SUPPORTING INFORMATION

High-throughput indirect quantitation of ¹³C enriched metabolites using ¹H NMR.

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Supporting figures

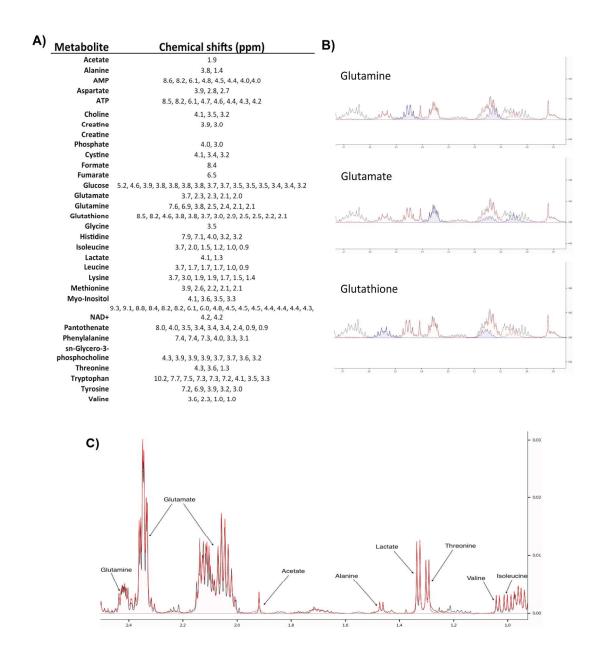


Figure S1. A) Table with all the metabolites fit in the spectra with their chemical shift. **B)** ¹H-NMR spectra of glutamine, glutamate and glutathione. Overlap deconvolution for C4 glutamine, glutamate and glutathione. Observed spectrum and the fitted spectrum are shown with black line and shade purple, respectively.

C) ¹H-NMR spectra of media samples after filtration. Acetate, alanine, isoleucine, lactate, threonine, valine, glutamate and glutamine are highlighted in the spectrum.

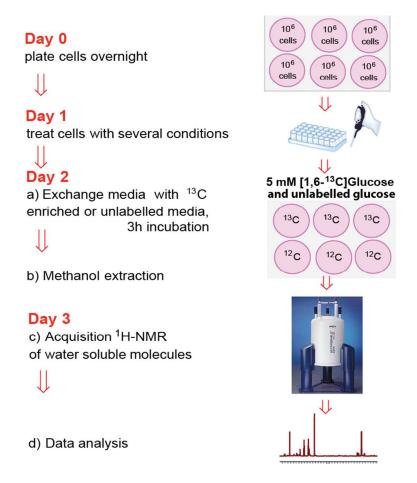


Figure S2. (A) Schematic representation of the experiment procedure for the tracing experiment using [1,6-¹³C]glucose.

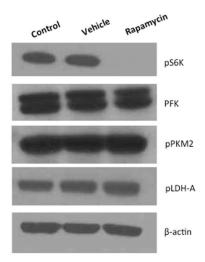


Figure S3. Western blots demonstrating target inhibition by rapamycin in UOK-262 cells. Western blot of tumors treated with either DMSO (vehicle) or 100mg/kg rapamycin for 24 hr (Rapamycin) probed with primary antibody for phospho-S6 at Ser235/236 and actin as a loading control

Acetate	4.38
Alanine	2.33
Aspartate	2.63
Creatine	3.50
Creatine phosphate	3.50
Cystine	1.75
Formate	3.00
Glucose	3.06
Glutamate	2.13
Glutamine	3.35
Glutathione	2.57
Glycine	3.06
Histidine	1.17
Isoleucine	2.10
Lactate	3.01
Leucine	2.50
Methionine	2.10
myo-Inositol	2.16
NAD+	3.50
Pantothenate	1.75
Phenylalanine	2.63
sn-Glycero-3-phosphocholine	1.75
Threonine	2.03
Tyrosine	4.67
Valine	2.33

Table S1. Conversion factors to apply for each metabolite analyzed.