

# AlphaScreen *SureFire* Cellular Kinase Assays

For rapid detection of  
full-length activated  
kinases in cell lysates —  
even with difficult targets

Combining PerkinElmer's versatile and highly sensitive AlphaScreen® technology with TGR Biosciences' *SureFire*® cellular kinase assays provide an innovative solution for screening kinase activation in whole cells.

## HTS compatible

Whether you are running single plates or hundreds of plates, AlphaScreen *SureFire* assays will work for you. These two advanced technologies provide the ultimate homogeneous non-radiometric assay for full-length activated phosphorylated kinases in cell lysates. Easily miniaturizable, AlphaScreen *SureFire* is the choice for rapid and accurate high throughput screening.

## Flexible Homogeneous Assays

AlphaScreen *SureFire* cellular kinase assays can be used with many different types of cells including; primary cells, cells recombinantly overexpressing cloned targets as well as cells endogenously expressing kinases or the target receptor.

These assays can be used to screen for therapeutic modulators of kinases, GPCRs, inflammatory targets and oncology targets. Use the AlphaScreen *SureFire* range of cellular kinase assays to analyze important pathways which include; insulin receptor and other tyrosine kinase receptor signaling via the PI 3-kinase pathway, MAP kinase-mediated pathways, inflammatory receptor signaling via p38 MAPK or JNK phosphorylation, tyrosine kinase receptor signaling through the ras/raf pathway, and cytokine receptor activation through JAK/STAT phosphorylation.

[www.perkinelmer.com](http://www.perkinelmer.com)



 TGR BioSciences PTY LTD  
from biology to business

  
PerkinElmer®  
precisely.

# AlphaScreen SureFire —

**Homogeneous cellular assays for phosphorylated kinases:** no-wash, non-radiometric, miniaturizable cell-based functional HTS assays for activated kinases.

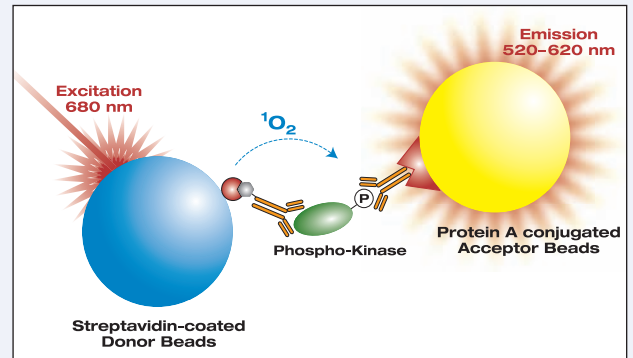
**Suitable for over-expressed and endogenous receptors:** extremely sensitive assay that can be used with cells over-expressing cloned targets or with cells expressing physiologically relevant endogenous levels of kinase or receptor target.

**Screen for small or large molecule therapeutics:** suitable for monitoring target modulation by monoclonal antibodies and small molecule inhibitors.

**Use in many types of cells, including primary cells:** flexible assay format and application use.

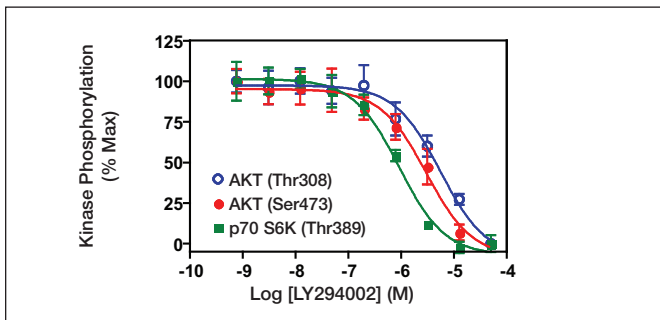
**Easy to automate with excellent Z' values:** utilizes HTS-proven AlphaScreen technology, which is easily automated and scalable to 384- and 1536-well microplate formats, while yielding outstanding S/Bs and Z' values.

## AlphaScreen SureFire Kinase Assay Principle



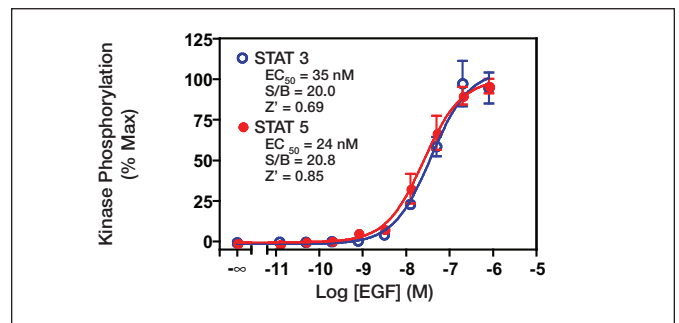
**AlphaScreen SureFire Kinase Assay Principle.** Detection of the activated kinase is enabled by immuno-sandwich capture of endogenous phosphorylated kinase in cell lysates. Antibody-coated AlphaScreen beads generate a highly amplified signal when in close proximity due to the binding of the phospho-kinase.

### Inhibition of PI3K Pathway Kinases by LY294002



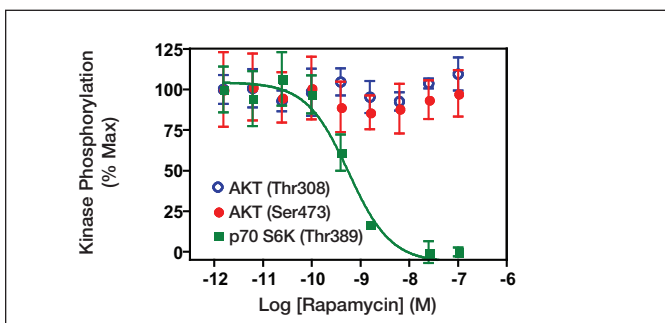
Confluent mouse NIH-3T3 cells were serum starved ON and stimulated with PDGF for 10 minutes in the presence of various concentrations of LY294002 inhibitor. Phosphorylation of AKT (Ser473), AKT (Thr308) and p70 S6K (Thr389) kinases were measured using AlphaScreen SureFire Phospho-kinase and AlphaScreen Protein A detection reagents on an EnVision Alpha plate reader.

### JAK/STAT Pathway Analysis STAT 3 & 5 Activation in A431 Cells



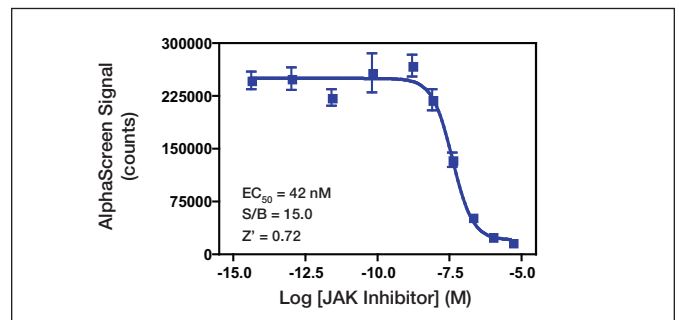
Human A431 cells were serum starved ON and stimulated with various concentrations of EGF. Phosphorylation of STAT 3 (Tyr705) and STAT 5 (Tyr694/Tyr699) kinases were measured using AlphaScreen SureFire Phospho-kinase and AlphaScreen Protein A detection reagents on an EnVision Alpha plate reader.

### Inhibition of PI3K Pathway Kinases by Rapamycin



Confluent mouse NIH-3T3 cells were serum starved ON and stimulated with PDGF for 10 minutes in the presence of various concentrations of rapamycin inhibitor. Phosphorylation of AKT (Ser473), AKT (Thr308) and p70 S6K (Thr389) kinases were measured using AlphaScreen SureFire Phospho-kinase and AlphaScreen Protein A detection reagents on an EnVision Alpha plate reader.

### Inhibition of Constitutive STAT 5 Phosphorylation in HEL Cells



Human HEL cells were serum starved ON and treated with various concentrations of JAK inhibitor. Phosphorylation of STAT 5 (Tyr694/Tyr699) kinase was measured using AlphaScreen SureFire Phospho-STAT 5 (Tyr694/Tyr699) and AlphaScreen Protein A detection reagents on an EnVision Alpha plate reader.

# kinase pathway analysis made easy

## AlphaScreen Technology — the ideal choice for accurately and economically screening a broad range of targets fast

The AlphaScreen *SureFire* cellular kinase assays are just a few of the many useful screening tools built on PerkinElmer's versatile AlphaScreen technology. AlphaScreen is a homogeneous, bead-based platform widely used in assay development and high throughput screening. The long excitation wavelength (680 nm) combined with a shorter emission wavelength (520 - 620 nm) reduces interference from biological samples or assay components, ensuring a very low background.

Unlike other proximity-based technologies, AlphaScreen beads can be up to 200 nm apart, allowing for the detection of very simple to very complex biological interactions, and substrates of many different sizes. **The result, high sensitivity at low attomole levels with a simple to set up and optimize, rapid assay.**

## Ordering Information

### AlphaScreen *SureFire* Kits

Product	Product No.	Size
<i>SureFire</i> Phospho-ERK 1/2 Assay Kits	TGRES500	500 assay points
	TGRES10K	10,000 assay points
	TGRES50K	50,000 assay points
<i>SureFire</i> Total-ERK Assay Kits	TGRTE500	500 assay points
	TGRTE10K	10,000 assay points
	TGRTE50K	50,000 assay points
<i>SureFire</i> Phospho-AKT (Ser473) Assay Kit	TGRAS500	500 assay points
	TGRAS10K	10,000 assay points
	TGRAS50K	50,000 assay points
<i>SureFire</i> Phospho-AKT (Thr308) Assay Kit	TGRA2S500	500 assay points
	TGRA2S10K	10,000 assay points
	TGRA2S50K	50,000 assay points
<i>SureFire</i> Phospho-p38 MAPK Assay Kit	TGR38S500	500 assay points
	TGR38S10K	10,000 assay points
	TGR38S50K	50,000 assay points
<i>SureFire</i> Phospho-p70 S6K (Thr389) Assay Kit	TGR70S500	500 assay points
	TGR70S10K	10,000 assay points
	TGR70S50K	50,000 assay points
<i>SureFire</i> Phospho-p70 S6K (Thr421/Ser424) Assay Kit	TGR702S500	500 assay points
	TGR702S10K	10,000 assay points
	TGR702S50K	50,000 assay points
<i>SureFire</i> Phospho-p70 S6K (Thr229) Assay Kit	TGR703S500	500 assay points
	TGR703S10K	10,000 assay points
	TGR703S50K	50,000 assay points
<i>SureFire</i> Phospho-PDK 1 (Ser241) Assay Kit	TGRPS500	500 assay points
	TGRPS10K	10,000 assay points
	TGRPS250K	50,000 assay points

## Optimized performance — use reagents with PerkinElmer instruments and plates

EnVision® Multilabel Microplate Reader. Get the speed and capacity your assay development or HTS lab needs with the EnVision Multilabel Plate Reader from PerkinElmer. Our fast application-oriented EnVision reader handles all non-isotopic detection technologies. The EnVision is optimized for our robust and sensitive AlphaScreen technology. Choose the AlphaScreen HTS Turbo option module for high throughput screening. Visit [www.perkinelmer.com/platereaders](http://www.perkinelmer.com/platereaders) to view our free interactive EnVision tutorial.

Better microplates mean better results. High quality microplates manufactured by the instrument and assay expert will give you better results, time after time.

You'll find the type, size and color you need for any assay application, detection technology and any plate reader. Don't waste time with general labware plates, choose plates developed and tested with your application in mind. Never forget – better plates, give better performance and better results. For up-to-date information on microplates visit [www.perkinelmer.com.microplates](http://www.perkinelmer.com.microplates) today.

Product	Product No.	Size
<i>SureFire</i> Phospho-MEK 1 Assay Kit	TGRMS500	500 assay points
	TGRMS10K	10,000 assay points
	TGRMS50K	50,000 assay points
<i>SureFire</i> Phospho-JNK Assay Kit	TGRJS500	500 assay points
	TGRJS10K	10,000 assay points
	TGRJS50K	50,000 assay points
<i>SureFire</i> Phospho-STAT 3 (Tyr705) Assay Kit	TGRS3S500	500 assay points
	TGRS3S10K	10,000 assay points
	TGRS3S50K	50,000 assay points
<i>SureFire</i> Phospho-STAT 4 (Tyr693) Assay Kit <i>Coming soon</i>	TGRS4S500	500 assay points
	TGRS4S10K	10,000 assay points
	TGRS4S50K	50,000 assay points
<i>SureFire</i> Phospho-STAT 5 (Tyr694/Tyr699) Assay Kit	TGRS5S500	500 assay points
	TGRS5S10K	10,000 assay points
	TGRS5S50K	50,000 assay points
<i>SureFire</i> Phospho-4EBP 1 (Thr70) Assay Kit	TGR4E2S500	500 assay points
	TGR4E2S10K	10,000 assay points
	TGR4E2S50K	50,000 assay points
<i>SureFire</i> Phospho-4EBP 1 (Thr37/Thr46) Assay Kit	TGR4ES500	500 assay points
	TGR4ES10K	10,000 assay points
	TGR4ES50K	50,000 assay points
<i>SureFire</i> Phospho-SMAD 2 (Ser465/Ser467) Assay Kit	TGRSM2S500	500 assay points
	TGRSM2S10K	10,000 assay points
	TGRSM2S50K	50,000 assay points
<i>SureFire</i> Phospho-GSK 3 $\alpha$ (Ser21) Assay Kit	TGRGAS500	500 assay points
	TGRGAS10K	10,000 assay points
	TGRGAS50K	50,000 assay points

## Complementary Products

### Microplates

Product	Product No.	Size
<i>SureFire</i> Phospho-GSK 3 $\alpha$ (Ser9) Assay Kit	TGRGBS500	500 assay points
	TGRGBS10K	10,000 assay points
	TGRGBS50K	50,000 assay points
<i>SureFire</i> Phospho-BAD (Ser112) Assay Kit	TGRBS500	500 assay points
	TGRBS10K	10,000 assay points
	TGRBS50K	50,000 assay points
<i>SureFire</i> Phospho-BAD (Ser136) Assay Kit	TGRB2S500	500 assay points
	TGRB2S10K	10,000 assay points
	TGRB2S50K	50,000 assay points
<i>SureFire</i> Phospho-NF $\kappa$ B p65 (Ser536) Assay Kit <i>Coming soon</i>	TGRNFS500	500 assay points
	TGRNFS10K	10,000 assay points
	TGRNFS50K	50,000 assay points
<i>SureFire</i> Phospho-S6 RP (Ser235/Ser236) Assay Kit	TGRS6PS500	500 assay points
	TGRS6PS10K	10,000 assay points
	TGRS6PS50K	50,000 assay points
<i>SureFire</i> Phospho-S6 RP (Ser240/Ser244) Assay Kit	TGRS6P2S500	500 assay points
	TGRS6P2S10K	10,000 assay points
	TGRS6P2S50K	50,000 assay points
<i>SureFire</i> Phospho-Caspase 9 (Ser196) Assay Kit <i>Coming soon</i>	TGRC9S500	500 assay points
	TGRC9S10K	10,000 assay points
	TGRC9S50K	50,000 assay points
AlphaScreen Protein A Kit	6760617C	500 assay points
	6760617M	10,000 assay points
	6760617R	50,000 assay points

*Note: Both SureFire kinase kits and AlphaScreen protein A kits are required in equal quantities to run the SureFire cellular kinase assays. One assay point provides sufficient reagents for a single well in standard 384-well microplate format*

Product	Product No.	Size
AlphaPlate™-1536, light gray, 1536-well	6004350	50
	6004859	200
AlphaPlate-384, light gray, 384-well	6008350	50
	6008359	200
OptiPlate™-24, white, 24-well	6005186	100
OptiPlate-96, white, 96-well	6005291	5
	6005290	50
	6005299	200
OptiPlate-384, white, 384-well	6007291	5
	6007290	50
	6007299	200
OptiPlate-1536, white opaque, 1536-well	6004291	5
	6004290	50
	6004299	200
OptiPlate-96 F, black, 96-well	6005271	5
	6005270	50
	6005279	200
½ Area ProxiPlate™-96, white, half-area 96-well	6005541	5
	6005540	50
	6005549	200
ProxiPlate-96, white, shallow 96-well	6006291	5
	6006290	50
	6006299	200
ProxiPlate-384 Plus, white, shallow 384-well	6008281	5
	6008280	50
	6008289	200

### AlphaScreen Optimization Tools

AlphaScreen Omnibeads® are used for optimizing instrument set-up and in assay development. AlphaScreen TruHits® beads are ideal for counter screening and identifying false positives.

Product	Product No.	Size
AlphaScreen Omnibeads	6760626D	1,000 assay points
	6760626M	10,000 assay points
	6760626R	50,000 assay points
AlphaScreen TruHits Kit	6760627D	1,000 assay points
	6760627M	10,000 assay points

For more information about *SureFire* technology, visit [www.tgr-biosciences.com.au](http://www.tgr-biosciences.com.au).

## Get the *SureFire* answer for your cellular functional kinase screens

HTS functional cell-based kinase assays are ready for your order!

For more information about AlphaScreen *SureFire* visit

[www.perkinelmer.com/surefire](http://www.perkinelmer.com/surefire) today!

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