The Protein Experts

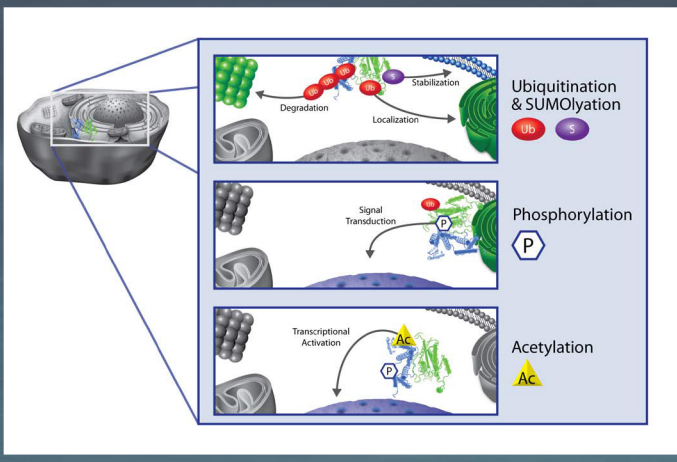
Cytoskeleton, Inc.

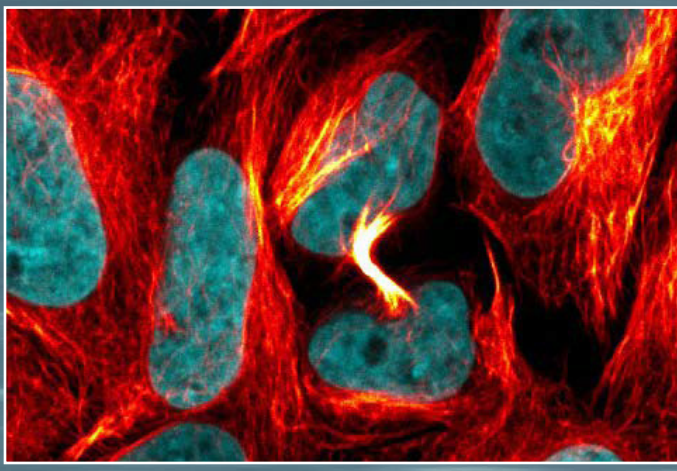
Cytoskeleton, Inc.

Minicatalog 2017

Axil Scientific
YOUR RESEARCH IS OUR BUSINESS

Signal Seeker™ Kits
Spirochrome™ Bioprobes
GLISA™ Activation Assays
Pulldown Activation Assays
GOBlot™ WB Processor
Custom Services
Actin Proteins
Motor Proteins
Small G-proteins
Tubulin Proteins





Helping advance science
one protein *at a time.*

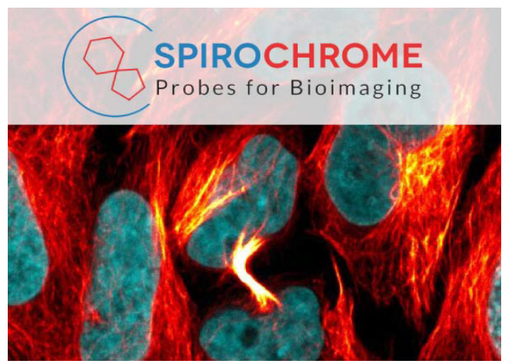
New Products Inside!
Signal Seeker™ Kits and Antibodies
GOBlot™ WB Processor Colors
Spirochrome™ Live Cell Probes

Table of Contents

Online Resources at Cytoskeleton.com

- Promotions and Discounts
- Technical Resources
- Monthly Newsletters
- Video Guides
- Protocols
- Webinars

New Products



NEW Far-Red bio-imaging probes!
Illuminate the cytoskeleton with low toxicity Ex/Em 690 nm / 720 nm wavelengths.
- Low toxicity
- Very high post-experimental viability
- Very bright

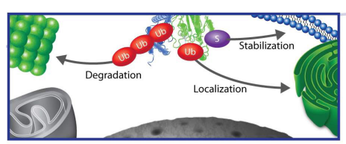
See p. 7 for more information

GOBlot™
Western Blot Processor

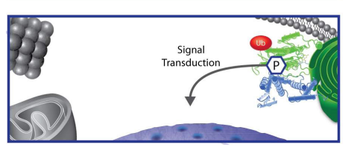
- Time saving - fully automated
- Choice of four routines
- Choice of six colors
- Ask about the no obligation trial period

See p. 6 for more information

Signal Seeker™ Kits
Looking deeper, and discovering more...



Ubiquitination & SUMOlyation

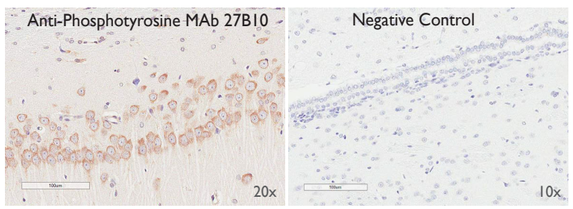


Phosphorylation

- User-friendly kits: One day, one lysate, real insight.
- Follow multiple signalling time profiles
- Measure endogenous signalling events
- Fully complemented kits - everything included.

See p. 4-5 for more information

Signal Seeker™ Antibodies



Cat. # APY03 MAb clone 27B10 at 1:125 dilution.

Non-specific mouse IgG2b

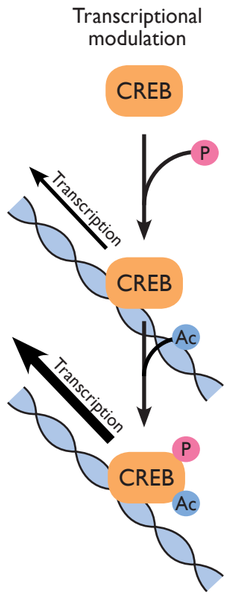
- Highly validated for immunohistochemistry
- High affinity & specificity for phosphotyrosine

Check out our full line of Signal Seeker antibodies for Phosphorylation, Acetylation, Ubiquitination, and SUMOylation post-translational modifications.

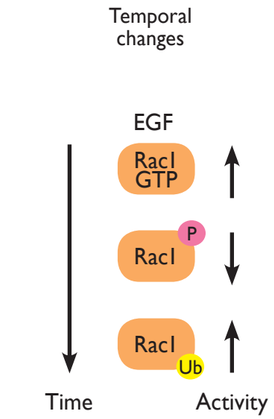
See p. 12 for more information

Signal Seeker Kits™ - looking deeper, and discovering more...

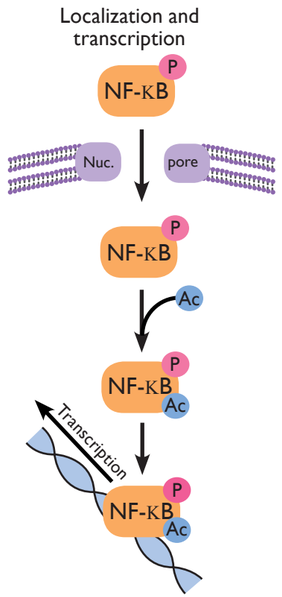
Discovering potentially novel regulatory mechanisms



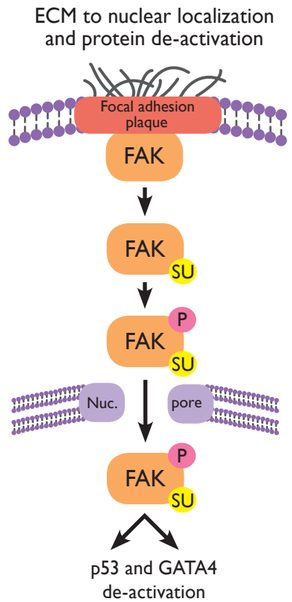
Paz et al. 2014. Combinatorial regulation of a signal-dependent activator by phosphorylation and acetylation PNAS, 111 (48), 17116–17121.



Law et al. 2015. Temporal regulation of phosphotyrosine-modified Rac1 in response to epidermal growth factor. Mol. Biol. Cell. ASCB Annual Meeting 2015, Poster P2126.



Ashburner et al 2001. The p65 (RelA) subunit of NF-kappaB interacts with... Mol Cell Biol. 2001 Oct;21(20):7065-77.
Chen and Greene, 2004. Shaping the nuclear action of NF-κB. Nat. Rev. Mol. Cell Biol. 5 p.392-401.



Lim 2013. Nuclear FAK: a New Mode of Gene Regulation from Cellular Adhesions. Mol. Cells. 36(1): 1–6.

User-friendly kits to investigate cutting edge signalling mechanisms

Uses in molecular biology

- Use different kits to build a temporal protein regulation profile.
- Investigate the role of known protein modifications in your system.
- Detect endogenous levels (vs. transfected amounts) of modified proteins.
- Discover novel biomarkers.

Advantages for immediate results

- User friendly kits speed your rate of discovery.
- Optimized buffers with PTM stabilizing composition.
- Kits contain all reagents and clarifying columns necessary to perform experiments.

Protein regulation during signal transduction and other cellular events is, by necessity, a rapid and dynamic process. Most often, these mechanisms involve modification of an extremely small fraction of the protein target. This makes the scientist's job of capturing key regulatory processes difficult and frustrating.

Signaling pathways were thought to depend on one protein modification per step, however recent developments have established some if not many key pathway steps are controlled by multiple types of modifications. Examples are a) co-acetylation and phosphorylation of CREB enhancing transcription (Paz et al. 2014), b) phosphorylation and SUMOylation of FAK creates a nuclear function for this archetypal focal adhesion protein (Lim 2012), and c) requirement for acetylation of an adjacent amino-acid of phospho-modified NF-κB p65 prior to transcriptional initiation (Ashburner 2001).

At Cytoskeleton, we have focused on generating accurate methods to measure these changes when they oscillate in a short timeframe and consequently generate a small percent of modified protein. Small percents of activated protein are well known to regulate pathways in the Small G-protein field where 0.5 to 1.0% of signal is considered significant, the underlying mechanism is thought to be rapid de-activation after the signaling event has passed through the step. The new kits allow end users to analyze endogenous modifications within one day and from one extract.

Signal Seeker SUMO 2/3 Enrichment Kit™ Cat. # BK162. 30 assays.

The functional component of the SUMO 2/3 Enrichment Kit is an anti-SUMO-2/3 antibody (clone: 11G2) which is provided as a chemically crosslinked Protein G bead conjugate. The affinity bead reagent was optimized to give no detectable leaching of either heavy or light chains in an IP assay, making the resulting data extremely specific, sensitive and clean (Fig. 1). The affinity beads immunoprecipitate a wide range of SUMO-2/3 targeted proteins in cell extracts (see manual at www.cytoskeleton.com/bk162). Both endogenous mono- and poly- SUMOylated proteins are easily detected with characteristic multiple bands in the Western Blot detection method.

Uses:

- Investigate transient regulatory mechanisms.
- Measure signalling events of multiple pathway member proteins.
- Discover new modifications of your protein of interest.

References

1. Barysch S. et al. 2014. Identification and analysis of endogenous SUMO1 and SUMO2/3 targets in mammalian cells and tissues using monoclonal antibodies. Nat Protoc. 9(4):896-909
2. Becker J. et al. 2013. Detecting endogenous SUMO targets in mammalian cells and tissues. Nature Struc. & Mol. Biol. 20, 525-531.

Example result: Identification of SUMOylated Ubc9 protein after heat shock.

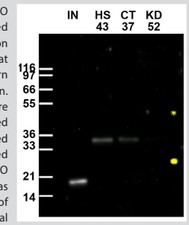


Figure 1 legend: The Signal Seeker SUMO 2/3 Enrichment Kit (Cat. BK162) was used to examine endogenous SUMOylation events in HeLa cells exposed to heat shock. The image shows a western blot result using anti-Ubc9 protein. Lane HS43; Denatured cell lysates were prepared from 42°C 10 min treated HeLa cells HS43: Heat Shock treated 42°C for 10 min, lane CT37: untreated cells, and lane KD52: HeLa siRNA SUMO knockdown extract. 1 mg of lysate was used for the immuno-precipitation of SUMO-2/3 conjugates. Note: Chemical conjugation of 11G2 to agarose beads prevents heavy and light chain leaching completely. A small or large dot indicate positions of putative heavy and light chains of antibodies if they were present. Unconjugated free SUMO is apparent at 18 kDal and a higher molecular-weight band is present in the heat shock lane, indicating that Ubc9 is conjugated by single SUMO-2/3 protein as previously reported (Refs. 1,2).

Signal Seeker Ubiquitin Enrichment Kit™ Cat. # BK161. 30 assays.

Cytoskeleton scientists have developed a proprietary formulation of ubiquitination binding domains (UBDs) that have the unique characteristic of capturing both mono-ubiquitinated and poly-ubiquitinated proteins with high affinity. As mono-, multi– and poly-ubiquitination often confer unique, non-redundant properties to their target protein it is crucial to obtain the complete ubiquitin profile of any given target protein. Cytoskeleton's Signal Seeker Ubiquitin Enrichment Kit is a powerful tool for the complete analysis of protein ubiquitination proflies.

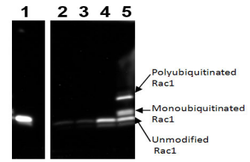
Uses:

- Detect highly transient regulation of protein modifications.
- Show endogenous mono- and poly-ubiquitinylation to confirm transfection or proteomic approaches.
- Use different kits to build a temporal protein regulation profile.

References

1. Luo H-B, et al. 2014. SUMOylation at K340 inhibits Tau degradation through deregulating its phosphorylation and ubiquitination. Proc. Natl. Acad. Sci. USA 111: 16586-16591.
2. Law et al. 2015 Temporal regulation of phosphotyrosine-modified Rac1 in response to epidermal growth factor. Mol. Biol. Cell. ASCB Annual Meeting 2015, Poster P2126.

Example result: Identification of mono- and poly-ubiquitinated forms of Rac1.



Legend: The Signal-Seeker Ubiquitin Enrichment Kit (Cat. # BK161) was used to examine ubiquitination of endogenous Rac1 in HeLa cells treated with CNF1 toxin (Cat. # CN04). The image shows a western blot result using anti-Rac1 antibody (Cat. # ARC03). Lane 1: 10 µg input lysate showing total Rac1 levels, Lane 2: BSA bead control plus untreated lysate , Lane 3: BSA bead control plus CN04 treated lysate, Lane 4: Ubiquitin Affinity beads plus untreated lysate, Lane 5: Ubiquitin Affinity beads plus CN04 treated lysate. Note: Both mono- and poly-ubiquitinated forms of Rac1 could be detected from 300 µg of HeLa cell lysate.

Signal Seeker Phosphotyrosine Enrichment Kit™ Cat. # BK160. 30 assays.

The Phosphotyrosine Enrichment Kit has been developed over the past four years to create an extremely robust and accurate method of measuring tyrosyl phosphorylation events. The included protocol is a very sensitive method to detect sub-nanogram amounts of modified proteins, and to enable high fidelity time courses to be analyzed. The exquisite accuracy means that subtle changes in signal, as seen in the adjacent figures) can easily be detected and reproduced for publication quality data .

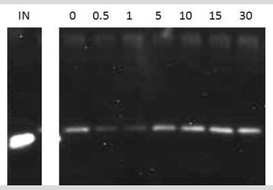
Uses:

- Pharmacological investigation of phospho-protein isoforms in your pathway of interest.
- Investigation of the relationship between phosphorylation events and other signalling events.
- Identification and Development of biomarkers.

References

1. Bunda S. et al. 2014. Src promotes GTPase activity of Ras via tyrosine 32 phosphorylation PNAS E3785-E3794.
2. Law et al. 2015. Temporal regulation of phosphotyrosine-modified Rac1 in response to epidermal growth factor. Mol. Biol. Cell. ASCB Annual Meeting 2015, Poster P2126.

Example result: Measurement of rapidly changing levels of Rac1 tyrosyl phosphorylation.



Legend: Anti-Rac1 antibody (Cat. # ARC03) was used to probe a blot containing samples from a time course of A431 cells treated with EGF. Time is noted in minutes at head of lane, IN = total Rac1 in extract. All other lanes indicate the concentrated immuno-precipitation purified phospho-form of Rac1 isolated by the Signal Seeker Kit. Note: Steady state basal levels of tyrosyl phosphorylated Rac1 was rapidly reduced after 30 s of EGF treatment, and returning to basal and then higher levels after 10 min. Each lane represents an input of 1000 µg of cell lysate.

Kit or Affinity Bead	Type	Reactions	Cat. #
Signal Seeker™ Phosphotyrosine Enrichment Kit	Kit	30	BK160
Phosphotyrosine Affinity Beads	Beads	40-80	APY03-beads
Signal Seeker™ SUMO 2/3 Enrichment Kit	Kit	30	BK162
SUMO 2/3 Affinity Beads	Beads	20-40	ASM24-beads
Signal Seeker™ Ubiquitin Enrichment Kit	Kit	30	BK161
Ubiquitin Affinity Beads	Beads	40	UBA01-beads
Control for Ippt IgG Beads	Beads	10	CIG01-beads
Control for Ubiquitin Affinity Beads	Beads	10	CUB01

GOBlot

The First Affordable Western Blot Processor

Save Time and Money

- Save up to 3 hours per day
- Fully automated
- Choice of four routines
- Choice of six colors
- Recycle primary antibody
- No obligation trial period

Recycle the Primary Antibody

Legend: A single anti-RhoA monoclonal antibody solution was used to probe four replicate membranes. Note how the 3rd re-cycle test has a reduced band intensity indicating the antibody is being depleted by the repeated process.

Development of the GOBlot

The GOBlot™ Western Blot Processor (patents pending) was developed to be an affordable and helpful device for all scientists. With input from over 300 research scientists, the flexible routines and capabilities of the processor were defined. Western blot automation with the GOBlot saves the average researcher 3 hours a day while improving the reproducibility of results. The GOBlot saves researchers money by providing the option to recycle primary antibody and it does not require machine-specific consumables. See the results for yourself with a no-obligation trial period or read customer reviews online at Biocompare.com.

GOBlot Options	Cat. #
GOBlot (1 Machine)	WBM01
GOBlot (4 Machine Bundle)	WBM01

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Ask about the no obligation trial period by e-mailing
tservice@cytoskeleton.com

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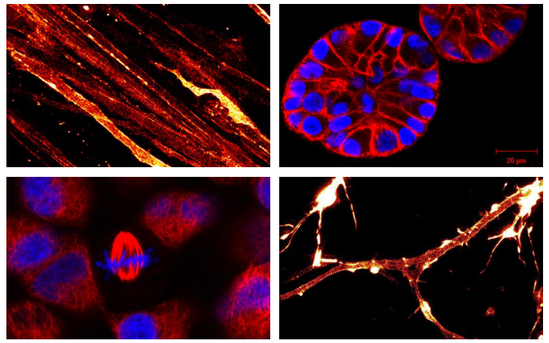


Top Left: STED image of dorsal root ganglion (rat) stained with SiR-actin. Courtesy Of Elisa D'Este, MPI Biophysical Chemistry, Göttingen. **Top Right:** fMCF10A cells expressing H2B-GFP (Blue) in Matrigel stained with SiR-actin (red). Image taken on an inverted LSM microscope. Courtesy of Christian Conrad and Katharina Jechow, Heidelberg. **Lower Left:** HeLa cells expressing H2B-mCherry (Blue) stained with SiR-Tubulin (Red), courtesy of Daniel Gerlich and Claudia Blaukopf, Inst. Mol. Biotech., Vienna). **Lower Right:** STED image of cultured rat hippocampal neurons stained with SiR-actin. Actin rings (stripes) with 180 nm periodicity can be seen. Courtesy Of Elisa D'Este, MPI Biophysical Chemistry, Göttingen.

SPIROCHROME
Probes for Bioimaging
www.cytoskeleton.com/spirochrome

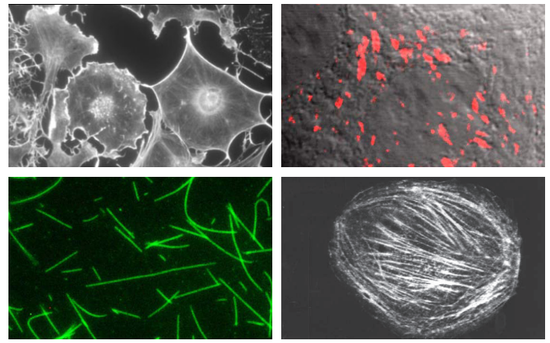
Tubulin Imaging

Description	Ex / Em	Cat. #	Amount
NEW **SiR700-Tubulin Kit** Includes SiR700-Tubulin and Verapamil	690 / 720 nm	CY-SC014	35 nmol
SiR-Tubulin Kit Includes SiR-Tubulin and Verapamil	630 / 680 nm	CY-SC002	50 nmol
Cytoskeleton Kit Includes SiR-Actin, SiR-Tubulin, and Verapamil	630 / 680 nm	CY-SC006	50 nmol each
AMCA Labeled Tubulin	350 / 440 nm	TL440M-A TL440M-B	5 x 20 µg 20 x 20 µg
HiLyte Fluor™ 488 Labeled Tubulin	460 / 520 nm	TL488M-A TL488M-B	5 x 20 µg 20 x 20 µg
TRITC Rhodamine Labeled Tubulin	535 / 590 nm	TL590M-A TL590M-B	5 x 20 µg 20 x 20 µg
X-Rhodamine Labeled Tubulin	560 / 620 nm	TL620M-A TL620M-B	5 x 20 µg 20 x 20 µg
HiLyte Fluor™ 647 Labeled Tubulin	620 / 670 nm	TL670M-A TL670M-B	5 x 20 µg 20 x 20 µg

ECM Imaging

Description	Ex / Em	Cat. #	Amount
Fibronectin Red fluorescent, rhodamine	535 / 590 nm	FNR01-A FNR01-B	5 x 20 µg 20 x 20 µg
Fibronectin Green fluorescent, HiLyte Fluor™ 488	460 / 520 nm	FNR02-A FNR02-B	5 x 20 µg 20 x 20 µg
Fibronectin Biotinylated	na	FNR03-A FNR03-B	5 x 20 µg 20 x 20 µg
Laminin Red fluorescent, rhodamine	535 / 590 nm	LMN01-A LMN01-B	5 x 20 µg 20 x 20 µg
Laminin Green fluorescent, HiLyte Fluor™ 488	460 / 520 nm	LMN02-A LMN02-B	5 x 20 µg 20 x 20 µg
Laminin Biotinylated	na	LMN03-A LMN03-B	5 x 20 µg 20 x 20 µg

SiR-Actin, Sir-Tubulin, SiR-DNA, SiR-Lysosome are trademarks of Spirochrome SA (Switzerland). HiLyteFluor is a trademark of Anaspec Inc. (CA, USA).



Top Left: Swiss 3T3 cells treated with cell permeable Rho inihibitor (Cat. # CT04) and stained with rhodamine phalloidin (Cat. PHDR1); note the lack of F-actin stress fibers in each cell. **Top right:** Fluorescent fibronectin (Cat. # FNR01) treated MCF10A cells (image kindly provided by A. Varadara and M. Karthykenyan, Univ. S.Carolina, Columbia, SC). **Lower left:** 488 HiLyte Fluor™ labeled tubulin polymerized *in vitro* (Cat. # TL488M). **Lower right:** Fluorescent non-muscle actin (Cat.# APHR) injected in to CHO cells; note the stress fibers across the whole cell width (kindly provided by Dr. Goldman's lab, Northwestern Univ, Chicago, IL).

Small G-protein Modulators and Actin Imaging

Description	Ex / Em	Cat. #	Amount
NEW **SiR700-Actin Kit** Includes SiR-Actin and Verapamil	690 / 720 nm	CY-SC013	35 nmol
SiR-Actin Kit Includes SiR700-Actin and Verapamil	630 / 680 nm	CY-SC001	50 nmol
Cytoskeleton Kit Includes SiR-Actin, SiR-Tubulin, and Verapamil	630 / 680 nm	CY-SC006	50 nmol each
Rhodamine Actin Protein Human platelet, non-muscle	535 / 590 nm	APHR-A APHR-C	4 x 10 µg 20 x 10 µg
Rhodamine Actin protein Rabbit skeletal muscle	535 / 590 nm	AR05-B AR05-C	10 x 20 µg 20 x 20 µg
Rho Activator II Deamidation of Rho Gln-63		CN03-A CN03-B	3 x 20 µg 9 x 20 µg
Rho Inhibitor I ADP ribosylation of Rho Asn-41		CT04-A CT04-B CT04-C	1 x 20 µg 5 x 20 µg 20 x 20 µg
Rho/Rac/Cdc42 Activator I Deamidation of Rho Gln-63 & Rac/Cdc42 Gln-61		CN04-A CN04-B	3 x 20 µg 9 x 20 µg
Rho Activator I SHP-2 phosphatase-mediated Rho activation		CN01-A CN01-B	5 x 10 units 20 x 10 units
Rac/Cdc42 Activator II EGF receptor-mediated Rac/Cdc42 activation		CN02-A CN02-B	5 x 10 units 20 x 10 units

Lysosome Imaging

DNA Live Cell Imaging Reagents	Ex / Em	Cat. #	Amount
NEW **SiR-Lysosome Kit** Includes SiR-Lysosome and Verapamil	630 / 680 nm	CY-SC012	50 nmol
NEW **SiR700-Lysosome Kit** Includes SiR700-Lysosome and Verapamil	690 / 720 nm	CY-SC016	35 nmol

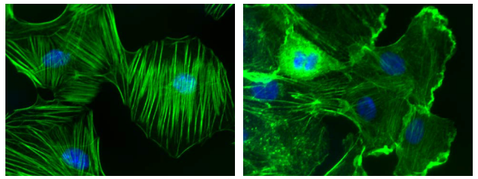
DNA Imaging

DNA Live Cell Imaging Reagents	Ex / Em	Cat. #	Amount
SiR-DNA Kit Includes SiR-DNA and Verapamil	630 / 680 nm	CY-SC007	50 nmol
NEW **SiR700-DNA Kit** Includes SiR700-DNA and Verapamil	690 / 720 nm	CY-SC015	35 nmol

About Activation Assays

Since 2001, Cytoskeleton has provided the scientific community with the most robust, accurate, and time-saving kits to measure Small GTP-binding protein (SmG) activation. Along the way, we have developed numerous versions for different SmGs, such as Rho, Rac, Arf1 & 6, Ras, and Ral. Also, the quantifiable GLISA versions enabled a new wave of more sensitive applications, e.g. measurement in limited primary cell numbers and Matrigel 3D matrix. We continue to develop and maintain these high standards, which allow you to produce the best results in the least amount of time.

SmGs are involved in regulating cell signaling pathways and impact a wide range of cellular processes, functions, and morphology. The Pulldown version of the assay uses affinity beads which are incubated with the extract and then separated by centrifugation. The pelleted products are separated by SDS-PAGE and blotted onto a membrane for western analysis of the SmG of interest. The GLISA® format is a modified ELISA which has the affinity reagent permanently attached to the well of a 96-well plate. The extract is incubated in the well which is then washed and probed with primary and secondary antibodies.



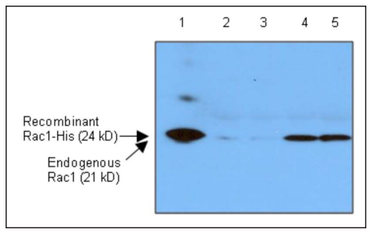
Legend: Rho activation (left) and Rac activation (right) in Swiss 3T3 cells. F-actin is visualized with fluorescent green phalloidin staining (Cat.# PHDG1) and nuclear blue DNA staining with Dapi. Cells were activated with Cat.# CN03 (left) and Cat. # CN04 (right).

Comparison of Pulldown and GLISA formats

Parameter	Pull-down	G-LISA®
Total protein per assay	500-2000 µg	10-50 µg
Assay time	10-12 h (2 days)	<3 h
Primary cells & 3D matrix compatible	No	Yes
Sample handling	10 Samples	96 Samples
Quantitative data*	Semi	Yes

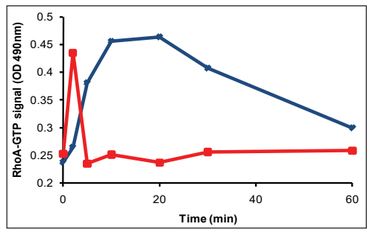
* Numerical readouts and fewer sample handling steps make G-LISA® assays more quantitative.

Pull-down Result Example



Swiss 3T3 cells were serum-starved for 24h; after this, a sample was treated with 10 ng/ml of EGF for 2 min (Lanes 4 & 5). Other cells were not treated and remained serum-starved (Lanes 2 & 3). Rac1 activation was measured using the Rac1 Activation pull-down assay. 500 µg of lysate were assayed with 10 µg of PAK-PBD beads (Lanes 2-5). Lane 1 shows 20 ng of recombinant Rac1-His protein run as a Western blot standard.

G-LISA® Result Example



Time course of activation of RhoA in Swiss 3T3 cells by CN01 and LPA. Serum-starved Swiss 3T3 cells were treated with Rho Activator I, Cat. # CN01 (blue diamonds) or LPA (red squares). RhoA activity was measured by reading signals at OD_{490nm}. Data are background subtracted.

Example Product Citations

RhoA G-LISA® (Cat. # BK124)
Chen W. et al., 2016. *Oncol. Lett.* **11**, 1375-1381.
Jones E.L. et al., 2016. *J. Immunol.* DOI: 10.4049/jimmunol.1500357.

Rac1,2,3 G-LISA® (Cat. # BK125)
Qin G. et al., 2016. *Cancer Lett.* **374**, 85-95.
Marusak C. et al., 2016. *Pharmacol. Res.* **113**, 515-520.

Rac1 G-LISA® (Cat. # BK128)
Ubba V. et al., 2016. *Reprod Sci.* DOI: 10.1177/1933719116669057.
Sayyad W.A. et al., 2016. *PLoS One.* **11,** e0146842.

Cdc42 G-LISA® (Cat. # BK127)
Jones E.L. et al., 2016. *J. Immunol.* DOI: 10.4049/jimmunol.1500357.
Croise P. et al., 2016. *Endocr. Relat. Cancer.* **23,** 281-293.

Ras G-LISA® (Cat. # BK131)
L.D. Camargo et al., 2013. *Free Radic. Biol. Med.* **65**, 1398-1407.
S.-J. Lee et al., 2013. *J. Biol. Chem.* **288**, 25244-25253.

More online!
www.cytoskeleton.com/activation-assays

Activation Assays

Pull-down Activation Assays

Pull-down assays utilize affinity beads linked to an effector protein that selectively binds active GTPase followed by quantitation with Western blotting.

Pull-down Activation Assays	Cat. #	Amount
Combo RhoA/Rac1/Cdc42 Activation Assay Biochem Kit™	BK030	3 x 10 assays
Arf1 Activation Assay Biochem Kit™	BK032-S	20 assays
Arf6 Activation Assay Biochem Kit™	BK033-S	20 assays
Cdc42 Activation Assay Biochem Kit™	BK034-S BK034	20 assays 50 assays
Rac1 Activation Assay Biochem Kit™	BK035-S BK035	20 assays 50 assays
RalA Activation Assay Biochem Kit™	BK040	50 assays
Ras Activation Assay Biochem Kit™	BK008-S BK008	20 assays 50 assays
RhoA Activation Assay Biochem Kit™	BK036-S BK036	20 assays 80 assays
Protease Inhibitor Cocktail (100x stock)	PIC02	1 ml

For isoforms not listed, see our information resources online.

G-LISA® Activation Assays

G-LISAs use a 96-well plate coated with effector protein that selectively binds the active GTPase followed by quantitation with ELISA techniques.

G-LISA Activation Assays	Cat. #	Amount
RhoA/Rac1/Cdc42 G-LISA Activation Assay Bundle BK135=BK124-S+BK127-S+BK128-S	BK135	3 Kits (24 assays/kit)
Arf1 G-LISA® Activation Assay, colorimetric	BK132	96 assays
Arf6 G-LISA® Activation Assay, colorimetric	BK133	96 assays
Cdc42 G-LISA® Activation Assay, colorimetric	BK127-S BK127	24 assays 96 assays
Rac1,2,3 G-LISA® Activation Assay, colorimetric	BK125	96 assays
Rac1 G-LISA® Activation Assay, colorimetric	BK128-S BK128	24 assays 96 assays
Rac1 G-LISA® Activation Assay, luminescence	BK126	96 assays
RalA G-LISA® Activation Assay, colorimetric	BK129	96 assays
Ras G-LISA® Activation Assay, colorimetric	BK131	96 assays
RhoA G-LISA® Activation Assay, colorimetric	BK124-S BK124	24 assays 96 assays
RhoA G-LISA® Activation Assay, luminescence	BK121	96 assays
Protease Inhibitor Cocktail (100x stock)	PIC02	1 ml

Related Activation Assay Products

Total RhoA ELISA

Rapidly measure Total RhoA from cell or tissue lysates using the extremely sensitive and linear Total RhoA ELISA.

ELISA	Cat. #	Amount
Total RhoA ELISA	BK150	96 assays

Activators & Inhibitors

G-switch™ small G-protein activators and inhibitors are highly potent reagents that target endogenous Rho family proteins and pathways.

See Pg. 10

Acti-stain Phalloidins

Acti-stain™ fluorescent phalloidins provide exceptionally bright and stable probes for F-actin at an economical price.

See Pg. 13

GTPase Affinity Beads & Proteins

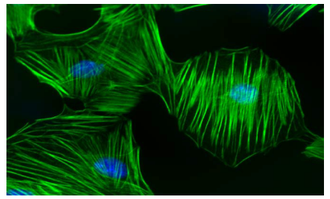
Specifically target the active form of small G-proteins with these brightly-colored GTPase affinity beads and proteins.

See Pg. 10

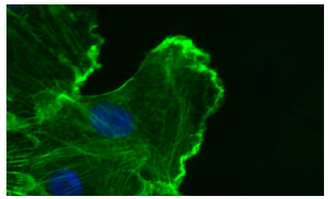
G-switch™ Activators & Inhibitors

The G-switch™ line of small G-protein tools are highly potent reagents that target endogenous Rho family proteins and pathways. In contrast to methods that rely on over-expression or knockdown of target proteins (e.g., DNA transfection of dominant-negative or constitutively-active Rho mutants, RNAi knockdown), G-switch™ reagents act rapidly on the endogenous target protein (in minutes to hours), thereby optimizing the chance of generating a more physiologically relevant response.

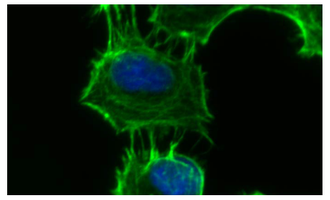
G-protein Modulator	Cell Entry Mechanism	Protein Modulation	Cat. #	Amount
Rho Activator II Deamidation of Rho Gln-63	Cell permeable	Direct	CN03-A CN03-B	3 x 20 µg 9 x 20 µg
Rho Inhibitor I Specific inhibitor of Rho activity, ADP ribosylation of Rho Asn-41 (very cell permeable)	Cell permeable	Direct	CT04-A CT04-B CT04-C	1 x 20 µg 5 x 20 µg 20 x 20 µg
C3 Transferase Protein Specific inhibitor of Rho activity, ADP ribosylation of Rho Asn-41 (limited cell permeability)	Pinocytosis	Direct	CT03-A CT03-C	1 x 25 µg 4 x 25 µg
Rho/Rac/Cdc42 Activator I Deamidation of Rho Gln-63 & Rac/Cdc42 Gln-61	Cell permeable	Direct	CN04-A CN04-B	3 x 20 µg 9 x 20 µg
Rho Activator I SHP-2 phosphatase-mediated Rho activation	Cell permeable	Indirect	CN01-A CN01-B	5 x 10 units 20 x 10 units
Rac/Cdc42 Activator II EGF receptor-mediated Rac/Cdc42 activation	Receptor mediated	Indirect	CN02-A CN02-B	5 x 10 units 20 x 10 units



Stress fibers caused by Rho activation using Cat. # CN03. Actin stained green with Cat. # PHDG1..



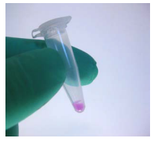
Membrane ruffles induced by Rac activation using Cat. # CN04. Actin stained green with Cat. # PHDG1.



Microspikes induced by Cdc42 activation using Cat. # CN02. Actin stained green with Cat. # PHDG1.

GTPase Affinity Beads & Proteins

GTPase Affinity Beads and Proteins	Purity	Cat. #	Amount
GGA3-PBD Beads Binds active (GTP-bound) Arf1 and Arf6	>85%	GGA05-A	1 x 500 µg
PAK-PBD Protein Binds active (GTP-bound) Cdc42 and Rac1,2,3	>80%	PAK01-A PAK01-B	1 x 250 µg 4 x 250 µg
PAK-PBD Beads Binds active (GTP-bound) Cdc42 and Rac1,2,3	>80%	PAK02-A PAK02-B	1 x 500 µg 4 x 500 µg
Raf-RBD Beads Binds active (GTP-bound) K-, N-, H-Ras	>80%	RF02-A RF02-B	1 x 2 mg 4 x 2 mg
Rhotekin-RBD Protein Binds active (GTP-bound) RhoA,B,C	>90%	RT01-A	1 x 500 µg
Rhotekin-RBD Beads Binds active (GTP-bound) RhoA,B,C	>85%	RT02-A RT02-B	2 x 2 mg 6 x 2 mg



Specifically target the active form of small G-proteins with these brightly-colored GTPase affinity beads and proteins.

G-protein Effector Proteins

G-protein Modulator & Effector Proteins	Purity	Cat. #	Amount
Dbs His Protein, RhoGEF domain (DH/PH) GEF for Cdc42 and RhoA	>80%	GE01-A	2 x 50 µg
p50RhoGAP GST Protein, full length GAP for Cdc42, Rac, and Rho	>90%	GAP01-A GAP01-B	1 x 50 µg 4 x 50 µg
p50RhoGAP GST Protein, GAP domain GAP for Cdc42, Rac, and Rho	>90%	GAS01-A GAS01-B	1 x 50 µg 4 x 50 µg
RhoGDI GST Protein Inhibitor of Cdc42, Rac, and Rho	>90%	GDI01-A	1 x 25 µg
SOS1 Ras GEF Domain Protein GEF for H-, K- or N-Ras	>90%	CS-SOS1-A CS-SOS1-B	1 x 100 µg 1 x 1 mg

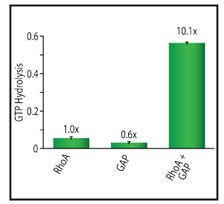
Bulk Quantities Available

Small G-protein Tools

RhoGAP Assay Biochem Kit™

Measures the amount of inorganic phosphate (Pi) produced by GTPase activating protein inducing RhoA GTPase.

- Phosphate detection reagents included
- Reaction buffer included
- Cdc42, Rac1, RhoA, and H-Ras GTPase control proteins
- RhoGAP (Positive control)



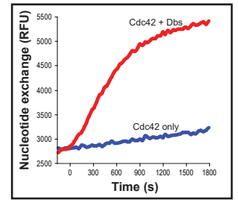
GAP assay using BK105. RhoA alone (RhoA), p50RhoGAP alone (GAP), or RhoA and p50RhoGAP (RhoA+GAP) were incubated and the GTP hydrolysis was measured.

Product	Cat. #	Amount
RhoGAP Assay Biochem Kit™	BK105	80-160 assays

RhoGEF Assay Biochem Kit™

Fluorophore-based assay measures Mant-GTP nucleotide exchange on small G-proteins.

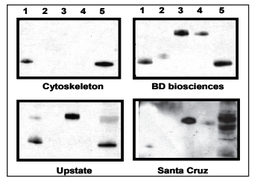
- Mant-GTP included
- Reaction buffer included
- Cdc42, Rac1, and RhoA GTPase control proteins
- GEF domain of Dbs (Positive control for GTP or GDP exchange on RhoA and Cdc42)
- Useful with all other small G-proteins



RhoGEF assay using BK100. Cdc42 alone and Cdc42 + Dbs was incubated and fluorescence intensity (nucleotide exchange) was measured.

Product	Cat. #	Amount
RhoGEF Exchange Assay Biochem Kit™	BK100	60-300 assays

Antibodies for Small G-proteins



Anti-Rac1 monoclonal antibody (Cat. # ARC03) does not cross-react with Rac2, 3, or Cdc42 (upper left blot), while all other commercially available Rac1 antibodies cross-react with GTPases other than Rac1.

Small G-protein Antibodies	Host	Type	Species Reactivity	Cat. #	Amount
Cdc42 Specific Antibody Human Cdc42 Peptide	Mouse	mAb	Hu, Ms, Rt, other extracts	ACD03 ACD03-S	2 x 200 µl 1 x 50 µl
Rac1 Specific Antibody Human C-terminal Peptide	Mouse	mAb	Hu, Ms, Rt, other extracts	ARC03-A ARC03-S	2 x 100 µl 1 x 50 µl
RhoA Specific Antibody Human RhoA Peptide	Mouse	mAb	Hu, Ms, Rt, other extracts	ARH04 ARH04-S	2 x 100 µl 1 x 50 µl

Additional Signal Transduction Reagents

Signal Transduction Reagents	Cat. #	Amount
Total RhoA ELISA Measures total RhoA levels	BK150	96 assays
GTPgS Non-hydrolyzable GTP analog, 50 µl of 20 mM	BS01	1 x 500 µg
GTPase CytoPhos™ Assay One step assay for enzyme Kcat 0.01 to 100	BK054	1000 assays

Bulk Discounts Available
Visit www.cytoskeleton.com/bulk

Purified G-proteins

Purified G-proteins	Purity	Cat. #	Amount
Cdc42 His Protein, constitutively-active (Q61L)	>70%	C6101-A	1 x 10 µg
Cdc42 GST Protein, dominant-negative (T17N)	>90%	C17G01-A	1 x 25 µg
Cdc42 GST Protein, wild-type	>90%	CDG01-C	8 x 25 µg
Cdc42 His Protein, wild-type	>90%	CD01-A CD01-C CD01-XL	1 x 100 µg 3 x 100 µg 1 x 1 mg
Rac1 His Protein, constitutively-active (Q61L)	>90%	R6101-A	1 x 10 µg
Rac1 GST Protein, dominant-negative (T17N)	>90%	R17G01-A	1 x 25 µg
Rac1 GST Protein, wild-type	>90%	RCG01-C	8 x 25 µg
Rac1 His Protein, wild-type	>90%	RC01-A RC01-C RC01-XL	1 x 100 µg 3 x 100 µg 1 x 1 mg
Rac2 His Protein, wild-type	>90%	RC02-A	1 x 100 µg
Rap1b His Protein, wild-type	>90%	RR02-A	1 x 100 µg
H-Ras His Protein, wild-type	>80%	RS01-A RS01-C	1 x 100 µg 3 x 100 µg
NEW **K-Ras4B Protein, wild-type**	>80%	CS-RS03	1 x 100 µg
RhoA His Protein, constitutively-active (Q63L)	>90%	R6301-A	1 x 10 µg
RhoA GST Protein, wild-type	>90%	RHG01-C	8 x 25 µg
RhoA His Protein, wild-type	>80%	RH01-A RH01-C RH01-XL	1 x 100 µg 3 x 100 µg 1 x 1 mg
RhoC His Protein, wild-type	>90%	RH03-A	1 x 100 µg

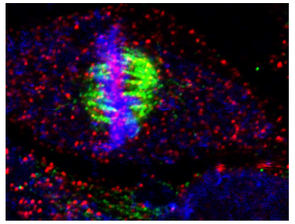
Axil Scientific
YOUR RESEARCH IS OUR BUSINESS

With Cytoskeleton's antibodies and reagents, you will benefit from several distinct advantages for your antibody based reagents:

- All antibodies developed in house
- All antibodies manufactured in house
- Extensive quality control that is visible to the user
- Specialist technical help

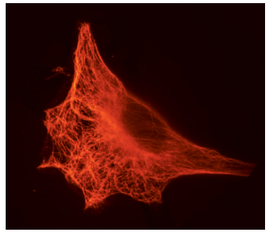
Learn More at
www.cytoskeleton.com/ptm-antibodies
Validation info, analysis, applications, and customer testimonials.

Anti-SUMO-2/3 immuno-fluorescence in mitotic cells



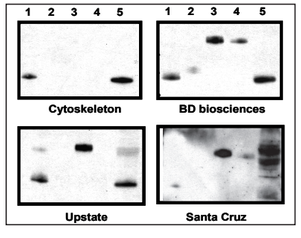
Immunofluorescence of HeLa cells in metaphase with SUMO-2/3 Antibody (Cat. # ASM23, red) and α/β-tubulin antibody (Cat. # ATN02, green). Chromosomal DNA stained with Dapi (blue).

Microtubule Visualizing Antibody (Cat. # ATN02)



Microtubule network in a NIH3T3 cell iluminated with Cytoskeleton's sheep anti-tubulin antibody (ATN02). ATN02 is a pan-tubulin sheep polyclonal antibody, hence it can be co-incubated with mouse, rat or rabbit antibodies for selective dual or triple antibody staining.

Rac1 Antibody Specificity (Cat. # ARC03)



Western blot analysis of small g-protein versus different Rac1 Antibodies. Anti-Rac1 monoclonal antibody (Cat. # ARC03) does not cross-react with Rac2, 3, or Cdc42 (upper left blot), while all other commercially available Rac1 antibodies cross-react with GTPases other than Rac1. Ln 1 - Rac1-6xHis, Ln 2- Rac2-6xHis, Ln 3 - Rac3-GST, Ln 4 - Cdc42-GST, Ln 5 - 50 µg platelet extract.

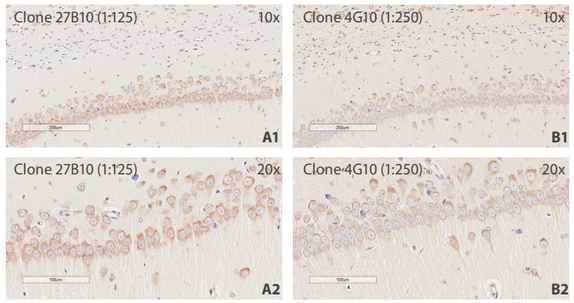
Small G-protein Antibodies

Small G-protein Antibodies	Host	Type	Species Reactivity	Cat. #	Amount
Cdc42 Specific Antibody Human Cdc42 Peptide	Mouse	mAb	Hu, Ms, Rt, other extracts	ACD03 ACD03-S	2 x 200 µl 1 x 50 µl
Rac1 Specific Antibody Human C-terminal Peptide	Mouse	mAb	Hu, Ms, Rt, other extracts	ARC03 ARC03-S	2 x 100 µl 1 x 50 µl
RhoA Specific Antibody Human RhoA Peptide	Mouse	mAb	Hu, Ms, Rt, other extracts	ARH04 ARH04-S	2 x 100 µl 1 x 50 µl

New Pathway Signaling Antibodies

In 2017 Cytoskeleton is expanding its offering of antibodies and reagents to study critical protein modifications. Reagents are available to study acetylation, tyrosyl phosphorylation, SUMOylation and ubiquitination. The products are rigorously QC'd and are particularly useful for enrichment studies of your protein of interest.

Immunohistochemical analysis of rat neuronal tissue: Anti-phosphotyrosine mAb 27B10 (Cat. # APY03) vs. 4G10



Anti-phosphotyrosine staining in rat neuronal tissue with Cytoskeleton's monoclonal antibody 27B10 (Cat. # APY03, A1, A2) vs. monoclonal antibody 4G10 (B1, B2). Proteinase K antigen retrieval used. Note the stronger and more specific anti-phosphotyrosine staining with Cytoskeleton's antibody 27B10 versus 4G10 antibody.

Pathway Signaling Antibodies

PTMtrue Antibody	Host	Type	Applications	Cat. #	Amount
Acetyl Lysine Antibody	Mouse	mAb	WB, IF, IP, ChIP	AAC01 AAC01-S	2 x 100 µl 1 x 25 µl
Phosphotyrosine Antibody	Mouse	mAb	WB, IP, IF, ELISA	APY03 APY03-S	2 x 100 µl 1 x 25 µl
Anti-Phosphotyrosine Affinity Beads	Mouse	mAb	IP	APY03-Beads	4 x 300 µl
Phosphotyrosine Antibody (HRP conjugate)	Mouse	mAb	WB	APY03-HRP APY03-HRP-S	1 x 100 µl 1 x 25 µl
NEW SUMO-2/3 Antibody (Clone 12F3)	Mouse	mAb	WB, IF, IP	ASM23 ASM23-S	2 x 100 µl 1 x 25 µl
NEW SUMO-2/3 Antibody (Clone 11G2)	Mouse	mAb	IF, IP	ASM24 ASM24-S	2 x 200 µl 1 x 150 µl
NEW SUMO-2/3 Affinity Beads	Mouse	mAb	IP	ASM24-Beads	2 x 400 µl
Ubiquitin Antibody	Mouse	mAb	WB, IF	AUB01 AUB01-S AUB01-XL	2 x 100 µl 1 x 25 µl 4 x 500 µl
Ubiquitin Affinity Beads (binds mono-/poly-ubiquitin tagged proteins)	n/a	n/a	IP	UBA01-beads	2 x 120 µl
NEW Control for Ippt IgG Beads	n/a	n/a	?	CIG01-beads	10 assays
NEW Control for Ubiquitin Affinity Beads	n/a	n/a	?	CUB01	10 assays

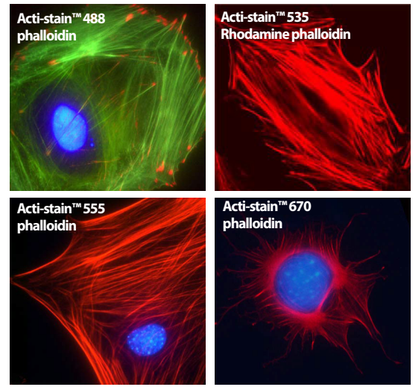
Cytoskeleton Protein Antibodies

Antibodies	Host	Type	Applications	Cat. #	Amount
Actin Antibody	Rabbit	pAb	WB, IP	AAN01-A AAN01-B	1 x 100 µg 3 x 100 µg
Tubulin Polyclonal Antibody	Sheep	pAb	WB, IF, ELISA	ATN02 ATN02-S	2 x 100 µl 1 x 25 µl
Cofilin Antibody	Rabbit	pAb	WB, IP, ICC	ACFL02-A ACFL02-B	1 x 50 µg 3 x 50 µg
Profilin Antibody	Rabbit	pAb	WB, ICC, ELISA, IP	APUF01-A	1 x 50 µg

Acti-stain™ Fluorescent Phalloidins and Spirochrome Live Cell Probes

The Acti-stain™ line of fluorescent phalloidins has been developed with an emphasis on creating exceptionally bright and stable probes for F-actin offered at an economical price. Side-by-side comparisons with similar products insure considerable savings without sacrificing quality when switching to an Acti-stain™ probe. The combination of in-house manufacturing, stringent quality control, and convenient packaging provides a great value. Give them a try and see for yourself.

For more information, citations and comparison to other fluorescent phalloidins, visit:
cytoskeleton.com/actin/acti-stain



Swiss 3T3 cells stained with Acti-stain™ Fluorescent Phalloidins

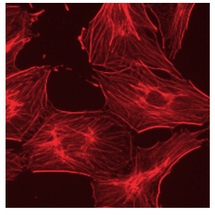
Product	Excitation	Emission	Signal stability * ($T_{1/2}$ in secs)	Cat. #	Amount**
Acti-stain™ 488 phalloidin	480 nm	535 nm	57	PHDG1-A	300 Slides
Acti-stain™ 535 phalloidin (Rhodamine phalloidin)	535 nm	585 nm	27	PHDR1	300 Slides
Acti-stain™ 555 phalloidin	535 nm	585 nm	46	PHDH1-A	300 Slides
Acti-stain™ 670 phalloidin	640 nm	670 nm	18	PHDN1-A	300 Slides
NEW **SiR700-Actin Kit** Includes SiR-Actin and Verapamil	690 nm	720 nm		CY-SC013	35 nmol
SiR-Actin Kit Includes SiR700-Actin and Verapamil	630 nm	680 nm		CY-SC001	50 nmol

* Stability measured without antifade. For comparison, fluorescein phalloidin has a T1/2 of 6 secs.
** One slide equals enough phalloidin to stain a 25 mm² coverslip

SiR-Actin is a trademarks of Spirochrome SA (Switzerland).

F-actin Visualization Biochem Kit™

Fix and permeabilize tissue culture cells while preserving structure of the F-actin cytoskeleton. Subsequently, the F-actin cytoskeleton is stained with fluorescent (rhodamine) phalloidin (Cat. # PHDR1) that is provided in the kit.

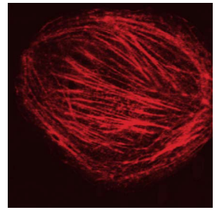


The F-actin cytoskeleton of Swiss 3T3 cells visualized with rhodamine phalloidin and using fixatives and cell permeabilizing reagents from the F-actin Visualization Biochem Kit™.

Product	Cat. #	Amount
F-actin Visualization Biochem Kit™	BK005	300 assays

Live Cell Actin Staining Products

In living cells, actin structures can be observed by incorporating fluorescently labeled actin, by expressing GFP-actin, or by expressing a fluorescently labeled actin binding protein sub-domain. Fluorescent actin is the most accurate reporter of actin structures.



Rhodamine-labeled actin microinjected into CHO cells. The labeled actin (Cat. # APHR) rapidly incorporates into the cellular actin cytoskeleton and allows real time observation of actin dynamics.

Labeled Actins	Source	Purity	Cat. #	Amount
Rhodamine Actin Protein	Human platelet, non-muscle	>99%	APHR-A APHR-C	4 x 10 µg 20 x 10 µg
Rhodamine Actin Protein	Rabbit skeletal muscle	>99%	AR05-B AR05-C	10 x 20 µg 20 x 20 µg
Spirochrome ™ SiR-Actin Kit	Chemical	>99%	CY-SC001	50 nmol

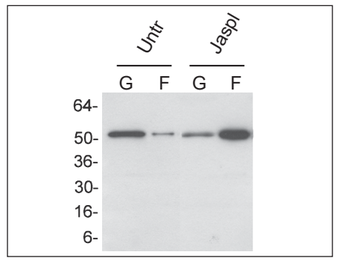
SiR-Actin is a trademarks of Spirochrome SA (Switzerland).

G-actin/F-actin *In Vivo* Assay Biochem Kit™

- Quantitates monomeric vs polymeric actin in cell/tissue lysates
- Reproducible and accurate method
- Contains all needed reagents

Lyse cells or tissue in the F-actin stabilizing buffer, preserving the G-actin:F-actin ratio. Centrifuge samples, separating supernatants (G-actin) and pellets (F-actin) which are then run on a gel for Western blot analysis.

Reorganization of actin after treatment with jasplakinolide



Swiss 3T3 cells were treated with jasplakinolide (Jaspl) or left untreated (Untr) and the G-actin (G) and F-actin (F) content was assayed using the G-actin/F-actin kit. Treatment with jasplakinolide resulted in a potent accumulation of F-actin.

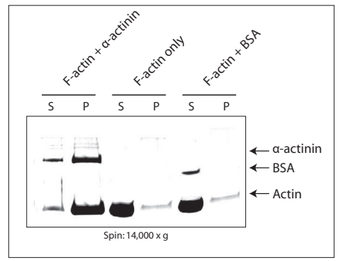
Product	Cat. #	Amount
G-actin/F-actin *In Vivo* Assay Biochem Kit™	BK037	30-100 assays
Protease Inhibitor Cocktail (100x solution)	PIC02	1 ml

Actin Binding Protein Spin-Down Assay Biochem Kit

- Identifies and characterizes Actin Binding Proteins (ABPs)
- Generation of saturation binding curves
- Muscle (BK001) or non-muscle (BK013) actin

This co-sedimentation assay will help you identify whether your ABP is a F-actin binding protein, a F-actin severing protein, has F-actin bundling activity, or is a G-actin binding protein.

Actin bundling assay using kit BK001



F-actin was incubated alone or together with α-actinin or BSA. Bundled F-actin was pelleted by a 14,000 x g centrifugation and pellets (P) and supernatants (S) were run on a SDS-PAGE gel. Only in the presence of the F-actin bundling protein α-actinin is actin pelleted at this centrifugation speed.

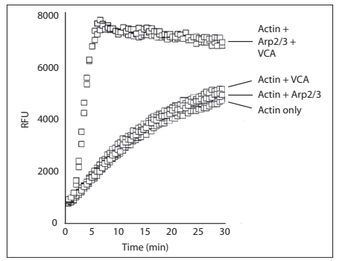
Product	Cat. #	Amount
Actin Binding Protein Spin-Down Assay Biochem Kit™ (skeletal muscle actin)	BK001	30-100 assays
Actin Binding Protein Spin-Down Assay Biochem Kit™ (non-muscle actin)	BK013	30-100 assays

Actin Polymerization Assay Biochem Kit™

- Utilizes fluorescent pyrene-actin
- F-actin polymerization and depolymerization
- Works with multiple sources of actin
- Valuable for characterizing ABPs

This kit is based upon the enhanced fluorescence of pyrene-conjugated actin that occurs during polymerization. Its versatility allows the study of the effects on polymerization (or depolymerization) of a compound, tissue extract, or protein of interest.

Characterization of ABPs using Actin Polymerization Biochem Kit™



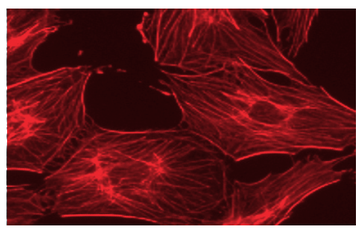
Effects of Arp2/3 (Cat. # RP01P) and the WASP VCA (Cat. # VCG03) domain on actin polymerization *in vitro*. Arp2/3 or the WASP VCA domain alone has little effect on the rate of actin polymerization, while the combination of the two leads to an activation of the actin nucleating Arp2/3 complex and a subsequent increased rate of actin polymerization.

Product	Cat. #	Amount
Actin Polymerization Assay Biochem Kit™	BK003	30-100 assays

F-actin Visualization Biochem Kit™

Fix and permeabilize tissue culture cells while preserving structure of the F-actin cytoskeleton. Subsequently, the F-actin cytoskeleton is stained with fluorescent (rhodamine) phalloidin (Cat. # PHDR1) that is also provided in the kit.

Product	Cat. #	Amount
F-actin Visualization Biochem Kit™	BK005	300 assays

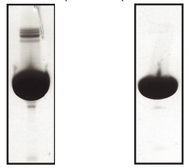


The F-actin cytoskeleton of Swiss 3T3 cells visualized with rhodamine phalloidin and using fixatives and cell permeabilizing reagents from the F-actin Visualization Biochem Kit™.

High Purity

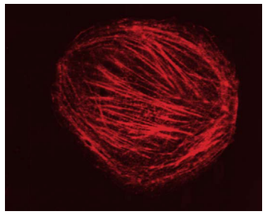
The highest purity actin available. Purities greater than 99% from most sources. Cited 100's of times in the literature.

>97% Pure (Cat. AKL95) **>99% Pure** (Cat. AKL99)



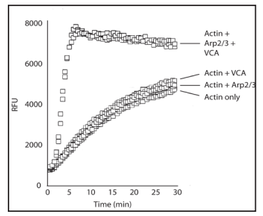
Labeled Actins

Highly pure, biologically active actins labeled with Rhodamine, Pyrene, and Biotin.



Biologically Active

Actin polymerization stimulated by Arp2/3 complex and the VCA domain of WASP measured with Pyrene Actin fluorescence (Cat.# AP05).



Unlabeled Actin Proteins

Unlabeled Actins	Source	Purity	Cat. #	Amount
Actin Protein	Rabbit skeletal muscle	>99%	AKL99-A AKL99-B AKL99-C AKL99-D AKL99-E	4 x 250 µg 2 x 1 mg 5 x 1 mg 10 x 1 mg 20 x 1 mg
Actin Protein	Rabbit skeletal muscle	>97%	AKL95-B AKL95-C	1 x 1 mg 5 x 1 mg
Actin Protein	Bovine cardiac muscle	>99%	AD99-A AD99-B	1 x 1 mg 5 x 1 mg
Actin Protein	Smooth muscle, chicken gizzard	>99%	AS99-A AS99-B	1 x 1 mg 5 x 1 mg
Actin Protein	Human platelet, non-muscle	>99%	APHL99-A APHL99-C APHL99-E	2 x 250 µg 1 x 1 mg 5 x 1 mg
Pre-formed Actin Filaments	Rabbit skeletal muscle	>99%	AKF99-A AKF99-B	1 x 1 mg 5 x 1 mg
Thin Filament (calcium sensitive tropomyosin /actin polymer	Bovine cardiac muscle	90%	CS-TFC01	1 x 1 mg
Ebashi Complex (complex of tromyosin/tropomodulin	Bovine cardiac muscle	70%	CS-TT05	1 x 1 mg

Labeled Actin Proteins

Labeled Actins	Source	Purity	Cat. #	Amount	Price ($)
Biotinylated Actin Protein	Rabbit skeletal muscle	>99%	AB07-A AB07-C	5 x 20 µg 20 x 20 µg	**285** **795**
Pyrene Actin Protein	Rabbit skeletal muscle	>99%	AP05-A AP05-B	1 x 1 mg 5 x 1 mg	**195** **785**
Rhodamine Actin Protein	Human platelet, non-muscle	>99%	APHR-A APHR-C	4 x 10 µg 20 x 10 µg	**265** **819**
Rhodamine Actin Protein	Rabbit skeletal muscle	>99%	AR05-B AR05-C	10 x 20 µg 20 x 20 µg	**263** **515**

Actin Antibodies

Antibodies	Antigen	Host	Grade	Cat. #	Amount
Actin Antibody	C-terminal of actin	Rabbit	Affinity Purified	AAN01-A AAN01-B	1 x 100 µg 3 x 100 µg
Cofilin Antibody	N-terminal of human cofilin1	Rabbit	Affinity Purified	ACFL02-A ACFL02-B	1 x 50 µg 3 x 50 µg
Profilin Antibody	Purified human profilin	Rabbit	Affinity Purified	APUF01-A	1 x 50 µg

Intermediate Filaments

Intermediate Filament	Source	Purity	Cat. #	Amount
Vimentin Protein Recombinant	Syrian Hamster	>90%	V01-A V01-C	2 x 50 µg 10 x 50 µg

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Actin Binding Proteins

Actin Binding Proteins	Source	Purity	Cat. #	Amount
***a*-Actinin Protein**	Rabbit skeletal muscle	>90%	AT01-A AT01-C	2 x 50 µg 10 x 50 µg
Arp2/3 Protein Complex	Porcine brain	>90%	RP01P-A RP01P-B	2 x 50 µg 6 x 50 µg
Cofilin Protein	Recombinant human cofilin 1	95%	CF01-A CF01-C	1 x 100 µg 4 x 100 µg
Gelsolin Protein	Recombinant human, plasma isoform	>95%	HPG6-A HPG6-B	4 x 20 µg 20 x 20 µg
Myosin II Cardiac Protein	Bovine cardiac muscle	95%	MY03-A MY03-B	5 x 1 mg 20 x 1 mg
S1 Myosin Protein	Rabbit skeletal muscle Chymotrypsin digest of Cat. # MY02 plus chromat	>90%	CS-MYS04	1 x 250 µg
S1 Myosin Protein	Bovine cardiac muscle Chymotrypsin digest of Cat. # MY03 plus chromat.	>90%	CS-MYS03	1 x 250 µg
Heavy Meromyosin Protein	Bovine cardiac muscle Chymotrypsin digest of Cat. # MY03 plus FPLC.	90%	CS-MH03	1 x 100 µg
Myosin II Protein	Rabbit skeletal muscle	95%	MY02-A MY02-B	5 x 1 mg 20 x 1 mg
Heavy Meromyosin Protein	Rabbit skeletal muscle Chymotrypsin digest of Cat. # MY02.	90%	MH01-A	4 x 50 µg
Profilin Protein	Recombinant human profilin 1	>95%	PR01-A	1 x 50 µg
WASP protein VCA Domain: GST tagged activates Arp2/3	Recombinant human	>95%	VCG03-A	1 x 500 µg

Labeled ECM Proteins

Labeled ECMs	Source	Purity	Cat. #	Amount
Fibronectin Red fluorescent, rhodamine	Bovine serum	>80%	FNR01-A FNR01-B	5 x 20 µg 20 x 20 µg
Fibronectin Green fluorescent, HiLyte Fluor™ 488	Bovine serum	>80%	FNR02-A FNR02-B	5 x 20 µg 20 x 20 µg
Fibronectin Biotinylated	Bovine serum	>80%	FNR03-A FNR03-B	5 x 20 µg 20 x 20 µg
Laminin Red fluorescent, rhodamine	Engelbreth-Holm-Swarm mouse tumor	>90%	LMN01-A LMN01-B	5 x 20 µg 20 x 20 µg
Laminin Green fluorescent, HiLyte Fluor™ 488	Engelbreth-Holm-Swarm mouse tumor	>90%	LMN02-A LMN02-B	5 x 20 µg 20 x 20 µg
Laminin Biotinylated	Engelbreth-Holm-Swarm mouse tumor	>90%	LMN03-A LMN03-B	5 x 20 µg 20 x 20 µg

HiLyte Fluor is a trademark of Anaspec, Inc. (CA).

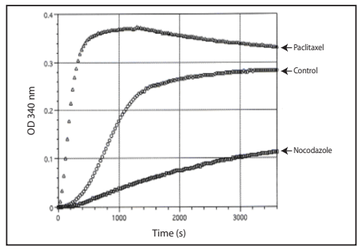
Actin Buffers

Actin Buffers	Cat. #	Amount
General Actin Buffer (10 ml or 100 ml when resuspended) For resuspending & diluting G-actin protein	BSA01-001 BSA01-010	1 x 10 ml 1 x 100 ml
Actin Polymerization Buffer (10X stock when resuspended) For the polymerization of actin	BSA02-001	1 x 2 ml
ATP (100 mM stock solution when resuspended) ATP is required for actin stability and polymerization	BSA04-001	1 x 1 ml

Tubulin Polymerization Assays

Tubulin polymerization assays are available in two formats: 1) the light scatter (also called absorbance or turbidometric) and 2) the fluorescence format based on the DAPI fluorophore. Both methods are sensitive to inhibitors and enhancers of polymerization. BK004P is an absorbance-based format used for hit or no hit screening results, whereas BK006P is for IC50 determinations which need more accuracy. BK011P, the fluorescent-based format, is used for screening and IC50s and is the most economical per assay.

Tubulin polymerization curves using Cat. # BK006P

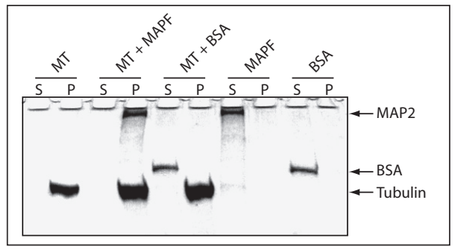


Product	Cat. #	Amount
Tubulin Polymerization Assay Biochem Kit™ Turbidometric-based, >99% pure tubulin	BK006P	24-30 assays
Tubulin Polymerization Assay Biochem Kit™ Turbidometric-based, >97% pure tubulin	BK004P	24-30 assays
Tubulin Polymerization Assay Biochem Kit™ Fluorescence-based, >99% pure tubulin	BK011P	96 assays

Tubulin Binding Assays

The Microtubule Binding Assay provides a robust method to identify and quantify how your test substance interacts with microtubules (see below). Biotinylated tubulin (Cat. # T333P) for use in subunit (heterodimer) binding assays is also available. See the SPA-based ligand competition assay described by Tahir et al. 2000 (Biotechniques, v29, pp156-160.).

Microtubule Binding Assay (Cat. # BK029) used to detect MAP binding to microtubules



Product	Cat. #	Amount
Tubulin (biotin labeled)	T333P-A T333P-B T333P-XL	5 x 20 µg 20 x 20 µg 1 x 500 µg
Microtubule Binding Protein Spin-Down Assay Biochem Kit™	BK029	30-100 assays

More Tubulin Biochem Kits™ & Antibodies

The Microtubule/Tubulin *In Vivo* Assay Kit measures the ratio of microtubules to tubulin in cell and tissue extracts. Samples are homogenized in lysis buffer, centrifuged, and then supernatant (tubulin) and pellet (microtubules) samples are run on a SDS-PAGE gel, blotted onto a membrane, and probed with anti-tubulin antibody. The tubulin antibody is ideal for dual and triple staining because the host animal is sheep, thus creating additional bandwidth for immunostaining.

Tubulin Biochem Kits™	Cat. #	Amount
Microtubule / Tubulin *In Vivo* Assay Biochem Kit™ Quantitates *in vivo* ratio of tubulin polymers & monomers	BK038	30-100 assays
Tubulin polyclonal antibody (host: sheep) Detects all species and isoforms of tubulin	ATN02 ATN02-S	2 x 100 µl 1 x 25 µl

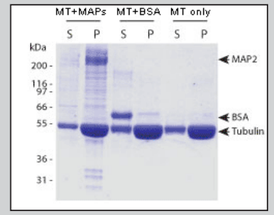
Specialized Tubulins For Pathogen Targeting

These specialized tubulins help exploit the diversity between host and pathogen tubulin isotypes. In combination with these proteins, micro-assays provide the most economical method of measuring drug interaction.

Products	Cat. #	Amount
Caki-1 Tumor Tubulin Protein	CS-TM001	1 x 250 µg
HeLa Cancer Cell Tubulin Protein (90% bI, 10% bIV isotypes)	CS-H001-B	1 x 250 µg
HeLa Cancer Cell Tubulin Protein (biotinylated) (90% bI, 10% bIV isotypes)	H003	1 x 40 µg
MCF-7 Cell Tubulin Protein (55% bI, 6% bIII, 39% bIV isotypes)	CS-H005	1 x 250 µg

Pre-formed Microtubules

- Substrate for discovery and characterization of microtubule binding proteins
- Determine IC50s for kinesin inhibitors
- Substrate for kinesin ATPases
- Ideal for HTS applications



MT binding spin-down assay using MT002. >80% of MT002 (arrow: Tubulin) is in pellet (P) after spin-down. MAPs bind to MTs and end up in pellet while BSA does not and stays in supernatant (S).

Unlabeled Tubulin Proteins

Unlabeled Proteins	Source	Purity	Cat. #	Amount
Tubulin Protein Lyophilized (no glycerol)	Porcine Brain	>99%	T240-A T240-B T240-C T240-DX	1 x 1 mg 5 x 1 mg 20 x 1 mg 1 x 10 mg
Tubulin Protein, MAP rich Lyophilized (no glycerol)	Porcine Brain	70% tubulin 30% MAPs	ML116-A ML116-B ML116-DX	1 x 1 mg 5 x 1 mg 1 x 10 mg
Tubulin for HTS Applications	Porcine Brain	97%	HTS03-A HTS03-B	1 x 4 mg 1 x 40 mg
Tubulin Protein Frozen (no glycerol)	Porcine Brain	>99%	T238P-A T238P-B T238P-C	1 x 1 mg 5 x 1 mg 20 x 1 mg
Microtubules pre-formed, lyophilized	Bovine brain	>99%	MT002-A MT002-XL	4 x 500 µg 1 x 10 mg
Caki-1 Tumor Tubulin Protein	Caki-1 Tumor Tissue	>90%	CS-TM001	1 x 250 µg
Cancer Cell Tubulin Protein	HeLa cells	>90%	CS-H001-B	1 x 250 µg
Cancer Cell Tubulin Protein	MCF-7 cells	>90%	CS-H005	1 x 250 µg

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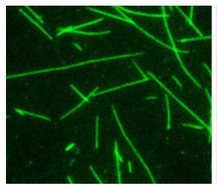
FtsZ Proteins

FtsZ Proteins	Source	Purity	Cat. #	Amount
FtsZ Protein	*S. aureus*, recombinant, 6xHis-tagged	>90%	FTZ02-A FTZ02-B	1 x 1 mg 5 x 1 mg
FtsZ Protein	*S. pneumoniae*, recombinant, 6xHis-tagged	>90%	FTZ03-A FTZ03-B	1 x 1 mg 5 x 1 mg
FtsZ Protein	*E. faecalis*, recombinant, 6xHis-tagged	>90%	FTZ04-A FTZ04-B	1 x 1 mg 5 x 1 mg
FtsZ Protein	*E. coli*, recombinant, 6xHis-tagged	>90%	FTZ05-A FTZ05-B	1 x 1 mg 5 x 1 mg

Tubulin Buffers, Reagents, & MAPs

Tubulin Buffers, Reagents, & MAPs	Cat. #	Amount
General Tubulin Buffer 10 ml or 100 ml when resuspended	BST01-001 BST01-010	1 x 10 ml 1 x 100 ml
GTP (100 mM stock when resuspended)	BST06-001 BST06-010	1 x 100 µl 10 x 100 µl
Tubulin Glycerol Buffer Enhances tubulin polymerization	BST05-001	1 x 10 ml
Microtubule-Associated Protein (MAP) Fraction Bovine brain MAP fraction, 70% MAP2	MAPF-A MAPF-C	1 x 100 µg 5 x 100 µg
Paclitaxel (2 mM stock when resuspended) Stabilizes microtubules	TXD01	10 x 100 µl
Tau Protein Bovine brain	TA01-A TA01-B	1 x 50 µg 3 x 50 µg

Labeled Tubulin Proteins



HiLyte Fluor™ 488 Labeled Tubulin - Cat. # TL488M

HiLyte Fluor is a trademark of Anaspec, Inc. (CA).

TRITC Rhodamine Labeled Tubulin - Cat. # TL590M

Labeled Tubulin Proteins	Ex / Em wavelength	$T_{1/2}$ of fluorescene (s)	Source	Purity	Cat. #	Amount
AMCA Labeled Tubulin	350 +/-20 nm 440 +/-20 nm	10	Porcine Brain	>99%	TL440M-A TL440M-B	5 x 20 µg 20 x 20 µg
HiLyte Fluor™ 488 Labeled Tubulin	460 +/-20 nm 520 +/-20 nm	300	Porcine Brain	>99%	TL488M-A TL488M-B	5 x 20 µg 20 x 20 µg
TRITC Rhodamine Labeled Tubulin	535 +/-20 nm 590 +/-20 nm	50	Porcine Brain	>99%	TL590M-A TL590M-B	5 x 20 µg 20 x 20 µg
X-Rhodamine Labeled Tubulin	560 +/- 20 nm 620 +/- 20 nm	70	Bovine Brain	>99%	TL620M-A TL620M-B	5 x 20 µg 20 x 20 µg
HiLyte Fluor™ 647 Labeled Tubulin	620 +/-20 nm 670 +/-20 nm	80	Porcine Brain	>99%	TL670M-A TL670M-B	5 x 20 µg 20 x 20 µg
Biotin Tubulin	na	na	Porcine Brain	>99%	T333P-A T333P-B T333P-XL	5 x 20 µg 20 x 20 µg 1 x 500 µg
Biotin Cancer Tubulin	na	na	HeLa cells	>90%	H003	1 x 40 µg

Kinesin & Dynein Proteins

Kinesin & Dynein Proteins	Source	Purity	Cat. #	Amount
CENP-E Motor Domain Protein	H. *sapiens*	>85%	CP01-A CP01-XL	2 x 25 µg 1 x 1 mg
Chromokinesin Motor Domain Protein	H. *sapiens*	>85%	CR01-A	2 x 25 µg
Dynein (cytoplasmic)	Porcine brain	>80%	CS-DN01	1 x 50 µg
Eg5 Motor Domain Protein	H. *sapiens*	>85%	EG01-A EG01-B EG01-XL	2 x 25 µg 10 x 25 µg 1 x 1 mg
Eg5 Homolog BimC Motor Domain Protein	A. *nidulans*	>85%	BM01-A	2 x 25 µg
Eg5 Homolog BimC Motor Domain Protein	A. *fumigatus*	>85%	EG02-A	2 x 15 µg
KIFC3 Motor Domain Protein	H. *sapiens*	>85%	KC01-A	2 x 25 µg
KIF3C Motor Domain Protein	H. *sapiens*	>85%	KF01-A	2 x 25 µg
KIF7 motor domain	H. *sapiens*	>85%	CS-KF51	1 x 100 µg
Kinesin Heavy Chain Motor Domain Protein	H. *sapiens*	>85%	KR01-A KR01-XL	2 x 25 µg 1 x 1 mg
MCAK Motor Domain Protein	H. *sapiens*	>85%	MK01-A	2 x 25 µg
MKLP1 Motor Domain Protein	H. *sapiens*	>85%	MP01-A MP01-XL	2 x 25 µg 1 x 1 mg
MKLP2 Motor Domain Protein	H. *sapiens*	>85%	CS-MP05	1 x 50 µg

Myosin & Thin Filament Proteins

Myosin Proteins	Source	Purity	Cat. #	Amount
NEW **Myosin S1 fragment (cardiac)**	Bovine	>85%	CS-MYS03	1 x 250 µg
NEW **Myosin S1 fragment (skeletal)**	Rabbit	>85%	CS-MYS04	1 x 250 µg
NEW **Myosin S1 fragment (smooth)**	Chicken	>85%	CS-MYS05	1 x 250 µg
Myosin II Skeletal Muscle Protein	Rabbit	>95%	MY02-A MY02-B	5 x 1 mg 20 x 1 mg
Myosin Cardiac Muscle Protein	Bovine	>95%	MY03-A MY03-B	5 x 1 mg 20 x 1 mg
Heavy Meromyosin Skeletal Muscle Protein	Rabbit	80%	MH01-A	4 x 50 µg
NEW **Heavy Meromyosin Cardiac Muscle Protein**	Bovine	80%	CS-MH03	1 x 100 µg
Pre-formed F-actin filaments	Rabbit	>99%	AKF99-A AKF99-B	1 x 1 mg 5 x 1 mg
NEW **Actin Thin Filaments (cardiac)** Calcium sensitive complex of F-actin, tropomyosin a/b & Troponin C,I,T	Bovine	>85%	CS-TFC01	1 x 1 mg
Tropomyosin / Troponin Complex Cardiac tropomyosin a/b & Troponin C,I,T	Bovine	>60%	CS-TT05	1 x 1 mg

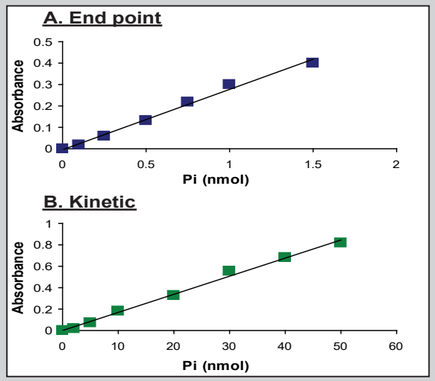
Pre-formed Microtubules & F-Actin Reagents

Microtubules and Other Reagents	Cat. #	Amount
NEW Thin Filament Protein (tropomyosin/tropomodulin and actin, calcium activated myosin ATPase	CS-TFC01	1 x 1 mg
Microtubules, Pre-formed, lyophilized, porcine source, substrate for kinesin ATPase assays	MT002-A MT002-XL	4 x 500 µg 1 x 10 mg
Microtubules, Pre-formed, lyophilized, bovine source	MT001-A MT001-XL	4 x 500 µg 1 x 10 mg
Actin Filaments, Pre-formed, lyophilized A ready to use substrate for myosin ATPase assays	AKF99-A AKF99-B	1 x 1 mg 5 x 1 mg
Paclitaxel (2 mM stock when resuspended) Stabilizes microtubules in motor assays	TXD01	10 x 100 µl

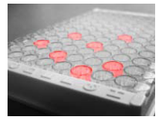
ATPase, GTPase, & Phosphatase Biochem Kits™

ATPases, GTPases, and other phosphatases liberate inorganic phosphate (Pi) from their respective triphosphate nucleotide or substrate. BK051-BK054 are suitable for HTS applications. BK051-BK054 and BK060 measure free phosphate via binding to a reporter dye or by enzymatic conversion into a reporter molecule. BK053 and BK054 are end-point assays suitable for measuring microtubule-induced kinesin ATPase or F-actin-induced myosin ATPase activity. BK051, BK052 and BK060 are kinetic assays, thus suitable for Vmax or Kcat determinations. These kits require a higher level activity ATPase or GTPase for sufficient sensitivity. BK060 is specialized for kinesins.

Phosphate Quantitation Biochem Kits™	Cat. #	Amount
ATPase ELIPA™ (enzyme-linked, colorimetric) Kinetic quantitation of ATP hydrolysis (Kcat 0.05 to >1.0)	BK051/052	96 assays
CytoPhos™ Phosphate Assay (endpoint assay) Colorimetric assay for ATPases & GTPases (Kcat 0.01 to >1.0)	BK054	1000 assays
GTPase ELIPA™ (enzyme-linked, colorimetric) Kinetic quantitation of GTP hydrolysis (Kcat 0.05 to >1.0)	BK051/052	96 assays
Kinesin ELIPA™ Biochem Kit For real time kinetic and Vmax kinesin ATPase measurements	BK060	96 assays
Kinesin ATPase Endpoint Assay For endpoint measurement of kinesin ATPase activity	BK053	1000 assays
Purine Nucleoside Phosphorylase protein Catalyzes the transfer of phosphate to MSEG reporter	ELP03	96 assays

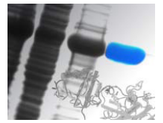


Comparison of standard curves of Cytoskeleton's endpoint (BK053 and BK054) and kinetic (BK051/52 and BK060) phosphate assays. Endpoint assays have a linear response between 0.1 and 1.5 nmol Pi. Kinetic assays give a linear response between 2 and 50 nmol Pi.



Compound Screening

Protein Purification



About Custom Services

Like our product offerings, the Custom Services department emphasizes quality products and services. We also understand ***accuracy*** and ***timeliness*** are critical elements for a successful project. The process starts with an experienced scientist asking for ***specifications and success factors*** for your project. Within 24 hours, the quotation will arrive and work can start at the next available schedule date. Regular updates are provided until project completion. Once complete, we continue support through timely citation-based advice and practical experience. Choose from over forty defined modules (full list is available online).

Combinations of Myosins and Actin Filaments

Cardiac S1 myosin / F-actin
Cardiac S1 myosin / Actin Thin Filament (calcium sensitive activation)
Cardiac Heavy Meromyosin / F-actin
Cardiac Heavy Meromyosin / Actin Thin Filament (calcium sens. actvn.)
Skeletal S1 myosin / F-actin
Skeletal S1 myosin / Actin Thin Filament (calcium sensitive activation)

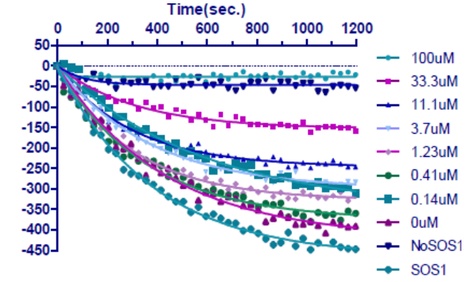
Inquire for other combinations not shown here, use the technical support e-mail tservice@cytoskeleton.com.

Combinations of Kinesin/Dyneins and Microtubules

Eg5 / microtubules
CenPE / microtubules
Kinesin HC / microtubules
Dynein / microtubules
MKLP1 / microtubules
MKLP2 / microtubules

Any motor from the opposite page or from your own labs can be configured in to the core screening format.

Measuring inhibition of SOS1-GEF-induced-GDP dissociation from K-Ras.



Legend: Inhibitors of GTP Exchange Factors (GEFs), which target small G-proteins such as Ras and Rho, are identified in a kinetic assay with either On or Off rates of GTP or GDP, or by measuring the equilibrium of the nucleotides to indicate a preference for GDP (inactive) or GTP (active).

Combinations of GEFs and Small G-Proteins

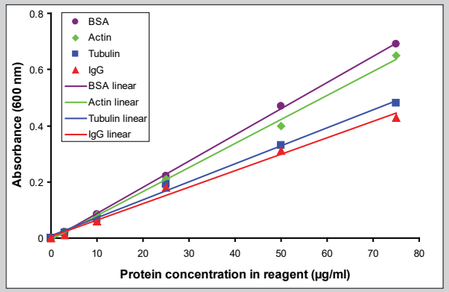
SOS1 / K-Ras4B
SOS1 / K-Ras4B G12V mutant
SOS1 / H-Ras
SOS1 / N-Ras
Tiam1 / Rac1
Tiam1 / Rac2
RasGRF1 / H-Ras
RasGRF1 / K-Ras4B
RasGRF1 / N-Ras
Dbs / RhoA
Dbs / Cdc42

Inquire for other combinations not shown here, use the technical support e-mail tservice@cytoskeleton.com.

For more information about Protein Purification Services please visit www.cytoskeleton.com/custom-services

Protein Assay Reagents

Since 1997, Cytoskeleton has been providing two standard protein assays that enable rapid, accurate, and detergent-compatible measurement of proteins in solution. Every batch is rigorously monitored and quality controlled for excellent batch to batch matching. The Advanced Protein Assay measures many different proteins with the same signal generation, and has very low detection capability (i.e. 1 µg/ml), whereas Precision Red is useful for measuring protein in cell extracts presented in detergent buffers.



Legend: ADV02 was used to measure four solutions of purified proteins, optical density was read at 600 nm. For both assays the color signal is developed in 1 min.

Used for measuring protein in:

- Cell Extracts in Detergent Buffers
- Purified Proteins and Antibodies
- High Protein Concentration Solutions
- Low Protein Concentration (ADV01 only)
- Serum Samples

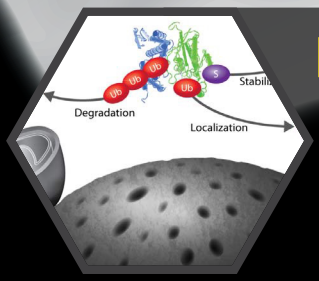
Detergent Compatibility

- Triton X-100
- NP40 or Igepal
- Tween 20
- SDS (ADV02 only)

- Color signal develops in 1 min
- OD is read at 600 nm with both reagents

Description	Cat. #	Amount
Advanced Protein Assay™ (5X stock reagent) Quantitates protein in the 0.025 - 1.0 mg/ml range	ADV01-A ADV01-B	1 x 500 ml 3 x 500 ml
Precision Red™ Advanced Protein Assay (1X stock reagent) Quantitates protein in the 0.25-50 mg/ml range	ADV02-A ADV02-B	1 x 500 ml 3 x 500 ml

International Customers
Locate your nearest distributor at:
cytoskeleton.com/distributors



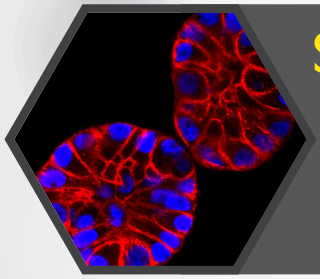
New Signal Seeker™ Kits and Antibodies

- Phosphotyrosine Enrichment Kit (p. 4-5)
- Mono-/Poly-Ubiquitin Enrichment Kit (p.4-5)
- SUMOylation Enrichment Kit (p.4-5)

GOBlot™ Western Blot Processor

- Save Time
- Save Money
- Save Antibody

Save 3 hours a day by utilizing the first affordable Western Blot Processor.



Spirochrome™ Live Cell Imaging Probes

- SiR-Tubulin Kit
- SiR-Actin Kit
- Cytoskeleton Kit
- SiR-Lysosome Kit
- SiR-DNA Kit
- NEW Far-red Probes

Contact Tebu-Bio to order Spirochrome™ products in Europe.

Email custcare@axilscientific.com or call us at 6775 7318 to enquire.

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