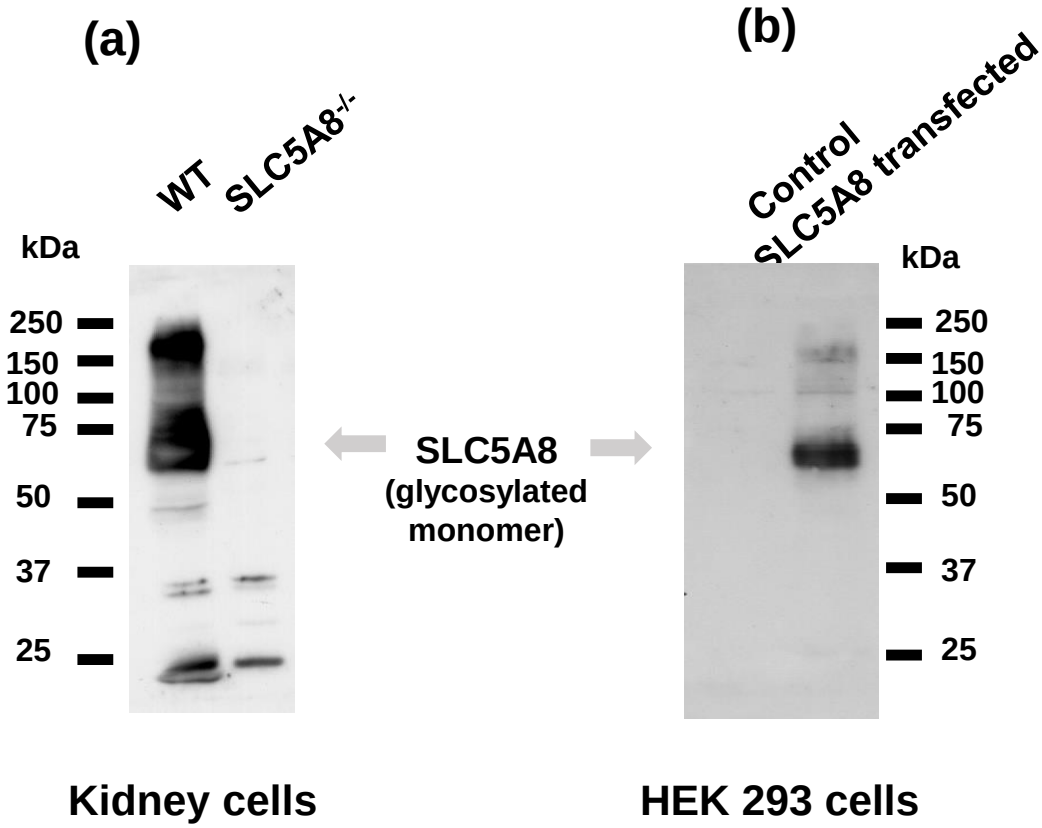
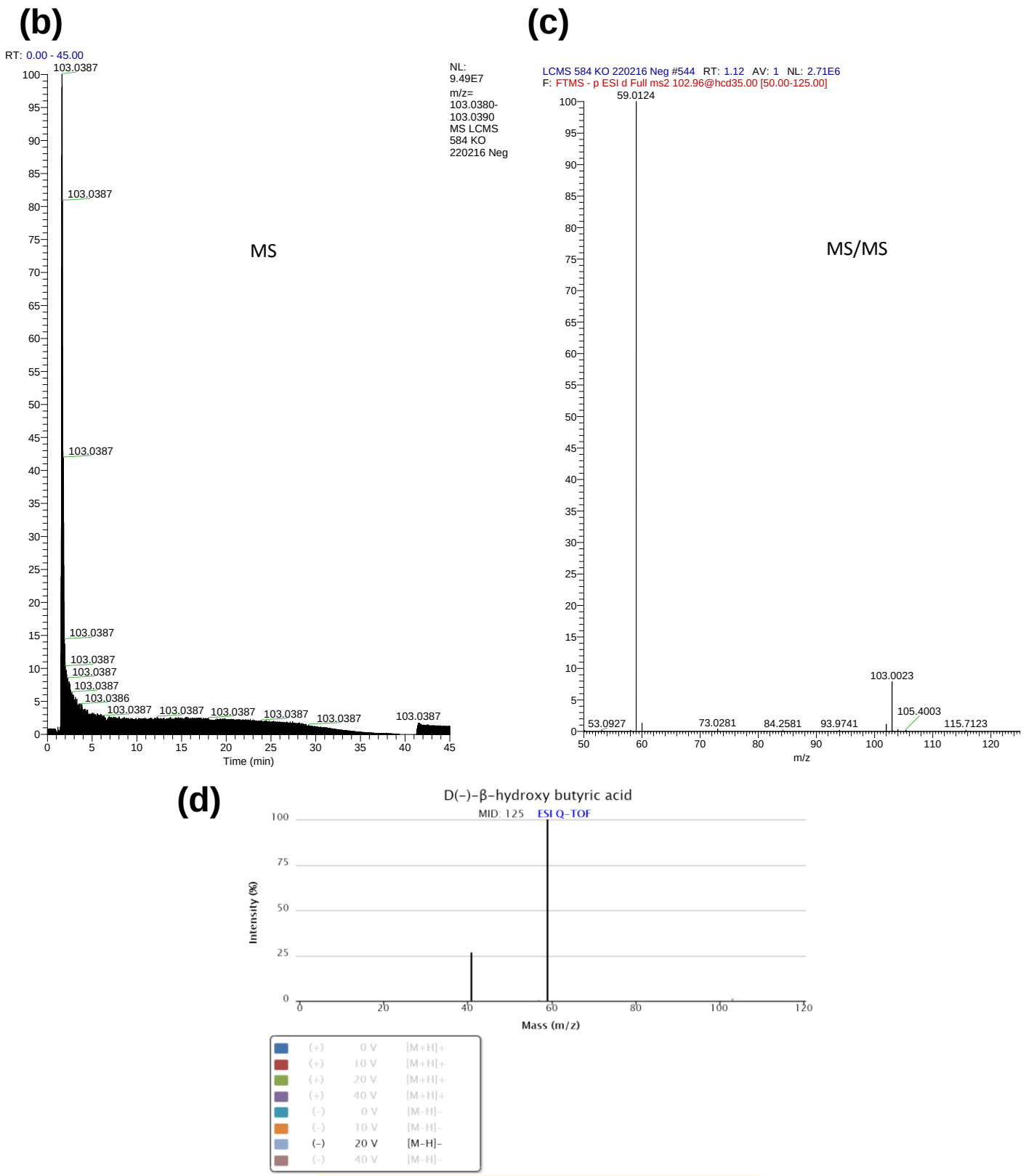


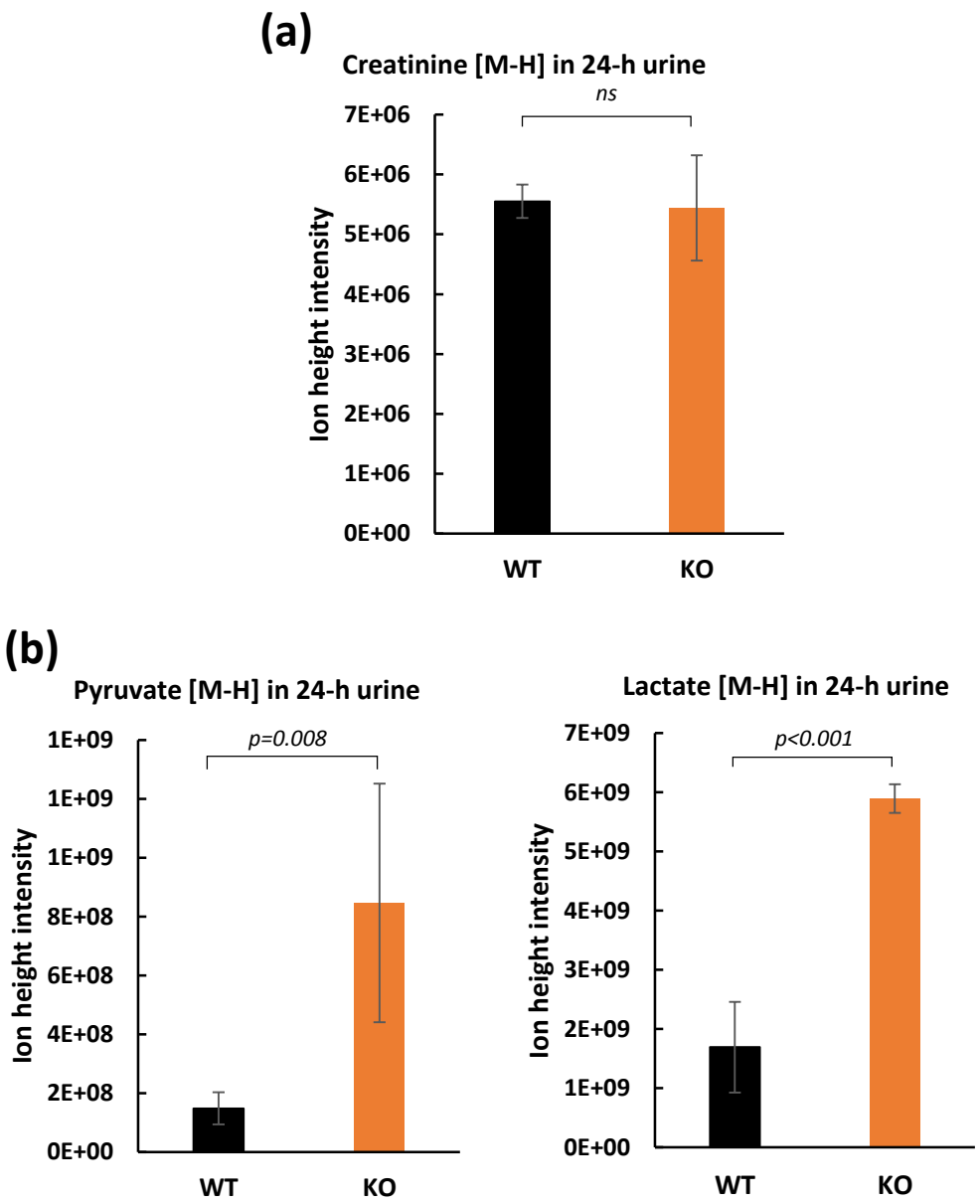
Supplementary figure 1. Western blot analysis of SLC5A8 expression. Membranes were probed with our anti-SLC5A8 antibody (2.32 mg/L, named 4R). 4R is an in house rat monoclonal antibody, raised against a mouse SLC5A8 peptide (amino acids 544 to 610). (a) Analyses were performed with membrane vesicles obtained from whole kidney of control mice (lane WT) and of SLC5A8-deficient mice (lane SLC5A8$^{-/-}$). Membrane vesicles were prepared as described previously, 25 µg proteins per lane were separated on SDS gels and transferred onto PVDF membranes (Huc-Brandt *et al*, 2011, Biochim Biophys Acta.;1808:65-77). (b) Western blot analyses were conducted on membranes obtained from non-transfected HEK 293 cells (lane Control) or transiently transfected HEK 293 cells with mouse SLC5A8-encoding plasmid (Lane mSLC5A8). Proteins (30 µg) were separated on SDS gels and transferred onto PVDF membranes. Main staining was obtained at an apparent molecular weight of about 70 kDa (lane WT (a) and lane mSLC5A8-/- (b)). According to our previous work on the highly SLC5A8-homologous membrane protein NIS (sodium-iodide-symporter, Huc-Brandt *et al*), we assume that this staining corresponds to fully glycosylated SLC5A8 monomers. Staining of protein migrating at higher molecular weights (mainly at about 150 kDa) observed in the same samples is most probably du to oligomeric forms of the SLC5A8 protein as is often observed in western blot studies of membrane proteins (see Huc-Brandt *et al*). The sharp 50-kDa band should then correspond to the non-glycosylated form of SCLC5A8. Staining at 37-kDa and 25-kDa band (a), found in both, WT and SLC5A8-/- lanes, should be du to unspecific staining.

(a)

m/z	Retention time	HMDB ID	Name	Monoisotopic mass	Adduct	Adduct m/z	Delta (ppm)	MS2 Validation
103.0387	1.12	HMDB0000011	Hydroxybutyric acid	104.0473	[M-H]	103.0401	13	Yes



Supplementary figure 2. Mass spectrometry identification of β-hydroxybutyrate. (a) main features of the mass identification in a typical experiment performed on urine samples. (b) Liquid chromatography/mass spectrometry (LC/MS) chromatogram (m/z 103.0387 [M - H]) of a typical experiment. (c) MS/MS mass spectra acquired for m/z 103.0387. The main fragmentation product is observed at m/z 59.0124. (d) The same fragment was described in METLIN metabolomics database source as illustrated by the MS/MS mass spectra for hydroxy butyric acid.

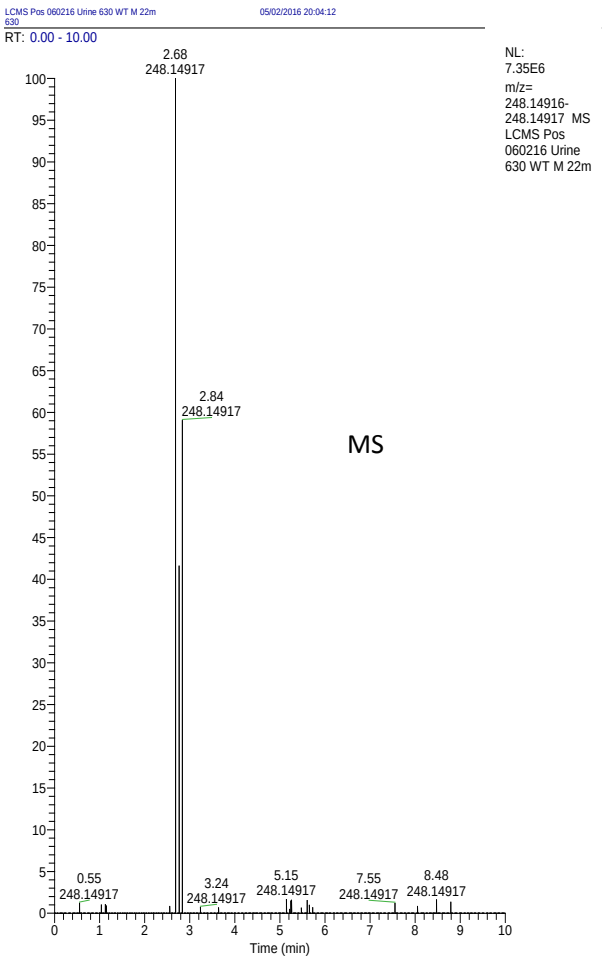


Supplementary figure 3. Level of (a) creatinine [M-H] m/z: 112.0503, (b) pyruvate [M-H] m/z: 87.0073 and (c) lactate [M-H] m/z: 88.0230 in 24-hour urine of 3-month-old wild-type (WT) and 3-month-old SLC5A8-deficient (KO) mice assessed by LC-MS (n=4).

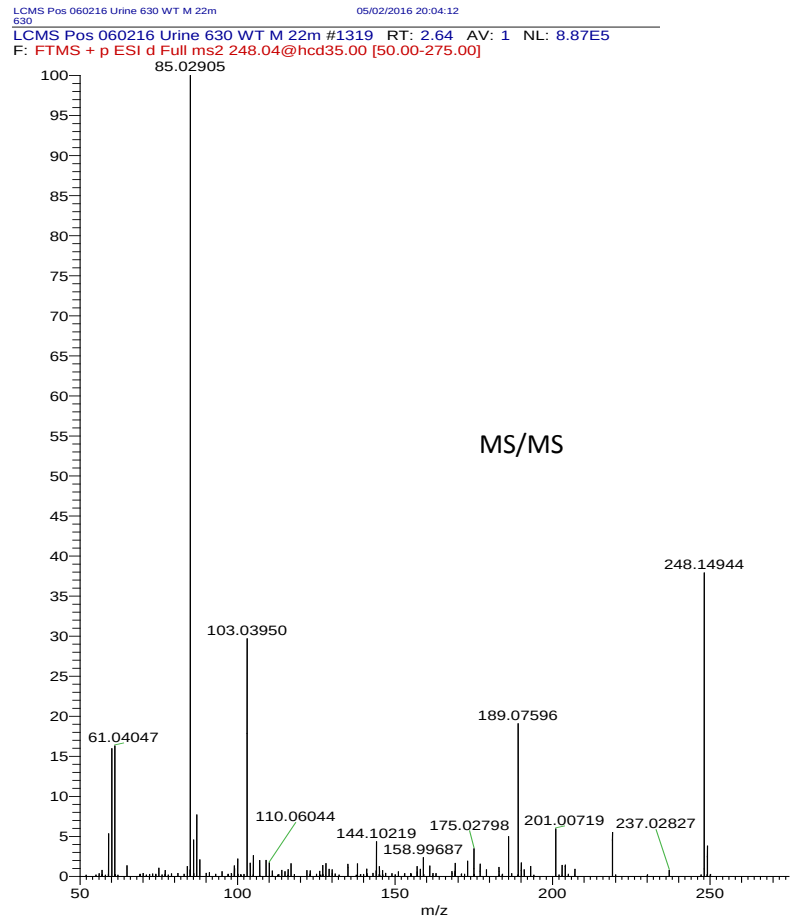
(a)

m/z	Retention time	HMDB ID	Name	Monoisotopic mass	Adduct	Adduct m/z	Delta (ppm)	MS2 Validation
248.1492	2.47	HMDB0013127	Hydroxybutyrylcarnitine	247.1420	[M+H]	248.1492	0	Yes

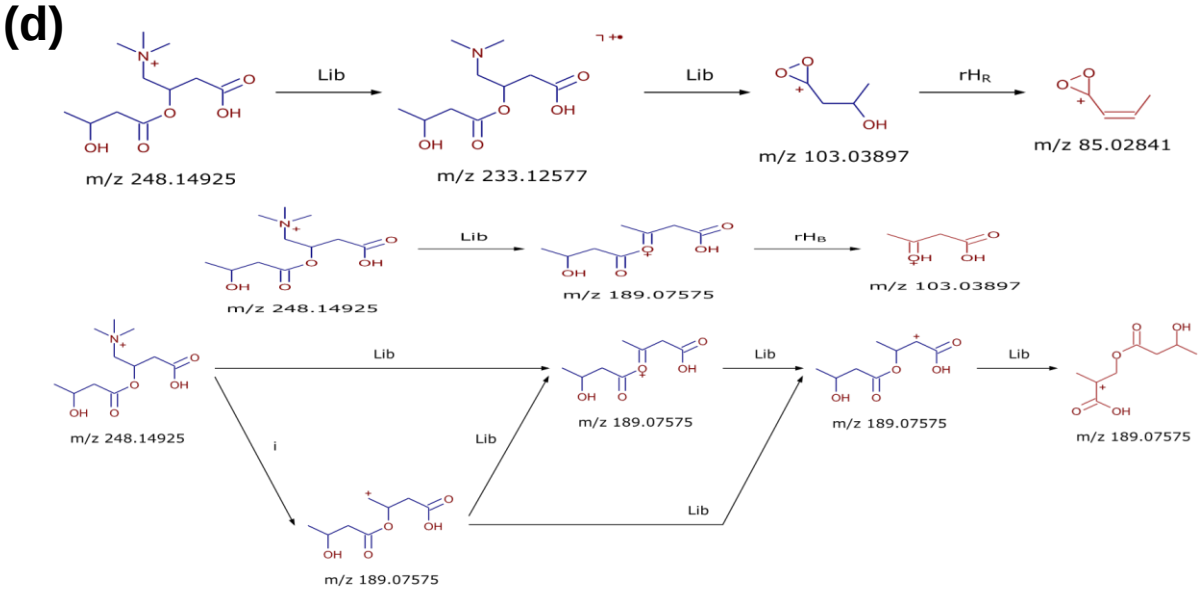
(b)



(c)



(d)

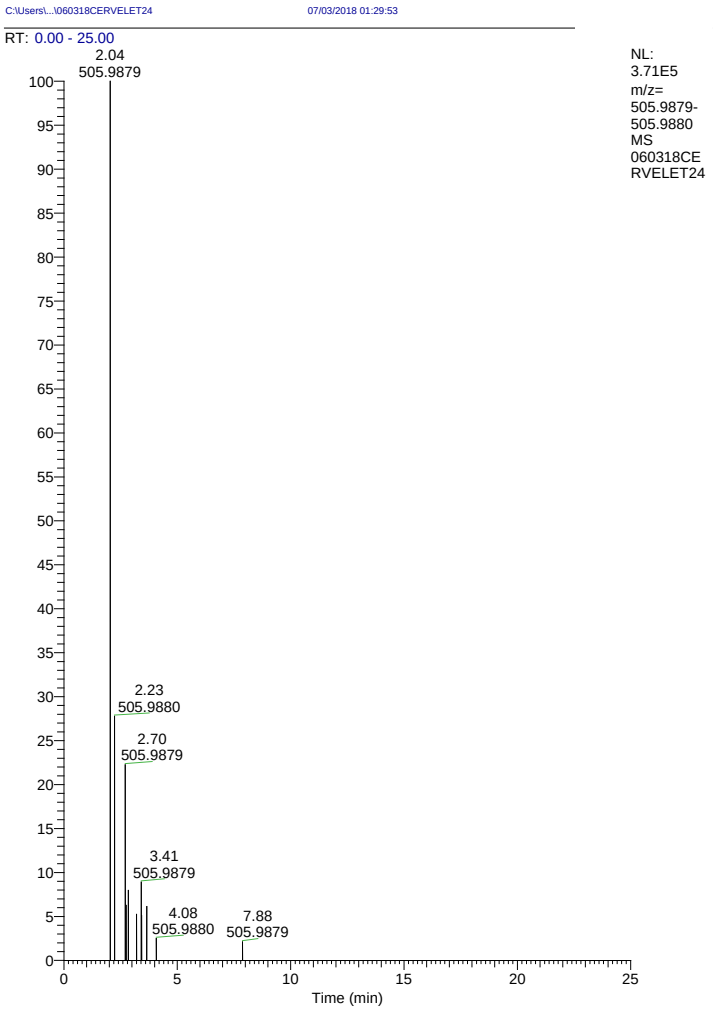


Supplementary figure 4. Identification of Hydroxybutyrylcarnitine by mass spectrometry . (a) main features of the mass identification in a typical experiment performed with urine. (b) Liquid chromatography/mass spectrometry (LC/MS) chromatogram (m/z 248.1492 [M + H]) of a typical experiment. (c) MS/MS mass spectra acquired for m/z 248.1492. (d) Experimental fragmentation of Hydroxybutyrylcarnitine using Mass Frontier software (ThermoFisher).

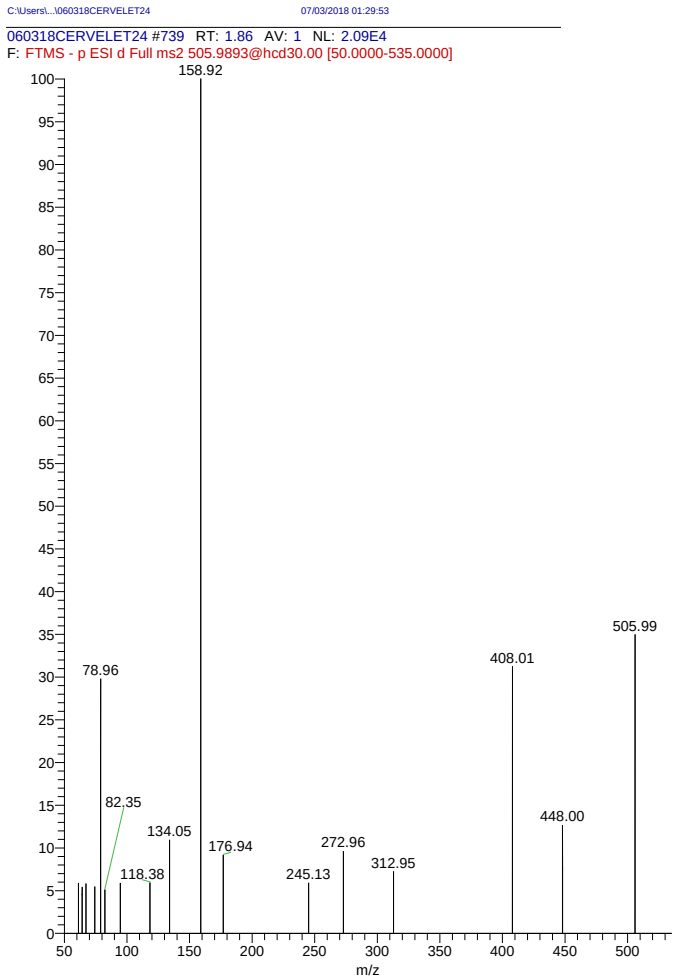
(a)

m/z	Retention time	HMDB ID	Name	Monoisotopic mass	Adduct	Adduct m/z	Delta (ppm)	MS2 Validation
505.9869	2.04	HMDB0000011	Adenosine triphosphate (ATP)	506.9957	[M-H]	505.9885	3	Yes

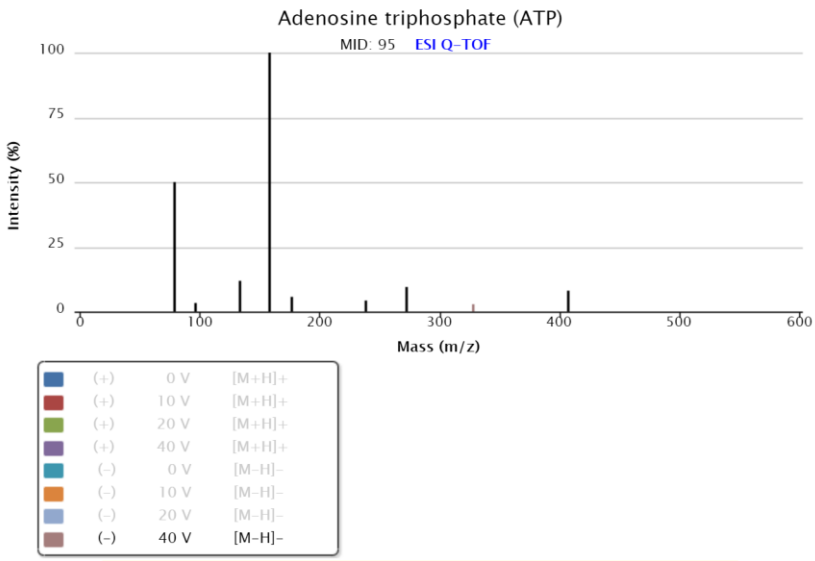
(b)



(c)



(d)

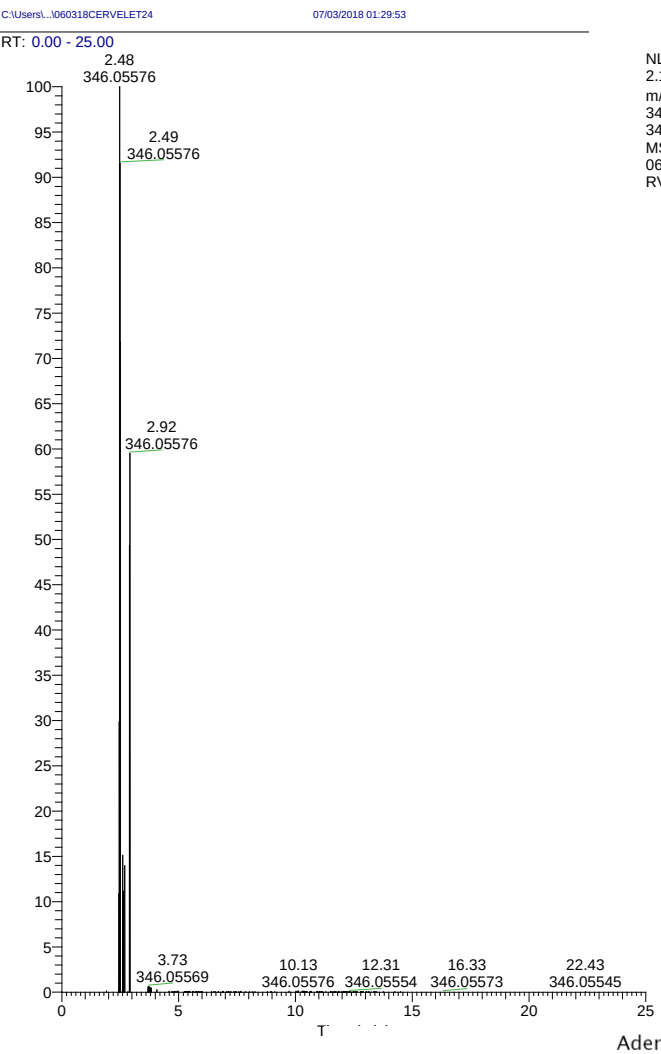


Supplementary figure 5. Mass spectrometry identification of Adenosine triphosphate (ATP). (a) main features of the mass identification in a typical experiment performed on urine samples. (b) Liquid chromatography/mass spectrometry (LC/MS) chromatogram (m/z 505.9869 [M - H]) of a typical experiment. (c) MS/MS mass spectra acquired for m/z 505.9869. (d) The MS/MS mass spectra for ATP in METLIN metabolomics database source.

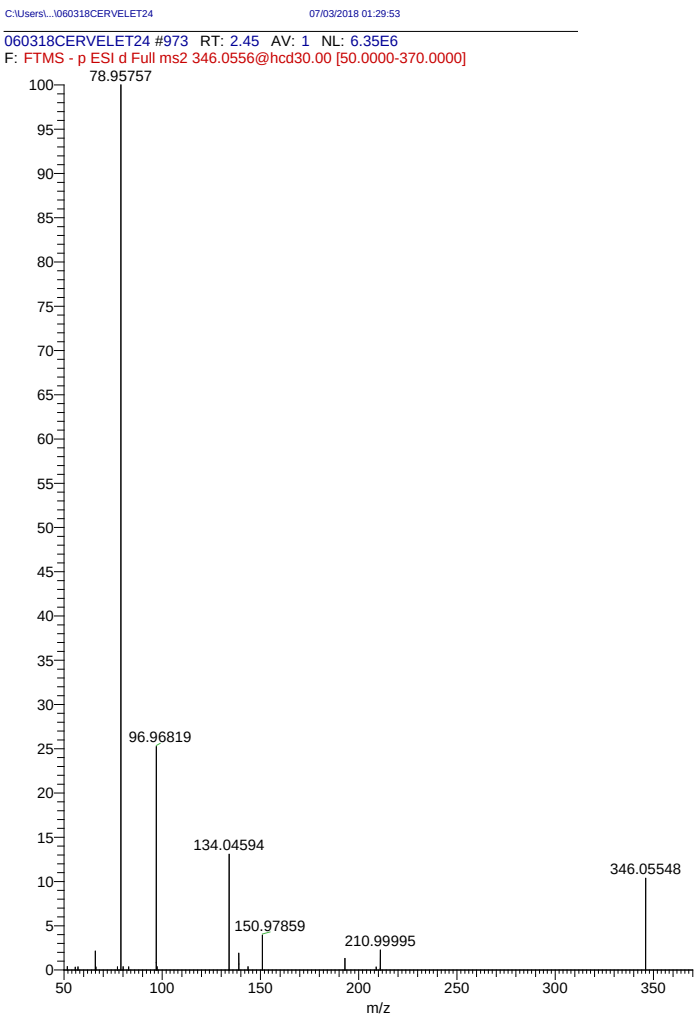
(a)

m/z	Retention time	HMDB ID	Name	Monoisotopic mass	Adduct	Adduct m/z	Delta (ppm)	MS2 Validation
346.0558	2.48	HMDB0000045	Adenosine monophosphate (AMP)	347.0631	[M-H]	346.0558	0	Yes

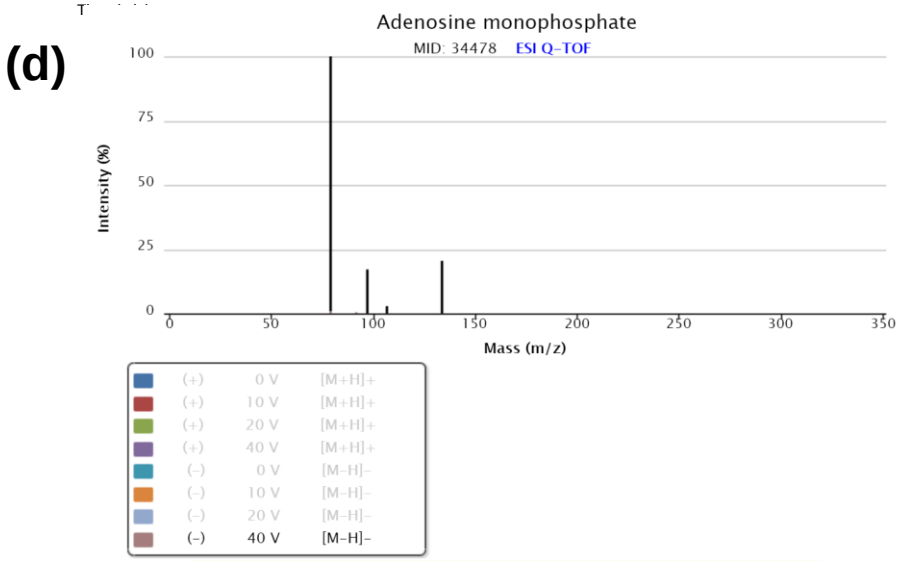
(b)



(c)



(d)



Supplementary figure 6. Mass spectrometry identification of Adenosine monophosphate (AMP). (a) main features of the mass identification in a typical experiment performed on urine samples. (b) Liquid chromatography/mass spectrometry (LC/MS) chromatogram (m/z 346.0558 [M - H]) of a typical experiment. (c) MS/MS mass spectra acquired for m/z 346.0558. (d) The MS/MS mass spectra for AMP in METLIN metabolomics database source.