MEDICAL SCHOOL METABOLOMICS CORE

Gly/TCA/nucleotide and NAD related metabolites Service Code: NAD

Summary: Profile of Central Metabolism, including glycolysis, pentose-phosphate shunt, TCA cycle, nucleotide pool and NAD related metabolites. One step organic solvent extraction of cultured cells or tissues, separated on a 1mm x150mm HILIC column in a 35 min cycle. All analytes and Internal Standards are measured by ESI⁻ ionization on a LC-QTOF mass spectrometer and reported as uM normalized to wet tissue weight or cell proteins. CV's are generally 15%.

Container: Eppendorf Tube or equivalent Normal Volume: Plasma (100 ul) Tissue (50-100 mgs); Cells (2E7). Minimal Volume: Plasma (50 uL)Tissue (30 mg); Cells (~5E6) Special Handling: If human or primate, note any known presence of infectious agents. Sample Collection: Snap freeze by liquid nitrogen. For tissues, resect and snap-freeze as soon as practical in tared centrifuge tube. Provide both sample weight and tared vial weight on sample submission

Reference: <u>Matthew A. Lorenz</u>, <u>Charles F. Burant</u>, and <u>Robert T. Kennedy</u> (2011) "Reducing Time and Increasing Sensitivity in Sample Preparation for Adherent Mammalian Cell Metabolomics", *Anal. Chem.* 83(9): 3406–3414.

Analyte	Abbr.	Mol Formula	LOQ(uM)
Acetyl-CoA	aCoA	C ₂₃ H ₃₈ N ₇ O ₁₇ P ₃ S	1
Citrate/Isocitrate combined	Cit/i-Cit	$C_6H_8O_7$	1
Succinate	Suc	$C_4H_6O_4$	1
Malate	Mal	$C_4H_6O_5$	1
2-Phosphoglycerate/3-Phosphoglycerate combined	2PG/3PG	C ₃ H ₇ O ₇ P	1
Phosphoenolpyruvate	PEP	$C_3H_5O_6P$	1
Adenosine monophosphate	AMP	$C_{10}H_{14}N_5O_7P$	1
Adenosine diphosphate	ADP	$C_{15}H_{23}N_5O_{14}P_2$	1
Adenosine triphosphate	ATP	$C_{10}H_{16}N_5O_{13}P_3$	1
Flavin adenine dinucleotide	FAD	$C_{27}H_{33}N_9O_{15}P_2$	1
Nicotinamide adenine dinucleotide	NAD	$C_{21}H_{28}N_7O_{14}P_2$	1
Nicotinamide adenine dinucleotide, reduced	NADH	$C_{21}H_{29}N_7O_{14}P_2$	1
Nicotinamide adenine dinucleotide phosphate	NADP	$C_{21}H_{29}N_7O_{17}P_3$	1
Nicotinamide adenine dinucleotide phosphate, reduced	NADPH	$C_{21}H_{30}N_7O_{17}P_3$	1

Table I: Analytes reported:



MEDICAL SCHOOL METABOLOMICS CORE UNIVERSITY OF MICHIGAN MEDICAL SCHOOL

Erythrose 4-phosphate*	E4P	C ₄ H ₉ O ₇ P	1
Ribulose 5-phosphate/Xylulose 5-phosphate/ribose-5-	R5P/X5P/	$C_5H_{11}O_8P$	1
phosphate combined*	Ru5P		
6-phosphogluconate*	6PG	$C_6H_{13}O_{10}P$	1
Sedoheptulose 7-phosphate*	S7P	C ₇ H ₁₅ O ₁₀ P	1
Fructose-6-phosphate + glucose-6-phosphate	F6P/G6P	$C_6H_{13}O_9P$	1
Fructose-bisphosphate	FBP	$C_6H_{14}O_{12}P_2$	1
Nicotinic acid (NA)	NA	$C_6NH_5O_2$	1
Nicotinic acid mononucleotide (NaMN)	NaMN	$C_{11}H_{15}N_2O_8P$	1
Quinolinic acid (QA)	QA	C ₇ H ₅ NO ₄	1
nicotinamide mononucleotide (NMN)	NMN	$C_{11}H_{15}N_2O_8P$	1
nicotinamide (NAM)	NAM	C ₆ H ₆ N ₂ O	1

*Metabolites are low concentrations and below detection limit in some samples.