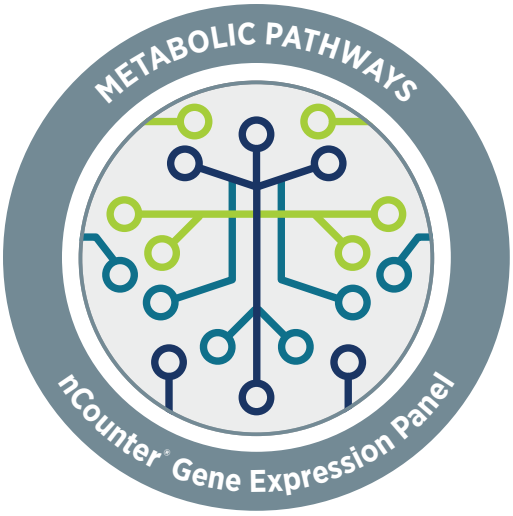
nCounter® Metabolic Pathways Panel

Gene Expression Panel

Cancer Metabolism • Immunometabolism • Metabolic Disease

Quantify the expression of hundreds of genes involved in core metabolic processes and signaling pathways in the context of cancer and immunity. The underlying molecular mechanisms behind alterations in metabolic pathways, signaling pathways, and cell stress can now be fully elucidated, giving researchers a complementary tool to traditional metabolite assays for profiling metabolic checkpoints and discovering potential therapeutic targets.



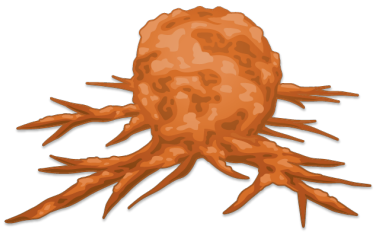
Product Highlights

- Profile 768 genes in human and mouse across 34 annotated pathways involved in five important themes for metabolism research
  - Biosynthesis and Anabolic Pathways
  - Nutrient Capture and Catabolic Pathways
  - Cell Stress
  - Metabolic Signaling
  - Transcriptional Regulation
- Understand mechanisms of metabolic adaptation, metabolic switching and metabolic alterations
- Study changes in mitochondrial respiration and glycolysis
- Quantify the presence and relative abundance of 14 different immune cell types for immunometabolism studies
- Advance efforts towards novel therapeutic target

Feature	Specifications
Number of Targets	768 (Human), 768 (Mouse), including internal reference genes
Standard Input Material (No amplification required)	25 ng–300 ng
Sample Input - Low Input	As little as 1 ng with nCounter RNA Low Input Kit (sold seperately)
Sample Type(s)	Cultured cells/cell lysates, sorted cells, FFPE-derived RNA, total RNA, fragmented RNA, PBMCs, and whole blood/plasma
Customizable	Add up to 55 unique genes with Panel Plus
Time to Results	Approximately 24 hours
Data Analysis	nSolver™ Analysis Software (RUO) and the ROSALIND® Platform

Key Applications with the nCounter Metabolic Pathways Panel

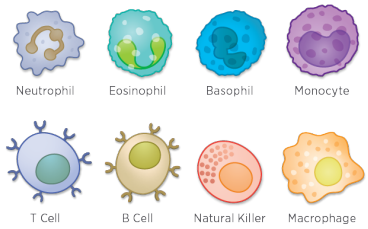
Cancer Metabolism



How does cancer impact normal metabolic function?

- Study key pathways involved in reprogrammed metabolism
- Identify metabolic changes that can be exploited for therapeutic development

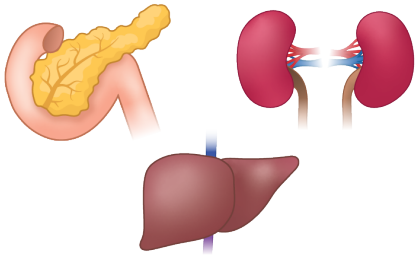
Immunometabolism



How does the immune response change as a result of alterations in metabolism

- Profile immune cell types
- Link pathway activity to immune phenotype

Metabolic Disease



How is metabolism altered in disease?

- Understanding disease pathogenesis
- Identify potential therapeutic targets

Theme	Description	Pathways	Number of Human Genes	Number of Mouse Genes
Biosynthesis and Anabolic Pathways	The processes involved in the production of complex macromolecules by enzyme-catalyzed biosynthetic pathways. The products of these pathways are required for nearly all cellular functions, including proliferation.	Amino Acid Synthesis, Arginine Metabolism, Cell Cycle, Fatty Acid Synthesis, Glutamine Metabolism, Glycolysis, IDH1/2 Activity, Mitochondrial Respiration, Nucleotide Synthesis, Pentose Phosphate Pathway, Tryptophan/ Kynurenine Metabolism, Vitamin and Cofactor Metabolism	354	348
Cell Stress	Cells are impacted by the availability of nutrients and presence of toxic compounds. Adaptive responses to the stress are required for tumorigenesis, metastasis, and immune responses.	DNA Damage Repair, Hypoxia, KEAP1/NRF2 Pathway, Reactive Oxygen Response	82	82
Nutrient Capture and Catabolic Pathways	The processes involved in the breakdown of macromolecules, scavenging of cellular materials, or import of nutrients in order to stimulate ATP production or fuel anabolic pathways.	Amino Acid Transporters, Autophagy, Endocytosis, Fatty Acid Oxidation, Glucose Transport, Lysosomal Degradation, Nucleotide Salvage	161	159
Metabolic Signaling	The pathways that are commonly disrupted in cancer cells or altered in immune cells that impact metabolic function. In the context of cancer, mutations allow these regulated signaling pathways to allow for metabolic change enabling tumorigenesis.	AMPK, mTOR, MAPK, Myc, NF-kB, p53 Pathway, PI3K, TCR and Costimulatory Signaling, TLR Signaling	237	235
Transcriptional Regulation	Processes involved in the alteration of epigenetic and transcriptional activity of the cell that enables sustained metabolic reprogramming. This reprogramming allows for tumorigenesis and underlies stable changes in immune cell phenotype.	Epigenetic Regulation, Transcriptional Regulation	77	69

Metabolic Pathways Panel Functional Annotations

Functional annotations for different pathways and processes were assigned to the genes in the Metabolic Pathways Panel. The pathways and processes that are included in this panel provide a comprehensive view of cell metabolism.

Annotation	Number of Human Genes	Number of Mouse Genes
AMPK	48	48
Amino Acid Synthesis	93	89
Amino Acid Transporters	9	9
Arginine Metabolism	16	16
Autophagy	54	54
Cell Cycle	65	65
DNA Damage Repair	31	31
Endocytosis	39	41
Epigenetic Regulation	26	26
Fatty Acid Oxidation	24	21
Fatty Acid Synthesis	11	11
Glucose Transport	6	5
Glutamine Metabolism	31	30
Glycolysis	40	37
Hypoxia	15	15
IDH1/2 Activity	15	15
KEAP1/NRF2 Pathway	5	5
Lysosomal Degradation	16	16
mTOR	57	57

Annotation	Number of Human Genes	Number of Mouse Genes
MAPK	52	52
Mitochondrial Respiration	73	72
Myc	20	20
NF-kB	27	27
Nucleotide Salvage	23	23
Nucleotide Synthesis	48	48
p53 Pathway	38	38
PI3K	82	82
Pentose Phosphate Pathway	19	19
Reactive Oxygen Response	37	37
TCR and Costimulatory Signaling	54	54
TLR Signaling	33	31
Transcriptional Regulation	59	51
Tryptophan/Kynurenine Metabolism	32	32
Vitamin and Cofactor Metabolism	28	28
Internal Reference	20	20

Immune Cell Profiling Feature

Genes included in the Human Metabolic Pathways Panel provide unique cell profiling data to measure the relative abundance of 14 different human immune cell types[1]. The table below summarizes each cell type represented by gene content in the panel, as qualified through biostatistical approaches and selected literature in the field of immunology.

Cell Type	Associated Human Genes
B cells	9
CD45	1
CD8 T cells	2
Cytotoxic Cells	10
Dendritic Cells	3
Exhausted CD8	4
Macrophages	4

Cell Type	Associated Human Genes
Mast cells	4
Neutrophils	7
NK CD56dim cells	4
NK Cells	2
T cells	6
Th1 Cells	1
Treg	1

nSolver™ Analysis Software

NanoString offers advanced software tools that address the continuous demands of data analysis and the need to get simple answers to specific biological questions easily. Genes included in the Metabolic Pathways panel are organized and linked to various advanced analysis modules to allow for efficient analysis of the 34 pathways involved in cellular metabolism.

ROSALIND® Platform

ROSALIND is a cloud-based platform that enables scientists to analyze and interpret differential gene expression data without the need for bioinformatics or programming skills. ROSALIND makes analysis of nCounter data easy, with guided modules for:

- Normalization
- Quality Control
- Individual Pathway Analysis
- Differential Expression
- Gene Set Analysis

nCounter customers can access ROSALIND at: rosalind.bio/nanostring

Ordering Information

Gene Expression Panels arrive ready-to-use and generally ship within 24 hours following purchase.

Product	Product Description	Quantity	Catalog Number
nCounter Human Metabolic Pathways Panel	Includes 768 genes; 20 internal reference genes for data normalization	12 Reactions	XT-CSO-HMP1-12
nCounter Mouse Metabolic Pathways Panel	Includes 768 genes; 20 internal reference genes for data normalization	12 Reactions	XT-CSO-MMP1-12
nCounter Analysis System Master Kit Reagents and Cartridges	Reagents, cartridges, and consumables necessary for sample processing on the nCounter Analysis System	12 Reactions	NAA-AKIT-012
nCounter SPRINT Cartridge 1 Cartridge, 12 lanes	Sample Cartridge for nCounter SPRINT System	12 Reactions	SPRINT-CAR-1.0
nCounter SPRINT Reagent Pack	nCounter SPRINT Reagent Pack containing Reagents A, B, C, and Hybridization Buffer	192 Reactions	SPRINT-REAG-KIT

Selected Panel References

1. Danaher, P et al. Gene Expression Markers of Tumor Infiltrating Leukocytes. J Immunother Cancer. 2017;21(5):18.
2. Peng, X et al. Molecular Characterization and Clinical Relevance of Metabolic Expression Subtypes in Human Cancers. Cell Reports. 2018;23(1):255-69.
3. DeBerardinis RJ, Chandel, NS. Fundamentals of Cancer Metabolism. Science Advances. 2016;2(5).
4. O'Neill LA et al. A Guide to Immunometabolism for Immunologists. Nature Reviews Immunology. 2016;16(9):553-65.
5. Stine, ZE et al. MYC, Metabolism, and Cancer. Cancer Discovery. 2015;10:1024-39.
6. Renner, K et al. Metabolic Hallmarks of Tumor and Immune Cells in the Tumor Microenvironment. Frontiers in Immunology. 2017;8:248.
7. Andrejava, G and Rathmell, JC. Similarities and Distinctions of Cancer and Immune Metabolism in Inflammation and Tumors. Cell Metabolism. 2017;26(1):49-70.

For more information, please visit nanostring.com/metabolic-pathways

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