Acute Phase Response
Cancer
Cardiovascular Disease
Diabetes
Cytokines, Chemokines, Growth Factors
Immunology/Inflammation
Immunoglobulin Isotyping
Cell Signaling
Toxicology

Bio-Plex Pro™ Cell Signaling Assays

MAGNETIC SEPARATION ENABLED

Akt, ATF-2, β-Actin, BAD, Btk, c-Abl, c-Jun, CREB, EGFR, Erk1/2, GSK-3α/β, HER-2, HSP27, GAPDH, IGF-1R, IκB-α, IR-β, IRS-1, JNK, Lyn, MEK1, mTOR, NF-κB p65, p38 MAPK, p53, P70 S6 Kinase, p90 RSK, PDGFR-α, PDGFR-β, PI3K p85, PTEN, S6 Ribosomal Protein, Smad2, Src, Stat1, Stat3, Syk, VEGFR-2, ZAP-70

- Magnetic or vacuum assay separation
- Convenient kit format
- Flexible ordering options
- Minimal sample volume
- Antibodies from CST

Cell Signaling Assays with Exceptional Sensitivity

The Bio-Plex Pro cell signaling assays are magnetic bead–based immunoassays designed to meet the sensitivity needs of the most discerning scientists. The multiplex format enables robust, reproducible, and simultaneous measurement of proteins involved in key intracellular signaling pathways. Design your own assay choosing from a broad selection of phosphoprotein and total protein targets to investigate pathways associated with:

- Cancer
- Inflammation
- Diabetes
- Neurological disorders
- Cardiovascular disorders
- Drug mechanism of action
- Toxicology

These assays incorporate several features to enhance both quality and ease of use:

- Panel includes house keeping proteins GAPDH and β-Actin
- Assay quick guide to get you started right away
- Assay protocol optimized for exceptional sensitivity and broad dynamic range
- Flexible ordering options — order singleplex sets or visit **www.bio-rad.com/assaybuilder** to configure a custom premixed kit

Benefits of Magnetic Bead–Based Assays

Magnetic bead–based assays enable automation of wash steps with a Bio-Plex Pro series or similar wash station. This innovation simplifies assay processing and eliminates the need for a vacuum manifold. After adopting the magnetic assay workflow, many users experience improved assay precision, in particular with viscous samples.

Rigorous Assay Validation

All Bio-Plex Pro cell signaling assays undergo a rigorous evaluation that includes the following parameters:

- Specificity and cross-reactivity
- Inter- and intra-assay precision
- Sensitivity (limit of detection, LOD)
- Dynamic range
- Performance characteristics in real samples

Assay Performance Definitions

Precision — the coefficient of variation (%CV) at concentrations within the assay working range

Sensitivity (LOD) — the amount of cell lysate protein for which the fluorescence intensity signal of the specific analyte is two standard deviations above the background signal

BIO-RAD

Available Assays

Table 1 shows a list of available Bio-Plex Pro cell signaling assays now offered for the semiquantitative detection of total target proteins and phosphoproteins in cell and tissue lysates.

Table 1. Available assays.

Total Target Assays	Bead Regions	Total Target Assays	Bead Regions
Akt*	75	JNK	34
Btk *	39	MEK1	27
c-Jun	56	mTOR*	46
CREB*	19	p38 MAPK	36
Erk1/2	38	P70 S6 Kinase	55
GSK-3β	18	PTEN*	22
HER-2	30	Smad2	14
IGF-1R	43	Src*	42
IκB-α	67	ZAP-70*	64

Housekeeping Proteins	Bead Regions	Housekeeping Proteins	Bead Regions
Human GAPDH	21	β-Actin	47

Phosphoprotein Assays	Bead Regions	Phosphoprotein Assays	Bead Regions
Akt (Ser^{473})	75	mTOR (Ser^{2448})*	46
Akt (Thr^{308})	75	NF-κB p65 (Ser^{536})	37
ATF-2 (Thr^{71})*	20	p38 MAPK (Thr^{180}/Tyr^{182})	36
BAD (Ser^{136})*	26	p53 (Ser^{15})*	53
Btk (Tyr^{223})*	39	p70 S6 Kinase (Thr^{389})	55
c-Abl (Tyr^{245})*	45	p70 S6 Kinase (Thr^{421}/Ser^{424})	55
c-Jun (Ser^{63})	56	p90 RSK (Ser^{380})*	35
CREB (Ser^{133})*	19	PDGFR-α (Tyr^{754})	28
EGFR (Tyr^{1068})	44	PDGFR-β (Tyr^{751})	57
EGFR (Tyr^{1173})	44	PI3K p85 (Tyr^{458})*	54
Erk1/2 (Thr^{202}/Tyr^{204}, Thr^{185}/Tyr^{187})	38	PTEN (Ser^{380})*	22
GSK-3α/β (Ser^{21}/Ser^{9})	18	S6 Ribosomal Protein* (Ser^{235}/Ser^{236})	74
HER-2 (Tyr^{1248})	30	Smad2 (Ser^{465}/Ser^{467})	14
HSP27 (Ser^{78})*	51	Src (Tyr^{416})*	42
IGF-1R (Tyr^{1131})	43	Stat1 (Tyr^{701})	61
IR-β (Tyr^{1146})*	43	Stat3 (Ser^{727})	52
IRS-1 (Ser^{636}/Ser^{639})*	76	Stat3 (Tyr^{705})	52
IκB-α (Ser^{32}/Ser^{36})	67	Syk (Tyr^{352})*	65
JNK (Thr^{183}/Tyr^{185})	34	VEGFR-2 (Tyr^{1175})	29
Lyn (Tyr^{507})*	33	ZAP-70 (Tyr^{319})*	64
MEK1 (Ser^{217}/Ser^{221})	27		

* Latest additions to the Bio-Plex® Pro cell signaling panel.

Assay Performance Characteristics

The Bio-Plex Pro cell signaling assays were evaluated for sensitivity (P/B ratio and LOD), intra- and inter-assay %CV, and species specificity (Table 2).

Table 2. Assay performance characteristics.

Total Target Assays	Intra-assay %CV	Inter-assay %CV	Sensitivity (LOD), µg/well	Reaction Species*
β-Actin	3.4	13.0	0.15	H, M, R, NHP
Akt	3.8	8.4	0.15	H, M, R
Btk	7.6	10.0	0.15	H, M
c-Jun	4.0	4.7	0.15	H
CREB	0.7	10.4	0.15	H, M, R, NHP
Erk1/2	3.4	9.2	0.15	H, M
GAPDH	1.9	4.8	0.15	H
GSK-3β	2.9	4.1	0.8	H, M, R
HER-2	5.6	4.2	0.04	H, M, R
IGF-1R	4.7	4.4	0.15	H
IκB-α	1.9	3.1	0.15	H
JNK	1.9	4.5	0.3	H
MEK 1	1.8	3.9	0.15	H, M, R
mTOR	4.1	3.3	0.15	H, M, R, NHP
p38 MAPK	4.3	9.6	0.3	H, M, R
p70 S6 Kinase	2.0	3.6	0.15	H, M, R
PTEN	5.0	3.2	0.15	H, M, R
Smad2	3.9	16.9	0.15	H
Src	11.3	5.4	0.15	H, M, R
ZAP-70	3.1	11.2	0.15	H

* H, human; M, mouse; R, rat; NHP, nonhuman primate.

Table 2. Assay performance characteristics (cont).

Phosphoprotein Assays	P/B Ratio*	Intra-assay %CV	Inter-assay %CV	Sensitivity (LOD), µg/well	Reaction Species**
Akt (Ser^{473})	65	2.9	4.5	0.15	H, M, R
Akt (Thr^{308})	34	3.3	6.1	0.15	H, M, R
ATF-2 (Thr^{71})	68	4.0	6.3	0.15	H, M, R, NHP
BAD (Ser^{136})	12	9.7	9.4	0.15	H, M, R, NHP
Btk (Tyr^{223})	99	3.2	1.3	0.15	H
c-Abl (Tyr^{245})	210	1.3	10.0	0.15	H, M
c-Jun (Ser^{63})	295	5.8	8.4	0.15	H, M, R
CREB (Ser^{133})	50	4.0	8.9	0.15	H, M, R
EGFR (Tyr^{1068})	98	14.1	17.7	0.15	H
EGFR (Tyr^{1173})	30	3.9	6.3	0.15	H
Erk1/2 (Thr^{202}/Tyr^{204}, Thr^{185}/Tyr^{187})	48	4.3	4.9	0.15	H, M
GSK-3α/β (Ser^{21}/Ser^{9})	19	2.9	5.4	0.15	H, M, R
HER-2 (Tyr^{1248})	49	10.1	9.1	0.15	H, M, R
HSP27 (Ser^{78})	58	2.7	4.4	0.15	H, NHP
IGF-1R (Tyr^{1131})	68	4.8	6.5	0.15	H
IR-β (Tyr^{1146})	74	1.5	3.2	0.15	H, M, R
IRS-1 (Ser^{636}/Ser^{639})	52	5.5	9.3	0.3	H, M, R
IκB-α (Ser^{32}/Ser^{36})	103	6.8	8.9	0.15	H
JNK (Thr^{183}/Tyr^{185})	73	4.7	4.1	0.15	H
Lyn (Tyr^{507})	125	4.2	4.4	0.15	H, M, R
MEK1 (Ser^{217}/Ser^{221})	44	1.9	5.5	0.15	H, M, R
mTOR (Ser^{2448})	40	3.3	9.2	0.3	H, M, R, NHP
NF-κB p65 (Ser^{536})	20	10.7	13.5	0.3	H, M, R
p38 MAPK (Thr^{180}/Tyr^{182})	62	3.1	6.4	0.15	H, M, R
p53 (Ser^{15})	59	8.0	12.2	0.15	H
p70 S6 Kinase (Thr^{389})	196	8.4	10.3	0.15	H, M, R
p70 S6 Kinase (Thr^{421}/Ser^{424})	11	2.9	20.1	0.15	H, M, R
p90 RSK (Ser^{380})	80	1.7	4.2	0.15	H, M, R, NHP
PDGFR-α (Tyr^{754})	40	5.0	6.2	0.04	H, M
PDGFR-β (Tyr^{751})	142	8.1	10.3	0.04	H, M, R
PI3K p85 (Tyr^{458})	42	9.3	11.4	1.6	H, M, R
PTEN (Ser^{380})	39	9.0	6.3	0.3	H, M, R
S6 Ribosomal Protein (Ser^{235}/Ser^{236})	26	4.3	5.2	0.15	H, M, R
Smad2 (Ser^{465}/Ser^{467})	14	4.4	8.8	0.3	H, M, R
Src (Tyr^{416})	178	5.7	9.1	0.15	H, M, R
Stat1 (Tyr^{701})	18	3.6	7.8	0.15	H
Stat3 (Ser^{727})	89	7.0	10.1	0.15	H, M, R
Stat3 (Tyr^{705})	18	2.5	6.8	0.15	H, M, R
Syk (Tyr^{352})	232	1.4	2.0	0.15	H, M, R
VEGFR-2 (Tyr^{1175})	614	8.3	11.7	0.15	H, M
ZAP-70 (Tyr^{319})	138	2.6	5.1	0.15	H, M

* Signal ratio (positive/background) of stimulated cell lysate/background cell lysate.
** H, human; M, mouse; R, rat; NHP, nonhuman primate.

Correlation and Sensitivity Comparison

Western blotting is a traditional technique for detecting phosphorylated proteins within a variety of cell culture and tissue sample lysates. Comparison of the Bio-Plex Pro cell signaling assays to that of western blotting shows a correlation in the expression pattern of phosphorylated proteins (Figure 1) as well as a greater sensitivity in the detection of these phosphorylated proteins (Figure 2).

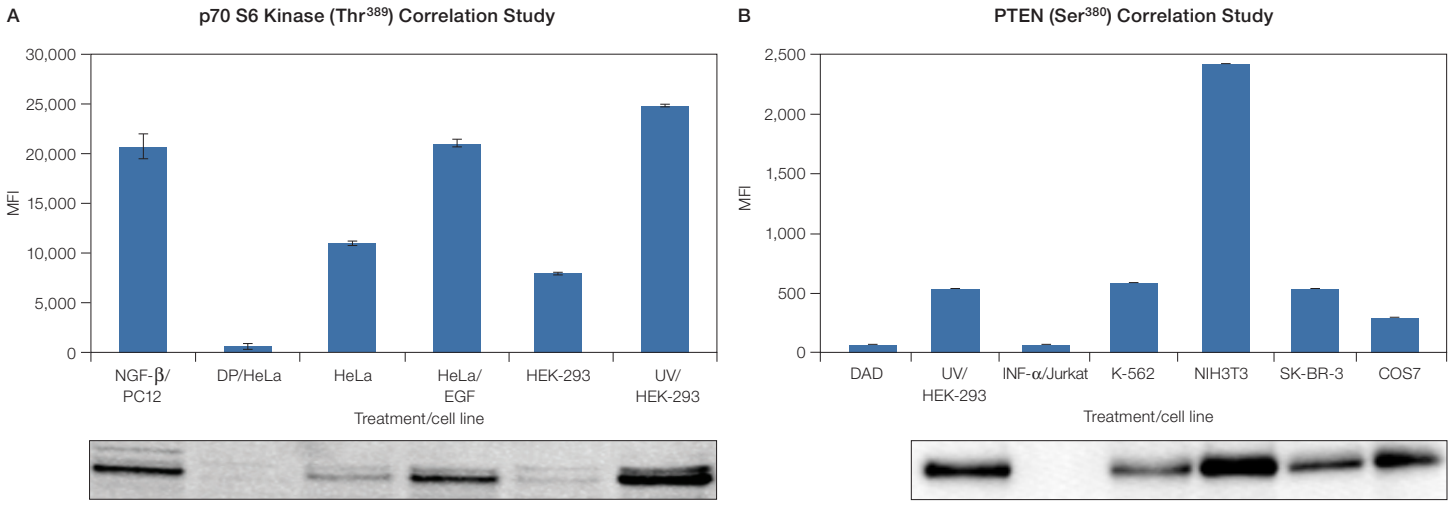

Fig. 1. Performance of Bio-Plex Pro cell signaling assays compared to western blotting. Measurements of p70 S6 Kinase (Thr^{389}) (**A**) and PTEN (Ser^{380}) (**B**) in a variety of cell lines. DAD indicates detection antibody diluent alone. In panel **A**, DP/HeLa indicates phosphatase-treated HeLa cells. This condition serves as a negative control.

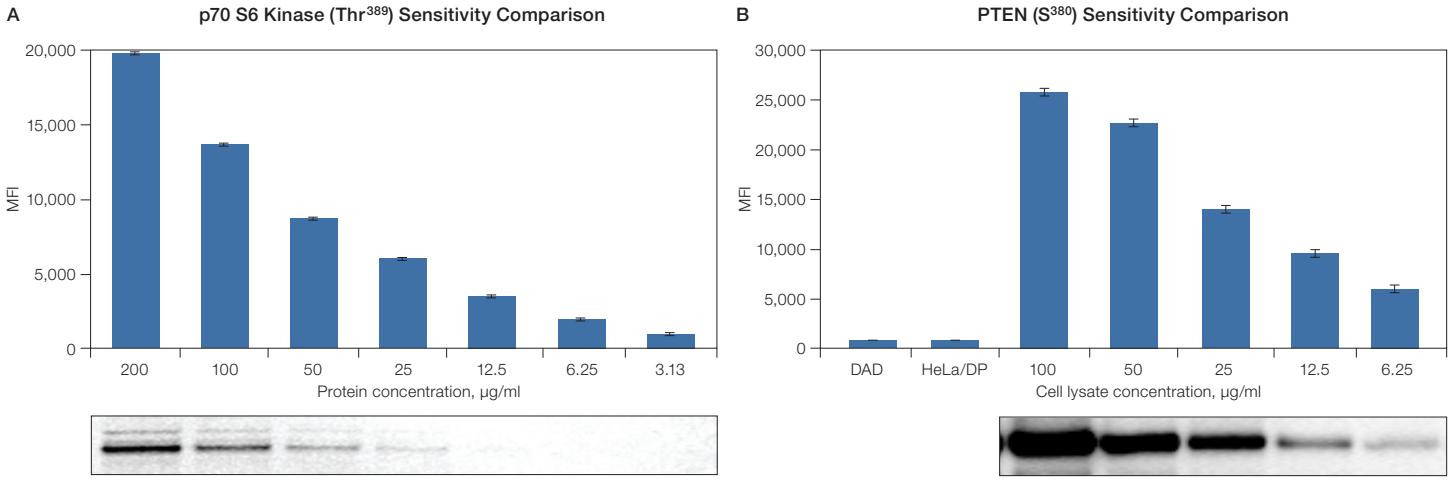

Fig. 2. Sensitivity of Bio-Plex Pro cell signaling assays. Phosphorylated p70 S6 Kinase (Thr^{389}) levels were measured in PC12 cells stimulated with NGF-β (**A**). The Bio-Plex assay was able to detect the analyte in as little as 3 µg/ml (150 ng/well) of total lysate protein. Phosphorylated PTEN (Ser^{380}) levels were measured in HeLa cells stimulated with TNF-α (**B**). The Bio-Plex assay was able to detect the analyte in as little as 6 µg/ml (300 ng/well) of total lysate protein.

Flexible Ordering Options

Premixed Panels

One kit with everything you need to run an experiment.

Individual Components

Select compatible singleplex sets and lysates. Use Table 3 to check for assay cross-reactivity. Refer to Table 4 to select the appropriate lysate controls for your assays. Complete your order with the Bio-Plex Pro cell signaling reagent kit for a mix-it-yourself multiplex solution.

x-Plex™ Custom Assay Service

A great choice for premium custom-mixed assays. Simply select your analytes of interest using the online Bio-Plex assay builder at **www.bio-rad.com/assaybuilder**. Assays are mixed for you at Bio-Rad and delivered as an all-in-one kit.

Ordering Information

Catalog #	Description
Premixed, All-in-One Kits	
LQ0-0000S6KL81S	**Bio-Plex Pro Cell Signaling MAPK Panel**, 9-plex, 1 x 96 includes coupled magnetic beads and detection antibodies, cell lysis buffer, cell lysis factor QG, cell wash buffer, bead resuspension buffer, detection antibody diluent, wash buffer, streptavidin-PE, 96-well flat bottom plate, sealing tape, and instructions for detecting ATF-2 (Thr^{71}), ERK1/2 (Thr^{202}/Tyr^{204}, Thr^{185}/Tyr^{187}), HSP27 (Ser^{78}), JNK (Thr^{183}/Tyr^{185}), MEK1 (Ser^{217}/Ser^{221}), p38 MAPK (Thr^{180}/Tyr^{182}), p53 (Ser^{15}), p90 RSK (Ser^{380}), and Stat3 (Ser^{727})
LQ0-0006JK0K0RR	**Bio-Plex Pro Cell Signaling Akt Panel**, 8-plex, 1 x 96 includes coupled magnetic beads and detection antibodies, cell lysis buffer, cell lysis factor QG, cell wash buffer, bead resuspension buffer, detection antibody diluent, wash buffer, streptavidin-PE, 96-well flat bottom plate, sealing tape, and instructions for detecting Akt (Ser^{473}), BAD (Ser^{136}), GSK-3α/β (Ser^{21}/Ser^{9}), IRS-1 (Ser^{636}/Ser^{639}), mTOR (Ser^{2448}), p70 S6 Kinase (Thr^{389}), PTEN (Ser^{380}), and S6 Ribosomal Protein (Ser^{235}/Ser^{236})
Bio-Plex Pro Cell Signaling Reagents	
171-304006M	**Bio-Plex Pro Cell Signaling Reagent Kit**, 1 x 96-well, includes cell lysis buffer, cell lysis factor QG, cell wash buffer, bead resuspension buffer, detection antibody diluent, wash buffer, streptavidin-PE, 96-well flat bottom plate, sealing tape, and instructions
171-304515	**Bio-Plex Pro Cell Signaling Wash Buffer**, 330 ml, for 1 x 96-well assay, for use with Bio-Plex Pro cell signaling assays only, compatible with both magnetic and vacuum separation methods
Wash Stations	
300-34376	**Bio-Plex Pro Wash Station**, includes magnetic plate carrier, waste bottle, 2 buffer bottles
Accessories	
171-020100	**Bio-Plex Handheld Magnetic Washer**, includes magnetic washer and adjustment hex tools for use in manual wash steps for all Bio-Plex magnetic assays
171-304502	**Filter Plate**, pkg of 1, 96-well plate, with clear plastic lid and tray, for Bio-Plex assays using the vacuum wash method, sealing tape not included
Software	
171-001510	**Bio-Plex Data Pro™ Software with Bio-Plex Manager™ Software**, Bio-Plex Data Pro software (5 seats), for multi-experiment analysis and advanced data visualization, and Bio-Plex Manager software (5 seats), for instrument data evaluation and optimization. CDs and security HASP key included
171-001513	**Bio-Plex Data Pro Software**, (5 seats), for multi-experiment analysis and advanced data visualization
171-STND01	**Bio-Plex Manager Software**, includes 1 user desktop license, to analyze Bio-Plex data and generate protocols, does not operate the instrument

Catalog #	Description
Phosphoprotein Singleplex Sets*	1 x 96-well, includes coupled magnetic beads and detection antibodies for detecting phosphorylated protein, requires cell signaling reagent kit and optional lysate controls
171-V50001M	Akt (Ser^{473})**
171-V50002M	Akt (Thr^{308})**
171-V50024M	ATF-2 (Thr^{71})**
171-V50025M	BAD (Ser^{136})
171-V50026M	Btk (Tyr^{223})**
171-V50027M	c-Abl (Tyr^{245})**
171-V50003M	c-Jun (Ser^{63})**
171-V50028M	CREB (Ser^{133})
171-V50004M	EGFR (Tyr^{1068})**
171-V50005M	EGFR (Tyr^{1173})**
171-V50006M	ERK1/2 (Thr^{202}/Tyr^{204},Thr^{185}/Tyr^{187})**
171-V50007M	GSK-3α/β (Ser^{21}/Ser^{9})
171-V50008M	HER-2 (Tyr^{1248})**
171-V50029M	HSP27 (Ser^{78})
171-V50009M	IGF-1R (Tyr^{1131})**
171-V50031M	IR-β (Tyr^{1146})**
171-V50030M	IRS-1 (Ser^{636}/Ser^{639})**
171-V50010M	IκB-α (Ser^{32}/Ser^{36})**
171-V50011M	JNK (Thr^{183}/Tyr^{185})
171-V50032M	Lyn (Tyr^{507})**
171-V50012M	MEK1 (Ser^{217}/Ser^{221})**
171-V50033M	mTOR (Ser^{2448})
171-V50013M	NF-κB p65 (Ser^{536})**
171-V50014M	p38 MAPK (Thr^{180}/Tyr^{182})**
171-V50034M	p53 (Ser^{15})
171-V50016M	p70 S6 Kinase (Thr^{389})**
171-V50015M	p70 S6 Kinase (Thr^{421}/Ser^{424})**
171-V50035M	p90 RSK (Ser^{380})
171-V50017M	PDGFR-α (Tyr^{754})**
171-V50018M	PDGFR-β (Tyr^{751})**
171-V50036M	PI3K p85 (Tyr^{458})**
171-V50037M	PTEN (Ser^{380})
171-V50038M	S6 Ribosomal Protein (Ser^{235}/Ser^{236})
171-V50019M	Smad2 (Ser^{465}/Ser^{467})
171-V50039M	Src (Tyr^{416})**
171-V50020M	Stat1 (Tyr^{701})**
171-V50021M	Stat3 (Ser^{727})**
171-V50022M	Stat3 (Tyr^{705})**
171-V50040M	Syk (Tyr^{352})**
171-V50023M	VEGFR-2 (Tyr^{1175})
171-V50041M	ZAP-70 (Tyr^{319})**
Total Target Singleplex Sets*	
171-V60001M	Akt**
171-V60012M	Btk
171-V60002M	c-Jun
171-V60013M	CREB
171-V60003M	Erk1/2
171-V60004M	GSK-3β
171-V60005M	Her2
171-V60014M	IGF-1R
171-V60006M	IκB-α
171-V60007M	JNK
171-V60008M	MEK1
171-V60015M	mTOR
171-V60009M	p38 MAPK**
171-V60010M	p70 S6 Kinase
171-V60016M	PTEN
171-V60011M	Smad2
171-V60017M	Src
171-V60018M	ZAP-70
Housekeeping Protein Sets*	
171-V60020M	β-Actin**
171-V60019M	Human GAPDH

* Refer to Table 4 to select lysate controls for multiplexing.

** Analytes with known cross-reactivity. Refer to Table 3 when selecting analytes for multiplexing.

Assay Multiplexibility

Cross-reactivity between antibodies should be considered when multiplexing immunoassays to obtain the best results. While these assays have been extensively optimized to minimize nonspecific binding, homologies among proteins in the same family, especially in the phosphorylated domains, make the multiplexing of some assays impractical. It is not recommended to multiplex assays with shared bead regions and 10% or greater cross-reactivity. Refer to Table 3 for assay multiplexibility.

Table 3. Assay multiplexibility. Only assays with known cross-reactivity are shown.

Assays	Cross-Reactivity >10%
β-Actin	Akt (Ser^{473}), Akt (Thr^{308}), Stat3 (Tyr^{705})
Akt (Total)	Smad2
Akt (Ser^{473})	β-Actin, Akt (Thr^{308})
Akt (Thr^{308})	β-Actin, ATF-2 (Thr^{71}), Stat3 (Tyr^{705}), Akt (Ser^{473})
ATF-2 (Thr^{71})	Akt (Thr^{308}), c-Jun (Ser^{63})
Btk (Tyr^{223})	HER-2 (Tyr^{1248}), Lyn (Tyr^{507}), PI3K p85 (Tyr^{458}), Src (Tyr^{416})
c-Abl (Tyr^{245})	EGFR (Tyr^{1173}), IR-β (Tyr^{1146}), PI3K p85 (Tyr^{458})
c-Jun (Ser^{63})	ATF-2 (Thr^{71}), P70 S6 Kinase (Thr^{421}/Ser^{424})
EGFR (Tyr^{1068})	HER-2 (Tyr^{1248}), IGF-1R (Tyr^{1131}), Stat1 (Tyr^{701}), Stat3 (Tyr^{705}), EGFR (Tyr^{1173})
EGFR (Tyr^{1173})	c-Abl (Tyr^{245}), HER-2 (Tyr^{1248}), IGF-1R (Tyr^{1131}), Stat1 (Tyr^{701}), EGFR (Tyr^{1068})
Erk1/2 (Thr^{202}/Tyr^{204}, Thr^{185}/Tyr^{187})	HER-2 (Tyr^{1248})
HER-2 (Tyr^{1248})	Btk (Tyr^{223}), EGFR (Tyr^{1068}), EGFR (Tyr^{1173}), Erk1/2 (Thr^{202}/Tyr^{204},Thr^{185}/Tyr^{187}), IGF-1R (Tyr^{1131}), Lyn (Tyr^{507}), PDGFR-α (Tyr^{754}), PDGFR-β (Tyr^{751}), PI3K p85 (Tyr^{458}), Stat1 (Tyr^{701})
IGF-1R (Tyr^{1131})	EGFR (Tyr^{1068}), EGFR (Tyr^{1173}), HER-2 (Tyr^{1248}), IR-β (Tyr^{1146}), PDGFR-α (Tyr^{754}), PDGFR-β (Tyr^{751})
IR-β (Tyr^{1146})	c-Abl (Tyr^{245}), IGF-1R (Tyr^{1131}), Lyn (Tyr^{507}), PI3K p85 (Tyr^{458}), Src (Tyr^{416}), ZAP-70 (Tyr^{319})
IRS-1 (Ser^{636}/Ser^{639})	PI3K p85 (Tyr^{458})
IκB-α (Ser^{32}/Ser^{36})	NF-κB p65 (Ser^{536})
Lyn (Tyr^{507})	Btk (Tyr^{223}), HER-2 (Tyr^{1248}), IR-β (Tyr^{1146}), PI3K p85 (Tyr^{458})
MEK1 (Ser^{217}/Ser^{221})	P70 S6 Kinase (Thr^{389}), P70 S6 Kinase (Thr^{421}/Ser^{424})
NF-κB p65 (Ser^{536})	IκB-α (Ser^{32}/Ser^{36}), Stat3 (Tyr^{705})
p38 MAPK (Total)	c-Jun
p70 S6 Kinase (Thr^{389})	MEK1 (Ser^{217}/Ser^{221}), p70 S6 Kinase (Thr^{421}/Ser^{424})
p70 S6 Kinase (Thr^{421}/Ser^{424})	c-Jun (Ser^{63}), MEK1 (Ser^{217}/Ser^{221}), p70 S6 Kinase (Thr^{389})
PDGFR-α (Tyr^{754})	HER-2 (Tyr^{1248}), IGF-1R (Tyr^{1131}), PDGFR-β (Tyr^{751})
PDGFR-β (Tyr^{751})	HER-2 (Tyr^{1248}), IGF-1R (Tyr^{1131}), PDGFR-α (Tyr^{754})
PI3K p85 (Tyr^{458})	Btk (Tyr^{223}), c-Abl (Tyr^{245}), HER-2 (Tyr^{1248}), IR-β (Tyr^{1146}), IRS-1 (Ser^{636}/Ser^{639}), Lyn (Tyr^{507}), Syk (Tyr^{352}), ZAP-70 (Y^{319})
Src (Tyr^{416})	Btk (Tyr^{223}), IR-β (Tyr^{1146})
Stat1 (Tyr^{701})	EGFR (Tyr^{1068}), EGFR (Tyr^{1173}), HER-2 (Tyr^{1248}), Stat3 (Ser^{727}), Stat3 (Tyr^{705})
Stat3 (Ser^{727})	Stat1 (Tyr^{701}), Stat3 (Tyr^{705})
Stat3 (Tyr^{705})	β-Actin, Akt (Thr^{308}), EGFR (Tyr^{1068}), NF-κB p65 (Ser^{536}), Stat1 (Tyr^{701}), Stat3 (Ser^{727})
Syk (Tyr^{352})	PI3K p85 (Tyr^{458})
ZAP-70 (Tyr^{319})	IR-β (Tyr^{1146}), PI3K p85 (Tyr^{458})

Cell Lysate Controls

To identify the appropriate controls for your Bio-Plex Pro cell signaling phosphoprotein, total target, or housekeeping protein assay of interest, refer to Table 4.

Table 4. Bio-Rad lysate controls for phosphoprotein, total target, and housekeeping protein assays.

Phosphoprotein	Lysate Control	Catalog #
Akt (Ser^{473}) Akt (Thr^{308}) Erk1/2 (Thr^{202}/Tyr^{204}, Thr^{185}/Tyr^{187}) GSK-3α/β (Ser^{21}/Ser^{9}) MEK1 (Ser^{217}/Ser^{221})	EGF-treated HEK-293	171-YZ0001
ATF-2 (Thr^{71}) c-Jun (Ser^{63}) CREB (Ser^{133}) JNK (Thr^{183}/Tyr^{185}) p38 MAPK (Thr^{180}/Tyr^{182}) p53 (Ser^{15})	UV-treated HEK-293	171-YZ0009
BAD (Ser^{136}) IRS-1 (Ser^{636}/Ser^{639}) mTOR (Ser^{2448}) PDGFR-α (Tyr^{754}) PDGFR-β (Tyr^{751}) PTEN (Ser^{380})	PDGF-treated NIH3T3	171-YZ0007
Btk (Tyr^{223}) Lyn (Tyr^{507}) PI3K p85 (Tyr^{458}) Syk (Tyr^{352})	H_2O_2-treated Ramos	171-YZ0011
c-Abl (Tyr^{245})	Untreated K-562	171-YZT003
EGFR (Tyr^{1068}) EGFR (Tyr^{1173})	EGF-treated HeLa	171-YZ0002
HER-2 (Tyr^{1248}) HSP27 (Ser^{78}) p90 RSK (Ser^{380}) S6 Ribosomal Protein (Ser^{235}/Ser^{236})	EGF-treated SK-BR-3	171-YZ0003
IGF-1R (Tyr^{1131}) IR-β (Tyr^{1146})	IGF-1-treated HEK-293	171-YZ0005
IκB-α (Ser^{32}/Ser^{36}) NF-κB p65 (Ser^{536}) Smad2 (Ser^{465}/Ser^{467})	TNF-α-treated HeLa	171-YZ0008
p70 S6 Kinase (Thr^{421}/Ser^{424}) p70 S6 Kinase (Thr^{389})	NGFβ-treated PC12	171-YZ0006
Src (Tyr^{416})	Src-transfected NIH3T3	171-YZ0013
Stat1 (Tyr^{701}) Stat3 (Ser^{727}) Stat3 (Tyr^{705})	IFN-α-treated HeLa	171-YZ0004
VEGFR-2 (Tyr^{1175})	VEGF-treated HUVEC	171-YZ0010
ZAP-70 (Tyr^{319})	H_2O_2-treated Jurkat	171-YZ0012
Negative control for all phosphoprotein assays	Phosphatase-treated HeLa	171-YZB001

Total Target	Lysate Control	Catalog #
Total Akt Total Erk1/2 Total GSK-3β Total IκB-α Total JNK Total MEK1 Total mTOR Total p38 MAPK Total p70 S6 Kinase Total PTEN Total Smad2 Total IGF-1R	Untreated HeLa	171-YZT002
Total Btk	H_2O_2-treated Ramos	171-YZ0011
Total ZAP-70	H_2O_2-treated Jurkat	171-YZ0012
Total c-Jun Total CREB	Untreated HEK-293	171-YZT001
Total HER-2	EGF-treated SK-BR-3	171-YZ0003
Total Src	Src-transfected NIH3T3	171-YZ0013
Negative control for all total target assays	Detection antibody diluent*	

Housekeeping Protein	Lysate Control	Catalog #
β-Actin	Untreated HeLa	171-YZT002
Human GAPDH	Untreated HeLa	171-YZT002
Negative control for all housekeeping protein assays	Detection antibody diluent*	

* Detection antibody diluent is included with the Bio-Plex Pro cell signaling reagent kit (171-304006M).

The Bio-Plex suspension array system includes fluorescently labeled microspheres and instrumentation licensed to Bio-Rad Laboratories, Inc. by the Luminex Corporation.

POWERFUL Cell Signaling TECHNOLOGY CONTENT

CST antibodies developed and validated for Bio-Plex cell signaling, phosphoprotein, and total target assays.

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