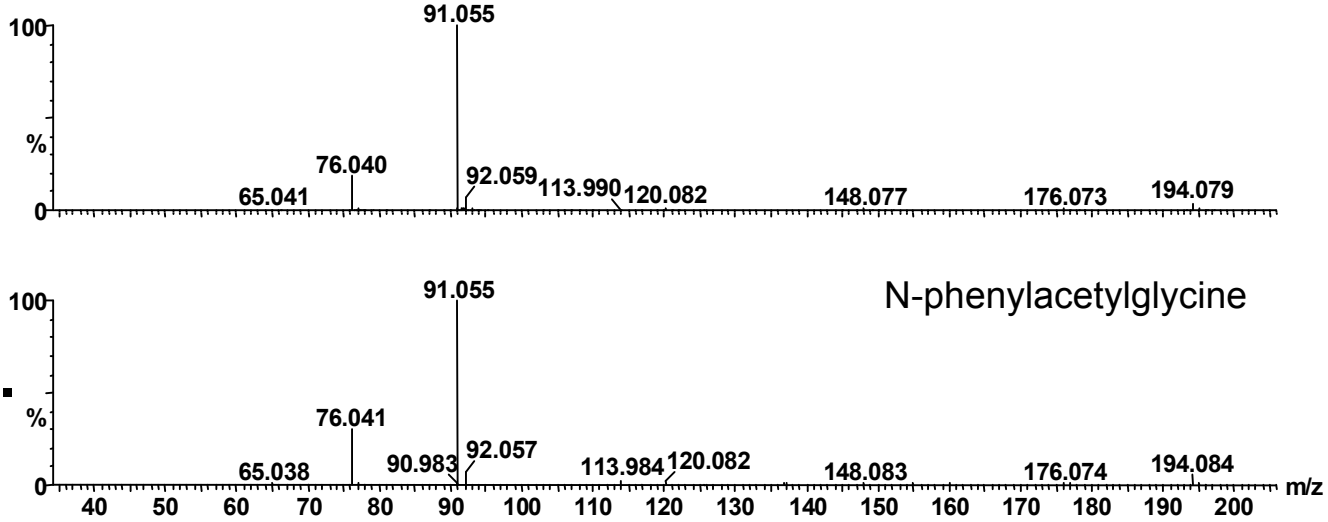
A. Ret. Time 1.76 min, TOFMS 166.07ESI+



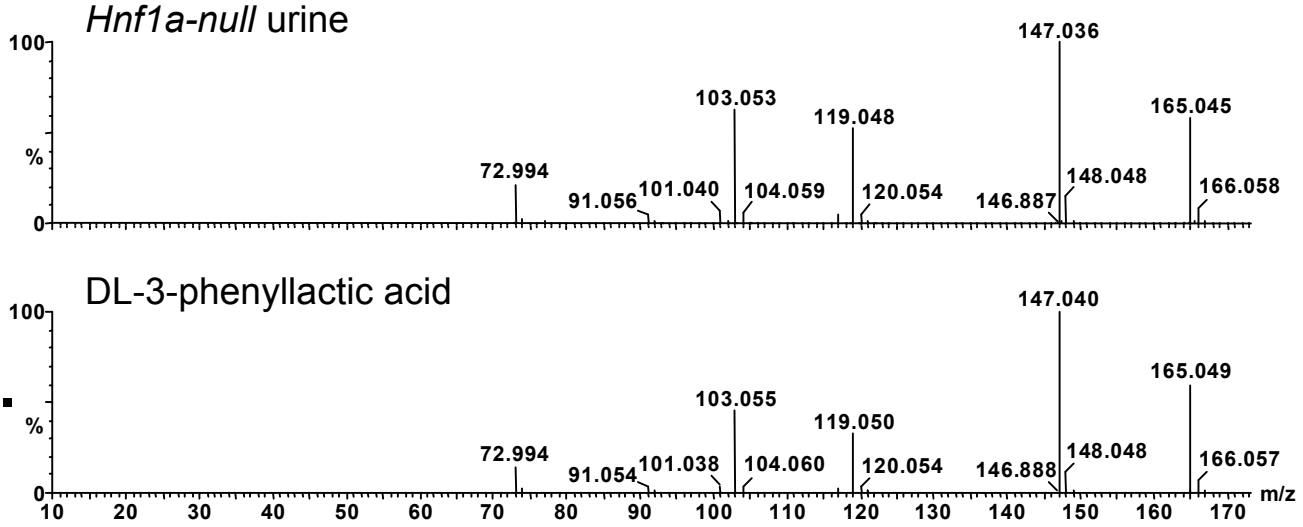
B. Ret. Time 3.78 min, TOFMS 208.08 ESI+



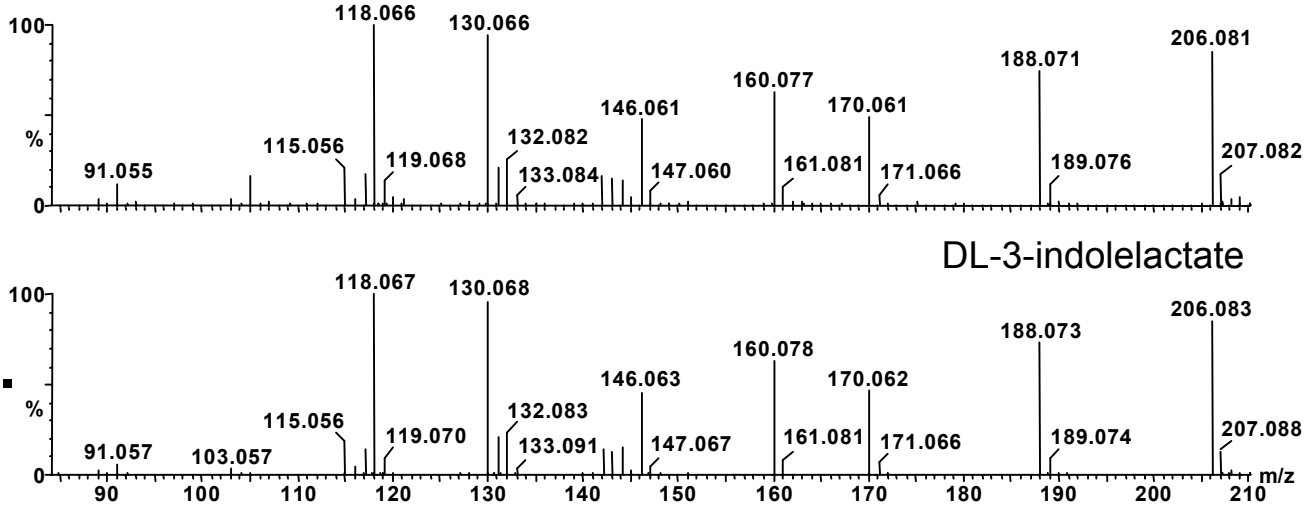
C. Ret. Time 2.98 min, TOFMS 194.07 ESI+



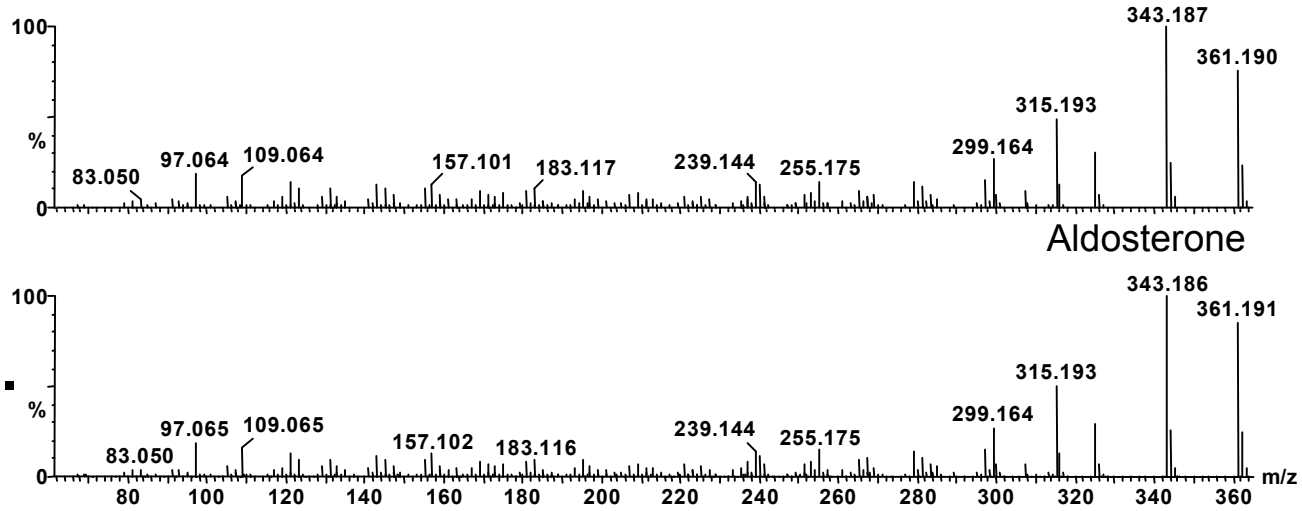
D. Ret. Time 3.44 min, TOFMS 165.06ESI-



E. Ret. Time 3.48 min, TOFMS 206.07 ESI+ *Hnf1a-null* urine

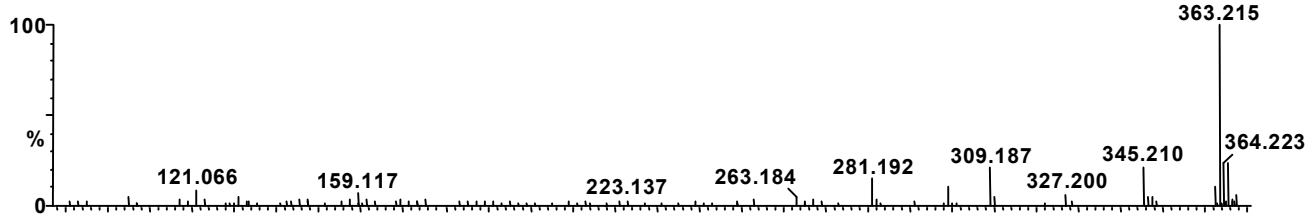


F. Ret. Time 4.98 min, TOFMS 361.20 ESI+ *Hnf1a-null* urine

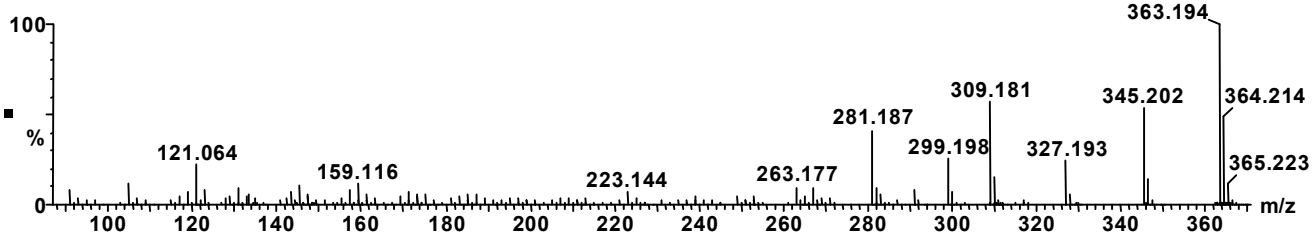


G. Ret. Time 5.24 min, TOFMS 363.00ESI+

Hnf1a-null urine

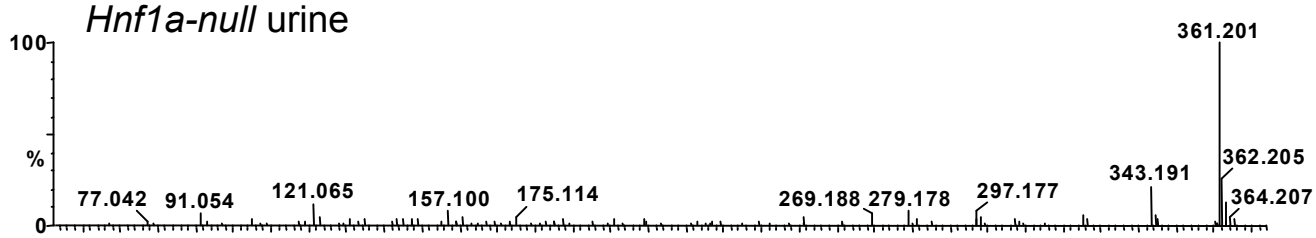


DHOPA

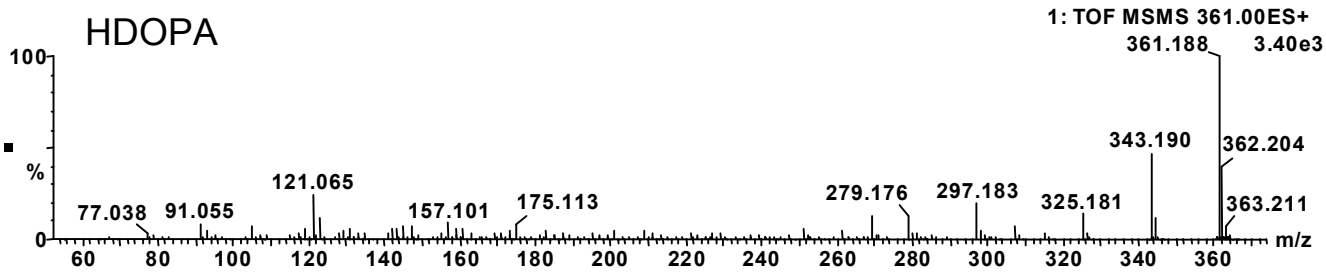


H. Ret. Time 5.28 min, TOFMS 361.00 ESI+

Hnf1a-null urine

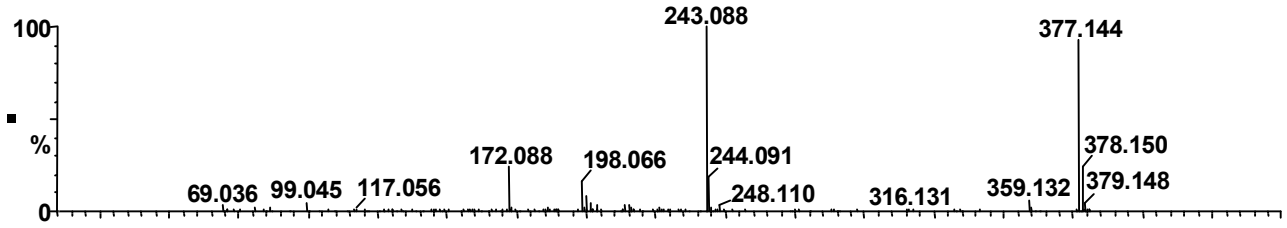


HDOPA

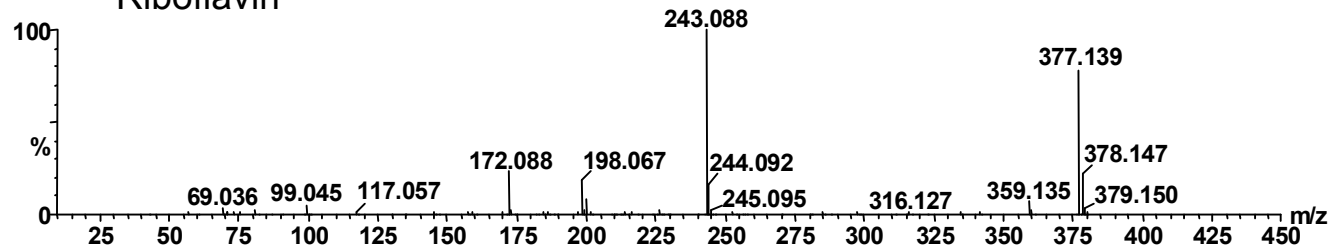


I. Ret. Time 2.88 min, TOFMS 377.20ESI+

Wild-type urine

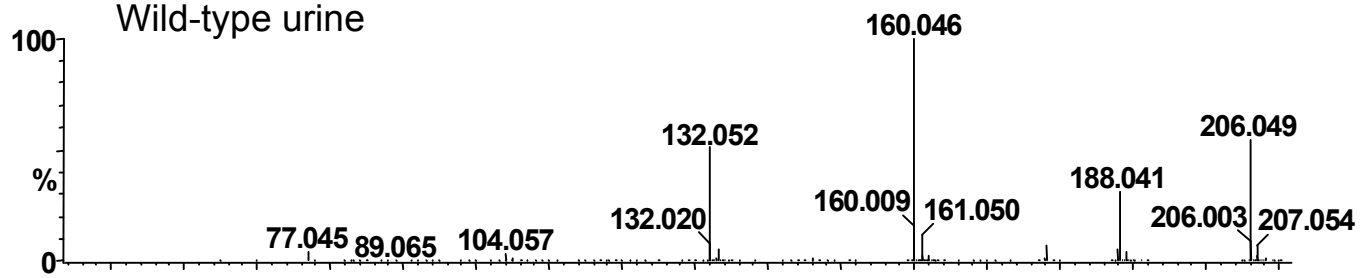


Riboflavin

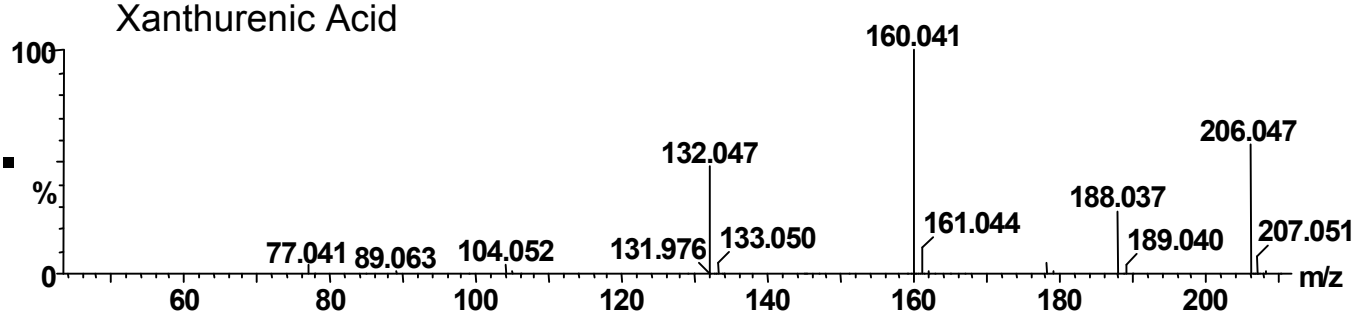


J. Ret. Time 1.79 min, TOFMS 206.07 ESI+

Wild-type urine



Xanthurenic Acid



Supplemental
Figure 2