Axela Biosciences

TRUSTED PARTNER ACCELERATING DRUG DISCOVERY & TRANSLATIONAL DEVELOPMENT

Reverse Phase Protein Array (RPPA)

High-Throughput Functional Proteomics for Discovery and Precision Medicine

- **High-throughput**
- **Functional insights**
- **Ultra-sensitive**
- **Minimal sample (µg)**

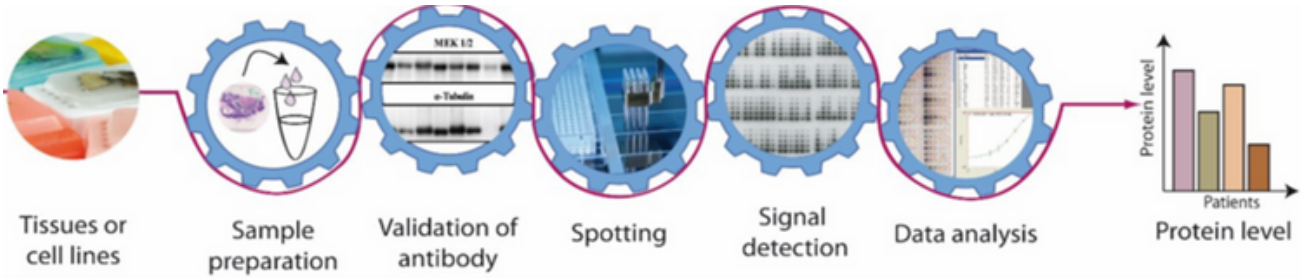
Integration of RPPA & nCounter Technology

Transcriptomics + Proteomics from One Sample: A Complete View of Biology

- **Profile ~600 proteins and ~770 transcripts simultaneously**
- **Uncover MoA and pathway activation from one sample**

WHAT IS RPPA & WHY IT MATTERS

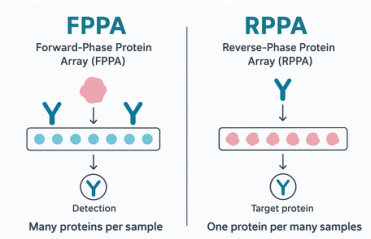
RPPA is a high-throughput, antibody-based proteomic array that quantitatively measures protein expression and post-translational modifications across thousands of samples. Unlike genomic or transcriptomic methods, RPPA captures functional protein activity, providing insights into signaling pathways, disease mechanisms, and treatment responses.



High sensitivity, minimal sample input, scalable, and actionable results

- **High multiplexity**: Enables the detection and quantification of hundreds of distinct proteins, including their expression levels and modification status, using only **40 µg of cell lysates (currently covering ~ 600 proteins from various signaling pathways).**
- **Cost-effective**: **$1,000 to 3,000 per sample**, depending on the quantity of samples and sample types.
- **High sensitivity**: Exhibits greater sensitivity than ELISA while requiring minimal sample consumption.
- **Truly quantitative:** RPPA enables precise measurement of protein expression and post-translational modifications, calibrated with recombinant proteins and phospho-peptide standards.
- **Simplified: No direct labeling or labor-intensive depletion or fractionation** of sample lysates is required.
- **Versatile sample types**: Various types of samples can be investigated, including those from cell cultures, live animals and patients (such as fine needle aspirates, laser capture microdissection, FFPE samples), sorted cells, as well as serum, plasma, saliva, urine, etc.

FPPA (Forward Phase Protein Array) vs. RPPA (Reverse Phase Protein Array)

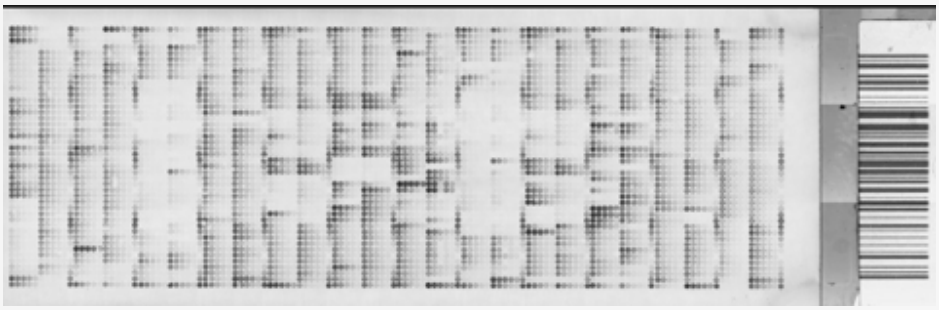


FEATURES	FPPA	RPPA	ADVANTAGES OF RPPA
Array Design	Antibodies are spotted on the array	Samples (protein lysates) are spotted on the array	Efficient use of precious samples
Sample Requirement	Requires larger amounts of sample	Very small sample needed (µg level)	Enables analysis of minimal samples
Throughput (Samples)	Dozens to ~100 samples per array	Dozens to thousands of samples per batch	Flexible for small to large cohort studies
Throughput (Proteins)	50–200 proteins per array (depends on antibodies)	Up to ~600 proteins per array (with validated antibodies)	Broad pathway coverage and MOA exploration
Sensitivity	Moderate	Very high, detects low-abundance proteins & PTMs	Detects subtle signaling changes
Reproducibility	Variable, antibody quality dependent	High reproducibility across batches	Robust for biomarker discovery & validation
Applications	General protein profiling	Multi-pathway analysis, biomarker discovery, MOA	Powerful in translational & clinical research

APPLICATIONS

RPPA empowers biopharma, clinicians, and researchers to drive discovery and precision medicine

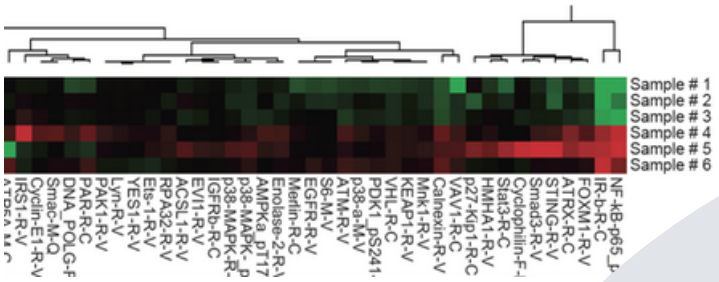
AREA	APPLICATIONS
Systems Biology	Network mapping, multi-omics integration
Drug Development	Target validation, biomarker discovery, MoA studies
Translational Development	Companion diagnostics, patient stratification, therapy monitoring
Various Diseases, e.g. Cancer	Pathway mapping, tumor subtyping, resistance profiling



Validated Targets

~ 600 targets, including ~100 phosphoproteins;
Please email info@axelabio.com for the target list.

Case Study: Investigating the MoA of Kinase Inhibitor A in Lymphoma



A single RPPA study revealed four novel pathways (MOAs) for Kinase Inhibitor A, showcasing the platform's exceptional sensitivity and reproducibility.

Antibody Name	14-3-3-beta	14-3-3-epsilon	Cadherin-6	Calnexin	FOXM1	FOXO3	Glucocorticoid-Receptor	HER3_pY1289	IR-b	IRF-1	IRF-3
Gene Name	YWHAB	YWHAE	CDH6	CANX	FOXM1	FOXO3	NR3C1	ERBB3	INSR	IRF1	IRF3
Antibody Name in Heatmap	14-3-3-beta-R-V	14-3-3-epsilon-M-C	Cadherin-6-R-C	Calnexin-R-V	FOXM1-R-V	FOXO3-R-V	Glucocorticoid-Receptor-R-V	HER3_pY1289-R-C	IR-b-R-C	IRF-1-R-V	IRF-3-R-V
Antigen Id	AGID00100	AGID00111	AGID02261	AGID00429	AGID02152	AGID00378	AGID00548	AGID00080	AGID00272	AGID00223	AGID00519
Slide no.	1	2	80	81	193	194	208	225	247	248	249
Sample description	14-3-3-beta-R-V	14-3-3-epsilon-M-C	Cadherin-6-R-C	Calnexin-R-V	FOXM1-R-V	FOXO3-R-V	Glucocorticoid-Receptor-R-V	HER3_pY1289-R-C	IR-b-R-C	IRF-1-R-V	IRF-3-R-V
Untreated, replicate 1	0.976	1.040	1.011	0.716	2.605	1.045	3.642	1.020	1.768	1.145	1.509
Untreated, replicate 2	0.977	1.045	1.017	0.725	2.963	1.152	3.527	1.031	1.732	1.169	1.451
Untreated, replicate 3	0.945	0.945	0.982	0.733	2.760	1.145	3.788	1.050	2.892	1.148	1.426
Treatment, replicate 1	0.904	0.909	1.005	1.183	4.033	1.071	0.972	1.042	6.630	1.058	1.529
Treatment, replicate 2	0.896	0.911	1.026	1.094	5.072	1.040	0.908	1.014	7.014	1.061	1.454
Treatment, replicate 3	1.004	0.957	1.047	1.150	3.482	1.031	0.914	1.032	7.379	1.047	1.552
Sample # 1-3, AVE	0.966	1.010	1.004	0.725	2.776	1.114	3.652	1.034	2.131	1.154	1.462
Sample # 4-6, AVE	0.935	0.925	1.026	1.142	4.195	1.047	0.931	1.030	7.007	1.055	1.512
TX1/Ctrl, Fold Change	0.97	0.92	1.02	1.58	1.51	0.94	0.25	1.00	3.29	0.91	1.03

INTEGRATED APPROACHES COMBINING PROTEOMICS & TRANSCRIPTOMICS

RPPA + nCounter: Comprehensive MOA Insights from One Sample

Key Benefits:

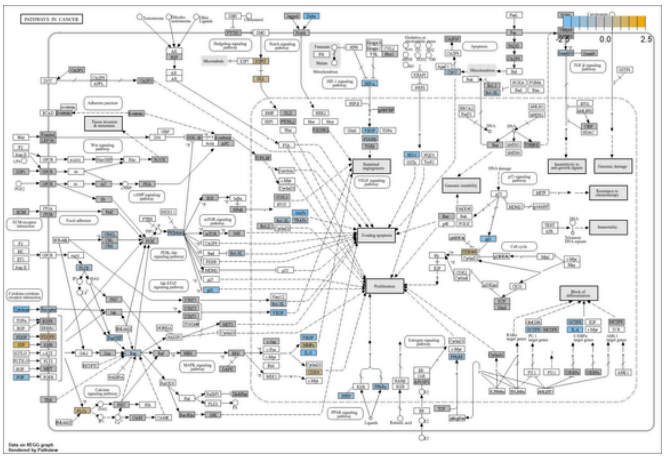
Dual-layer profiling: transcriptomics & proteomics from one sample

Mechanistic insight: link protein activity to gene expression

High throughput: hundreds of samples in parallel

Efficient MOA assessment: minimizes sample use and experimental variability

Layer	Technology	Targets per Sample
Transcriptome	nCounter	~770 genes
Proteome	RPPA	~600 proteins



Case Study: nCounter Analysis of Cancer Pathways: Upregulated (orange) and Downregulated (blue) mRNA Levels in Response to Drug B

Exact fold-change values are provided in the accompanying Excel file

FAQ for RPPA

Q: How many protein targets can be quantitated in one RPPA array?
A: Our current array can quantitate ~600 well-validated targets.

Q: What is the turnaround time for performing one batch of RPPA?
A: Our normal turnaround time is about 2-3 months for each batch.

Q: How many samples can be run in a batch?
A: We run the array in batches. Up to 1000 samples will be run in each array slide.

Q: What is the cost per sample?
A: Pricing ranges from $1,000 to $3,000 per sample, depending on sample type and volume. A one-time fee covers data analysis and reporting.

Q. What assays are available to confirm the RPPA data?
A. We offer Simple Western (automatic western blot), ELISA/AlphaLISA, Pathway-focused Antibody Arrays and flow cytometric analysis to confirm target of interest if needed.

We can be reached at

scientist.com
ScienceExchange

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