



Z'-LYTE™ Screening Protocol and Assay Conditions

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|                            |    |                                |    |                           |    |                      |    |
|----------------------------|----|--------------------------------|----|---------------------------|----|----------------------|----|
| ABL1                       | 7  | DYRK1B                         | 13 | KIT V559D V654A           | 19 | PLK1                 | 25 |
| ABL1 E255K                 | 7  | DYRK3                          | 13 | KIT V560G                 | 19 | PLK2                 | 26 |
| ABL1 F317I                 | 7  | DYRK4                          | 13 | KSR2                      | 19 | PLK3                 | 26 |
| ABL1 F317L                 | 7  | EEF2K                          | 13 | LCK                       | 20 | PRKACA (PKA)         | 26 |
| ABL1 G250E                 | 7  | EGFR (ErbB1)                   | 13 | LTK (TYK1)                | 20 | PRKCA (PKC alpha)    | 26 |
| ABL1 T315I                 | 7  | EGFR (ErbB1) C797S             | 13 | LYN A                     | 20 | PRKCB1 (PKC beta I)  | 26 |
| ABL1 Y253F                 | 7  | EGFR (ErbB1) G719C             | 13 | LYN B                     | 20 | PRKCB2 (PKC beta II) | 26 |
| ABL2 (Arg)                 | 7  | EGFR (ErbB1) G719S             | 14 | MAP3K19 (YSK4)            | 20 | PRKCD (PKC delta)    | 26 |
| ACVR1B (ALK4)              | 7  | EGFR (ErbB1) L858R             | 14 | MAP3K9 (MLK1)             | 20 | PRKCE (PKC epsilon)  | 26 |
| ADRBK1 (GRK2)              | 7  | EGFR (ErbB1) L861Q             | 14 | MAP4K2 (GCK)              | 20 | PRKCG (PKC gamma)    | 26 |
| ADRBK2 (GRK3)              | 7  | EGFR (ErbB1) T790M             | 14 | MAP4K4 (HGK)              | 20 | PRKCH (PKC eta)      | 26 |
| AKT1 (PKB alpha)           | 8  | EGFR (ErbB1) T790M C797S L858R | 14 | MAP4K5 (KHS1)             | 20 | PRKCI (PKC iota)     | 27 |
| AKT2 (PKB beta)            | 8  | EGFR (ErbB1) T790M L858R       | 14 | MAPK1 (ERK2)              | 20 | PRKCN (PKC delta)    | 27 |
| AKT3 (PKB gamma)           | 8  | EPHA1                          | 14 | MAPK11 (p38 beta)         | 20 | PRKCC (PKC theta)    | 27 |
| ALK                        | 8  | EPHA2                          | 14 | MAPK12 (p38 gamma)        | 21 | PRKCZ (PKC zeta)     | 27 |
| AMPK (A1/B2/G2)            | 8  | EPHA4                          | 14 | MAPK13 (p38 delta)        | 21 | PRKD1 (PKC mu)       | 27 |
| AMPK (A1/B2/G3)            | 8  | EPHA5                          | 14 | MAPK14 (p38 alpha) Direct | 21 | PRKD2 (PKD2)         | 27 |
| AMPK (A2/B1/G2)            | 8  | EPHA8                          | 15 | MAPK3 (ERK1)              | 21 | PRKG1                | 27 |
| AMPK (A2/B1/G3)            | 8  | EPHB1                          | 15 | MAPK7 (ERK5)              | 21 | PRKG2 (PKG2)         | 27 |
| AMPK (A2/B2/G3)            | 8  | EPHB2                          | 15 | MAPKAPK2                  | 21 | PRKX                 | 27 |
| AMPK A1/B1/G1              | 8  | EPHB3                          | 15 | MAPKAPK3                  | 21 | PTK2 (FAK)           | 27 |
| AMPK A2/B1/G1              | 8  | EPHB4                          | 15 | MAPKAPK5 (PRAK)           | 21 | PTK2B (FAK2)         | 27 |
| AURKA (Aurora A)           | 9  | ERBB2 (HER2)                   | 15 | MARK1 (MARK)              | 21 | PTKG (Brk)           | 28 |
| AURKB (Aurora B)           | 9  | ERBB4 (HER4)                   | 15 | MARK2                     | 21 | RET                  | 28 |
| AURKC (Aurora C)           | 9  | FER                            | 15 | MARK3                     | 21 | RET A883F            | 28 |
| AXL                        | 9  | FES (FPS)                      | 15 | MARK4                     | 22 | RET S891A            | 28 |
| BLK                        | 9  | FGFR1                          | 15 | MATK (HYL)                | 22 | RET V804E            | 28 |
| BMX                        | 9  | FGFR2                          | 15 | MELK                      | 22 | RET V804L            | 28 |
| BRSK1 (SAD1)               | 9  | FGFR2 N549H                    | 16 | MERTK (cMER)              | 22 | RET Y791F            | 28 |
| BTK                        | 9  | FGFR3                          | 16 | MET (cMet)                | 22 | ROCK1                | 28 |
| CAMK1D (CaMKI delta)       | 9  | FGFR3 K650E                    | 16 | MET (cMet) Y1235D         | 22 | ROCK2                | 28 |
| CAMK1G (CaMKI gamma)       | 9  | FGFR3 V555M                    | 16 | MET M1250T                | 22 | ROS1                 | 28 |
| CAMK2A (CaMKII alpha)      | 9  | FGFR4                          | 16 | MINK1                     | 22 | RPS6KA1 (RSK1)       | 28 |
| CAMK2B (CaMKII beta)       | 10 | FGR                            | 16 | MKNK1 (MNK1)              | 22 | RPS6KA2 (RSK3)       | 29 |
| CAMK2D (CaMKII delta)      | 10 | FLT1 (VEGFR1)                  | 16 | MST1R (RON)               | 22 | RPS6KA3 (RSK2)       | 29 |
| CAMK4 (CaMKIV)             | 10 | FLT3                           | 16 | MST4                      | 22 | RPS6KA4 (MSK2)       | 29 |
| CDC42 BPA (MRCKA)          | 10 | FLT3 D835Y                     | 16 | MUSK                      | 23 | RPS6KA5 (MSK1)       | 29 |
| CDC42 BPB (MRCKB)          | 10 | FLT4 (VEGFR3)                  | 16 | MYLK2 (skMLCK)            | 23 | RPS6KA6 (RSK4)       | 29 |
| CDC42 BPG (MRCKG)          | 10 | FRAP1 (mTOR)                   | 17 | NEK1                      | 23 | RPS6KB1 (p70S6K)     | 29 |
| CDK1/cyclin B              | 10 | FRK (PTK5)                     | 17 | NEK2                      | 23 | RPS6KB2 (p70S6Kb)    | 29 |
| CDK17/cyclin Y             | 10 | FYN                            | 17 | NEK4                      | 23 | SBK1                 | 29 |
| CDK18/cyclin Y             | 10 | GRK4                           | 17 | NEK6                      | 23 | SGK (SGK1)           | 29 |
| CDK2/cyclin A              | 10 | GRK5                           | 17 | NEK7                      | 23 | SGK2                 | 29 |
| CDK5/p25                   | 10 | GRK6                           | 17 | NEK9                      | 23 | SGKL (SGK3)          | 29 |
| CDK5/p35                   | 11 | GRK7                           | 17 | NIM1K                     | 23 | SNF1LK2              | 30 |
| CDKL5                      | 11 | GSK3A (GSK3 alpha)             | 17 | NTRK1 (TRKA)              | 23 | SRC                  | 30 |
| CHEK1 (CHK1)               | 11 | GSK3B (GSK3 beta)              | 17 | NTRK2 (TRKB)              | 23 | SRC N1               | 30 |
| CHEK2 (CHK2)               | 11 | HCK                            | 17 | NTRK3 (TRKC)              | 24 | SRMS (Srm)           | 30 |
| CLK1                       | 11 | HIPK1 (Myak)                   | 17 | PAK1                      | 24 | SRPK1                | 30 |
| CLK2                       | 11 | HIPK2                          | 18 | PAK2 (PAK65)              | 24 | SRPK2                | 30 |
| CLK3                       | 11 | HIPK3 (YAK1)                   | 18 | PAK3                      | 24 | STK22B (TSSK2)       | 30 |
| CSF1R (FMS)                | 11 | HIPK4                          | 18 | PAK4                      | 24 | STK22D (TSSK1)       | 30 |
| CSK                        | 11 | IGF1R                          | 18 | PAK6                      | 24 | STK23 (MSSK1)        | 30 |
| CSNK1A1 (CK1 alpha 1)      | 11 | IKBKB (IKK beta)               | 18 | PAK7 (KIAA1264)           | 24 | STK24 (MST3)         | 30 |
| CSNK1A1L                   | 11 | IKBKE (IKK epsilon)            | 18 | PASK                      | 24 | STK25 (YSK1)         | 30 |
| CSNK1D (CK1 delta)         | 12 | INSR                           | 18 | PDGFRA (PDGFR alpha)      | 24 | STK3 (MST2)          | 31 |
| CSNK1E (CK1 epsilon)       | 12 | INSRR (IRR)                    | 18 | PDGFRA D842V              | 24 | STK4 (MST1)          | 31 |
| CSNK1E (CK1 epsilon) R178C | 12 | IRAK4                          | 18 | PDGFRA T674I              | 24 | SYK                  | 31 |
| CSNK1G1 (CK1 gamma 1)      | 12 | ITK                            | 18 | PDGFRA V561D              | 25 | TAO2 (TAO1)          | 31 |
| CSNK1G2 (CK1 gamma 2)      | 12 | JAK1                           | 18 | PDGFRB (PDGFR beta)       | 25 | TBK1                 | 31 |
| CSNK1G3 (CK1 gamma 3)      | 12 | JAK2                           | 19 | PDK1 Direct               | 25 | TEK (Tie2)           | 31 |
| CSNK2A1 (CK2 alpha 1)      | 12 | JAK2 JH1 JH2                   | 19 | PEAK1                     | 25 | TEK (Tie2) Y897S     | 31 |
| CSNK2A2 (CK2 alpha 2)      | 12 | JAK2 JH1 JH2 V617F             | 19 | PHKG1                     | 25 | TNK1                 | 31 |
| DAPK3 (ZIPK)               | 12 | JAK3                           | 19 | PHKG2                     | 25 | TKX                  | 31 |
| DCAMKL1 (DCLK1)            | 12 | KDR (VEGFR2)                   | 19 | PIM1                      | 25 | TYK2                 | 31 |
| DCAMKL2 (DCK2)             | 13 | KIT                            | 19 | PIM2                      | 25 | TYRO3 (RSE)          | 31 |
| DNA-PK                     | 13 | KIT T670I                      | 19 | PIM3                      | 25 | YES1                 | 32 |
| DYRK1A                     | 13 | KIT V559D                      | 19 | PKN1 (PRK1)               | 25 | ZAP70                | 32 |

**KINASE-SPECIFIC ASSAY CONDITIONS - CASCADE FORMAT \_\_\_\_\_ 33**

|               |    |               |    |                    |    |                         |    |
|---------------|----|---------------|----|--------------------|----|-------------------------|----|
| BRAF          | 33 | MAP2K2 (MEK2) | 33 | MAPK10 (JNK3)      | 33 | MAPK9 (JNK2)            | 34 |
| BRAF V599E    | 33 | MAP2K6 (MKK6) | 33 | MAPK14 (p38 alpha) | 33 | PKD1                    | 34 |
| MAP2K1 (MEK1) | 33 | MAP3K8 (COT)  | 33 | MAPK8 (JNK1)       | 33 | RAF1 (cRAF) Y340D Y341D | 34 |

**TABLE OF KINASE ATP KM BINS AND INHIBITOR VALIDATION \_\_\_\_\_ 35**

## Assay Theory

The Z'-LYTE biochemical assay employs a fluorescence-based, coupled-enzyme format and is based on the differential sensitivity of phosphorylated and non-phosphorylated peptides to proteolytic cleavage (Figure 1). The peptide substrate is labeled with two fluorophores—one at each end—that make up a FRET pair.

In the primary reaction, the kinase transfers the gamma-phosphate of ATP to a single tyrosine, serine or threonine residue in a synthetic FRET-peptide. In the secondary reaction, a site-specific protease recognizes and cleaves non-phosphorylated FRET-peptides. Phosphorylation of FRET-peptides suppresses cleavage by the Development Reagent. Cleavage disrupts FRET between the donor (i.e., coumarin) and acceptor (i.e., fluorescein) fluorophores on the FRET-peptide, whereas uncleaved, phosphorylated FRET-peptides maintain FRET. A ratiometric method, which calculates the ratio (the Emission Ratio) of donor emission to acceptor emission after excitation of the donor fluorophore at 400 nm, is used to quantitate reaction progress, as shown in the equation below.

$$\text{Emission Ratio} = \frac{\text{Coumarin Emission (445 nm)}}{\text{Fluorescein Emission (520 nm)}}$$

A significant benefit of this ratiometric method for quantitating reaction progress is the elimination of well-to-well variations in FRET-peptide concentration and signal intensities. As a result, the assay yields very high Z'-factor values (>0.7) at a low percent phosphorylation.

Both cleaved and uncleaved FRET-peptides contribute to the fluorescence signals and therefore to the Emission Ratio. The extent of phosphorylation of the FRET-peptide can be calculated from the Emission Ratio. The Emission Ratio will remain low if the FRET-peptide is phosphorylated (i.e., no kinase inhibition) and will be high if the FRET-peptide is non-phosphorylated (i.e., kinase inhibition).

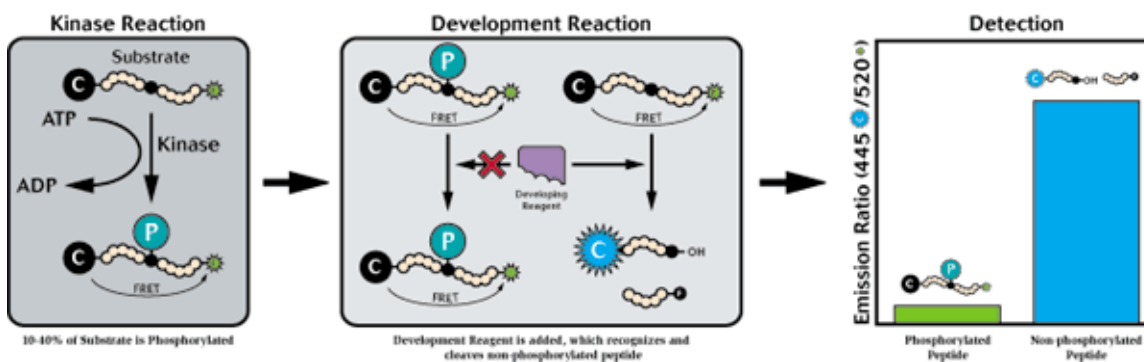


Figure 1: Z'-LYTE Illustration

## Z'-LYTE Assay Conditions

### Test Compounds

The Test Compounds are screened in 1% DMSO (final) in the well. For 10 point titrations, 3-fold serial dilutions are conducted from the starting concentration of the customer's choosing.

### Peptide/Kinase Mixtures

All Peptide/Kinase Mixtures are diluted to a 2X working concentration in the appropriate Kinase Buffer (see section *Kinase Specific Assay Conditions* for a complete description).

### ATP Solution

All ATP Solutions are diluted to a 4X working concentration in Kinase Buffer (50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA).

ATP Km apparent is previously determined using a Z'-LYTE assay.

### Development Reagent Solution

The Development Reagent is diluted in Development Buffer (see section *Kinase-Specific Assay Conditions - Direct and Cascade* for a complete description).

**10X Novel PKC Lipid Mix:** 2 mg/ml Phosphatidyl Serine, 0.2 mg/ml DAG in 20 mM HEPES, pH 7.4, 0.3% CHAPS

#### For 5 mL 10X Novel PKC Lipid Mix:

1. Add 10 mgs Phosphatidyl Serine (Avanti Polar Lipids Part# 8400032C or 840039C) and 1 mg DAG (Avanti Polar Lipids Part# 800811C) to a glass tube.
2. Remove the chloroform from lipid mixture by evaporating to a clear, thin film under a stream of nitrogen. Continuous rotation of the tube, at an angle to ensure maximum surface area of the lipid solution, will promote the thinnest film.
3. Add 5 mLs resuspension buffer, 20 mM HEPES, 0.3% CHAPS, pH 7.4, to the dried lipid mix
4. Heat gently to 50-60 °C for 1-2 minutes and vortex in short intervals until the lipids are dissolved to a clear or slightly hazy solution. The lipids are typically in solution after 2-3 heat/vortex cycles. .
5. Cool to room temperature, aliquot into single use volumes and store at -20 °C.

### Assay Protocol

*Bar-coded Corning, low volume NBS, black 384-well plate (Corning Cat. #4514)*

1. 100 nL – 100X Test Compound in 100% DMSO
2. 2.4 µL – Kinase buffer
3. 5 µL – 2X Peptide/Kinase Mixture
4. 2.5 µL – 4X ATP Solution
5. 30-second plate shake
6. 60-minute Kinase Reaction incubation at room temperature
7. 5 µL – Development Reagent Solution
8. 30-second plate shake
9. 60-minute Development Reaction incubation at room temperature
10. Read on fluorescence plate reader and analyze the data

## **Z'-LYTE Assay Controls**

*The following controls are made for each individual kinase and are located on the same plate as the kinase:*

### **0% Phosphorylation Control (100% Inhibition Control)**

The maximum Emission Ratio is established by the 0% Phosphorylation Control (100% Inhibition Control), which contains no ATP and therefore exhibits no kinase activity. This control yields 100% cleaved peptide in the Development Reaction.

### **100% Phosphorylation Control**

The 100% Phosphorylation Control, which consists of a synthetically phosphorylated peptide of the same sequence as the peptide substrate, is designed to allow for the calculation of percent phosphorylation. This control yields a very low percentage of cleaved peptide in the Development Reaction.

The 0% Phosphorylation and 100% Phosphorylation Controls allow one to calculate the percent Phosphorylation achieved in a specific reaction well. Control wells do not include any kinase inhibitors.

### **0% Inhibition Control**

The minimum Emission Ratio in a screen is established by the 0% Inhibition Control, which contains active kinase. This control is designed to produce a 10–50%\* phosphorylated peptide in the Kinase Reaction.

\* Cascade assays may produce up to 70% phosphorylated peptide.

### **Known Inhibitor**

A known inhibitor control standard curve, 10 point titration, is run for each individual kinase on the same plate as the kinase to ensure the kinase is inhibited within an expected IC<sub>50</sub> range previously determined.

*The following controls are prepared for each concentration of Test Compound assayed:*

### **Development Reaction Interference**

The Development Reaction Interference is established by comparing the Test Compound Control wells that do not contain ATP versus the 0% Phosphorylation Control (which does not contain the Test Compound). The expected value for a non-interfering compound should be 100%. Any value outside of 90% to 110% is flagged.

### **Test Compound Fluorescence Interference**

The Test Compound Fluorescence Interference is determined by comparing the Test Compound Control wells that do not contain the Kinase/Peptide Mixture (zero peptide control) versus the 0% Inhibition Control. The expected value for a non-fluorescence compound should be 0%. Any value > 20% is flagged.

## Z'-LYTE Data Analysis

The following equations are used for each set of data points:

|  | Equation  |
|--|---|
| <b>Correction for Background Fluorescence</b>  | $FI_{\text{Sample}} - FI_{\text{TCFI Ctl}}$   |
| <b>Emission Ratio</b><br>(using values corrected for background fluorescence)                            | $\frac{\text{Coumarin Emission (445 nm)}}{\text{Fluorescein Emission (520 nm)}}$  |
| <b>% Phosphorylation (% Phos)</b>  | $\left\{ 1 - \frac{(\text{Emission Ratio} \times F_{100\%}) - C_{100\%}}{(C_{0\%} - C_{100\%}) + [\text{Emission Ratio} \times (F_{100\%} - F_{0\%})]} \right\} * 100$      |
| <b>% Inhibition</b>  | $\left\{ 1 - \frac{\% \text{ Phos}_{\text{Sample}}}{\% \text{ Phos}_{0\% \text{ Inhibition Ctl}}} \right\} * 100$   |
| <b>Z'</b><br>(using Emission Ratio values)   | $1 - \frac{3 * \text{Stdev}_{0\% \text{ Phos Ctl}} + 3 * \text{Stdev}_{0\% \text{ Inhibition}}}{\text{Mean}_{0\% \text{ Phos Ctl}} - \text{Mean}_{0\% \text{ Inhibition}}}$ |
| <b>Difference Between Data Points</b><br>(single point only)   | $  \% \text{ Inhibition}_{\text{Point 1}} - \% \text{ Inhibition}_{\text{Point 2}}  $   |
| <b>Development Reaction Interference (DRI)</b><br>(no ATP control)                                       | $\frac{\text{Emission Ratio}_{\text{DRI Ctl}}}{\text{Emission Ratio}_{0\% \text{ Phos Ctl}}}$   |
| <b>Test Compound Fluorescence Interference (TCFI)</b><br>(check both Coumarin and Fluorescein emissions) | $\frac{FI_{\text{TCFI Ctl}}}{FI_{0\% \text{ Inhibitor Ctl}}}$   |

**FI** = Fluorescence Intensity

**C<sub>100%</sub>** = Average Coumarin emission signal of the 100% Phos. Control

**C<sub>0%</sub>** = Average Coumarin emission signal of the 0% Phos. Control

**F<sub>100%</sub>** = Average Fluorescein emission signal of the 100% Phos. Control

**F<sub>0%</sub>** = Average Fluorescein emission signal of the 0% Phos. Control

**DRI** = Development Reaction Interference

**TCFI** = Test Compound Fluorescence Interference

### Graphing Software

SelectScreen Kinase Profiling Service uses *XLfit* from IDBS. The dose response curve is curve fit to model number 205 (sigmoidal dose-response model). If the bottom of the curve does not fit between -20% & 20% inhibition, it is set to 0% inhibition. If the top of the curve does not fit between 70% and 130% inhibition, it is set to 100% inhibition.

## ***Kinase-Specific Assay Conditions - Direct Format***

### **ABL1**

The 2X ABL1 / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.29 - 1.26 ng ABL1 and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

### **ABL1 E255K**

The 2X ABL1 E255K / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.75 - 4.06 ng ABL1 E255K and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

### **ABL1 F317I**

The 2X ABL1 F317I / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.65 - 8 ng ABL1 F317I and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

### **ABL1 F317L**

The 2X ABL1 F317L / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.5 - 5 ng ABL1 F317L and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

### **ABL1 G250E**

The 2X ABL1 G250E / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.62 - 4.28 ng ABL1 G250E and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

### **ABL1 T315I**

The 2X ABL1 T315I / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.36 - 12 ng ABL1 T315I and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

### **ABL1 Y253F**

The 2X ABL1 Y253F / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.73 - 5.76 ng ABL1 Y253F and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

### **ABL2 (Arg)**

The 2X ABL2 (Arg) / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.42 - 5.3 ng ABL2 (Arg) and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

### **ACVR1B (ALK4)**

The 2X ACVR1B (ALK4) / Ser/Thr 16 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 9.15 - 60 ng ACVR1B (ALK4) and 2 µM Ser/Thr 16 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

### **ADRBK1 (GRK2)**

The 2X ADRBK1 (GRK2) / Ser/Thr 16 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.86 - 67.2 ng ADRBK1 (GRK2) and 2 µM Ser/Thr 16 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

### **ADRBK2 (GRK3)**

The 2X ADRBK2 (GRK3) / Ser/Thr 16 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 9.8 - 39.7 ng ADRBK2 (GRK3) and 2 µM Ser/Thr 16 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**AKT1 (PKB alpha)**

The 2X AKT1 (PKB alpha) / Ser/Thr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.82 - 12 ng AKT1 (PKB alpha) and 2 µM Ser/Thr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**AKT2 (PKB beta)**

The 2X AKT2 (PKB beta) / Ser/Thr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.78 - 40 ng AKT2 (PKB beta) and 2 µM Ser/Thr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**AKT3 (PKB gamma)**

The 2X AKT3 (PKB gamma) / Ser/Thr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.5 - 8.3 ng AKT3 (PKB gamma) and 2 µM Ser/Thr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**ALK**

The 2X ALK / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 4.25 - 96 ng ALK and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**AMPK (A1/B2/G2)**

The 2X AMPK (A1/B2/G2) / Ser/Thr 25 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.32 - 2.4 ng AMPK (A1/B2/G2) and 2 µM Ser/Thr 25 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**AMPK (A1/B2/G3)**

The 2X AMPK (A1/B2/G3) / Ser/Thr 25 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.77 - 6.5 ng AMPK (A1/B2/G3) and 2 µM Ser/Thr 25 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**AMPK (A2/B1/G2)**

The 2X AMPK (A2/B1/G2) / Ser/Thr 25 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.66 - 5.24 ng AMPK (A2/B1/G2) and 2 µM Ser/Thr 25 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**AMPK (A2/B1/G3)**

The 2X AMPK (A2/B1/G3) / Ser/Thr 25 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.48 - 15.8 ng AMPK (A2/B1/G3) and 2 µM Ser/Thr 25 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**AMPK (A2/B2/G3)**

The 2X AMPK (A2/B2/G3) / Ser/Thr 25 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 1.19 - 15.2 ng AMPK (A2/B2/G3) and 2 µM Ser/Thr 25 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**AMPK A1/B1/G1**

The 2X AMPK A1/B1/G1 / Ser/Thr 23 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.21 - 2.86 ng AMPK A1/B1/G1 and 2 µM Ser/Thr 23 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**AMPK A2/B1/G1**

The 2X AMPK A2/B1/G1 / Ser/Thr 23 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.75 - 40 ng AMPK A2/B1/G1 and 2 µM Ser/Thr 23 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.



**AURKA (Aurora A)**

The 2X AURKA (Aurora A) / Ser/Thr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.75 - 3.64 ng AURKA (Aurora A) and 2 µM Ser/Thr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**AURKB (Aurora B)**

The 2X AURKB (Aurora B) / Ser/Thr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.79 - 34.8 ng AURKB (Aurora B) and 2 µM Ser/Thr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**AURKC (Aurora C)**

The 2X AURKC (Aurora C) / Ser/Thr 19 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 7.44 - 100 ng AURKC (Aurora C) and 2 µM Ser/Thr 19 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**AXL**

The 2X AXL / Tyr 06 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 2 - 40 ng AXL and 2 µM Tyr 06 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**BLK**

The 2X BLK / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.08 - 0.75 ng BLK and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**BMX**

The 2X BMX / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.5 - 45.9 ng BMX and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**BRSK1 (SAD1)**

The 2X BRSK1 (SAD1) / Ser/Thr 21 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.8 - 41.7 ng BRSK1 (SAD1) and 2 µM Ser/Thr 21 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**BTK**

The 2X BTK / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.04 - 10.4 ng BTK and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**CAMK1D (CaMKI delta)**

The 2X CAMK1D (CaMKI delta) / Ser/Thr 25 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 800 U/ml Calmodulin, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 15.9 - 133 ng CAMK1D (CaMKI delta) and 2 µM Ser/Thr 25 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 400 U/ml Calmodulin, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**CAMK1G (CAMKI gamma)**

The 2X CAMK1G (CAMKI gamma) / Ser/Thr 23 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 800 U/ml Calmodulin, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 7.5 - 30 ng CAMK1G (CAMKI gamma) and 2 µM Ser/Thr 23 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 400 U/ml Calmodulin, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**CAMK2A (CaMKII alpha)**

The 2X CAMK2A (CaMKII alpha) / Ser/Thr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 800 U/ml Calmodulin, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.27 - 1.2 ng CAMK2A (CaMKII alpha) and 2 µM Ser/Thr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 400 U/ml Calmodulin, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**CAMK2B (CaMKII beta)**

The 2X CAMK2B (CaMKII beta) / Ser/Thr 17 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 800 U/ml Calmodulin, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.19 - 4.15 ng CAMK2B (CaMKII beta) and 2 µM Ser/Thr 17 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 400 U/ml Calmodulin, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**CAMK2D (CaMKII delta)**

The 2X CAMK2D (CaMKII delta) / Ser/Thr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 800 U/ml Calmodulin, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.08 - 0.67 ng CAMK2D (CaMKII delta) and 2 µM Ser/Thr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 400 U/ml Calmodulin, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**CAMK4 (CaMKIV)**

The 2X CAMK4 (CaMKIV) / Ser/Thr 13 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 800 U/ml Calmodulin, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 9.84 - 60 ng CAMK4 (CaMKIV) and 2 µM Ser/Thr 13 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 400 U/ml Calmodulin, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**CDC42 BPA (MRCKA)**

The 2X CDC42 BPA (MRCKA) / Ser/Thr 13 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 9.75 - 57.4 ng CDC42 BPA (MRCKA) and 2 µM Ser/Thr 13 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**CDC42 BPB (MRCKB)**

The 2X CDC42 BPB (MRCKB) / Ser/Thr 13 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3.59 - 21.7 ng CDC42 BPB (MRCKB) and 2 µM Ser/Thr 13 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**CDC42 BPG (MRCKG)**

The 2X CDC42 BPG (MRCKG) / Ser/Thr 13 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 5.5 - 25 ng CDC42 BPG (MRCKG) and 2 µM Ser/Thr 13 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**CDK1/cyclin B**

The 2X CDK1/cyclin B / Ser/Thr 18 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3.5 - 46.4 ng CDK1/cyclin B and 2 µM Ser/Thr 18 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**CDK17/cyclin Y**

The 2X CDK17/cyclin Y / Ser/Thr 12 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.44 - 30 ng CDK17/cyclin Y and 2 µM Ser/Thr 12 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**CDK18/cyclin Y**

The 2X CDK18/cyclin Y / Ser/Thr 12 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.46 - 30 ng CDK18/cyclin Y and 2 µM Ser/Thr 12 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**CDK2/cyclin A**

The 2X CDK2/cyclin A / Ser/Thr 12 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.22 - 10.3 ng CDK2/cyclin A and 2 µM Ser/Thr 12 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**CDK5/p25**

The 2X CDK5/p25 / Ser/Thr 12 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.18 - 1.46 ng CDK5/p25 and 2 µM Ser/Thr 12 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**CDK5/p35**

The 2X CDK5/p35 / Ser/Thr 12 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.14 - 1.3 ng CDK5/p35 and 2 µM Ser/Thr 12 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**CDKL5**

The 2X CDKL5 / Ser/Thr 18 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 10 - 40 ng CDKL5 and 2 µM Ser/Thr 18 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**CHEK1 (CHK1)**

The 2X CHEK1 (CHK1) / Ser/Thr 19 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.38 - 100 ng CHEK1 (CHK1) and 2 µM Ser/Thr 19 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**CHEK2 (CHK2)**

The 2X CHEK2 (CHK2) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 5.51 - 32.2 ng CHEK2 (CHK2) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**CLK1**

The 2X CLK1 / Ser/Thr 09 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 8.1 - 120 ng CLK1 and 2 µM Ser/Thr 09 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:512 dilution of Development Reagent A is added.

**CLK2**

The 2X CLK2 / Ser/Thr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.97 - 32 ng CLK2 and 2 µM Ser/Thr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**CLK3**

The 2X CLK3 / Ser/Thr 18 mixture is prepared in 50 mM Tris pH 8.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 9.57 - 41.5 ng CLK3 and 2 µM Ser/Thr 18 in 50 mM Tris / HEPES pH 8.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**CSF1R (FMS)**

The 2X CSF1R (FMS) / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.2 - 40 ng CSF1R (FMS) and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**CSK**

The 2X CSK / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.5 - 35.4 ng CSK and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**CSNK1A1 (CK1 alpha 1)**

The 2X CSNK1A1 (CK1 alpha 1) / Ser/Thr 11 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 2.6 - 13.5 ng CSNK1A1 (CK1 alpha 1) and 2 µM Ser/Thr 11 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**CSNK1A1L**

The 2X CSNK1A1L / Ser/Thr 11 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 8.75 - 40 ng CSNK1A1L and 2 µM Ser/Thr 11 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**CSNK1D (CK1 delta)**

The 2X CSNK1D (CK1 delta) / Ser/Thr 11 mixture is prepared in 50 mM Tris pH 8.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 9.97 - 72.9 ng CSNK1D (CK1 delta) and 2 µM Ser/Thr 11 in 50 mM Tris / HEPES pH 8.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**CSNK1E (CK1 epsilon)**

The 2X CSNK1E (CK1 epsilon) / Ser/Thr 11 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.35 - 9.44 ng CSNK1E (CK1 epsilon) and 2 µM Ser/Thr 11 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**CSNK1E (CK1 epsilon) R178C**

The 2X CSNK1E (CK1 epsilon) R178C / Ser/Thr 11 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3.75 - 60 ng CSNK1E (CK1 epsilon) R178C and 2 µM Ser/Thr 11 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**CSNK1G1 (CK1 gamma 1)**

The 2X CSNK1G1 (CK1 gamma 1) / Ser/Thr 05 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.04 - 14.7 ng CSNK1G1 (CK1 gamma 1) and 2 µM Ser/Thr 05 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**CSNK1G2 (CK1 gamma 2)**

The 2X CSNK1G2 (CK1 gamma 2) / Ser/Thr 05 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.75 - 12 ng CSNK1G2 (CK1 gamma 2) and 2 µM Ser/Thr 05 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**CSNK1G3 (CK1 gamma 3)**

The 2X CSNK1G3 (CK1 gamma 3) / Ser/Thr 05 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3.1 - 17.5 ng CSNK1G3 (CK1 gamma 3) and 2 µM Ser/Thr 05 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**CSNK2A1 (CK2 alpha 1)**

The 2X CSNK2A1 (CK2 alpha 1) / Ser/Thr 11 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.68 - 25.7 ng CSNK2A1 (CK2 alpha 1) and 2 µM Ser/Thr 11 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**CSNK2A2 (CK2 alpha 2)**

The 2X CSNK2A2 (CK2 alpha 2) / Ser/Thr 11 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.6 - 31.5 ng CSNK2A2 (CK2 alpha 2) and 2 µM Ser/Thr 11 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**DAPK3 (ZIPK)**

The 2X DAPK3 (ZIPK) / Ser/Thr 13 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 5.63 - 34.4 ng DAPK3 (ZIPK) and 2 µM Ser/Thr 13 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**DCAMKL1 (DCLK1)**

The 2X DCAMKL1 (DCLK1) / Ser/Thr 23 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 3.75 - 15 ng DCAMKL1 (DCLK1) and 2 µM Ser/Thr 23 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**DCAMKL2 (DCK2)**

The 2X DCAMKL2 (DCK2) / Ser/Thr 17 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 8.8 - 50.4 ng DCAMKL2 (DCK2) and 2 µM Ser/Thr 17 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**DNA-PK**

The 2X DNA-PK / Ser/Thr 26 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 5.0 µg/ml CT-DNA. The final 10 µL Kinase Reaction consists of 3.88 - 27.3 ng DNA-PK and 2 µM Ser/Thr 26 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 2.5 µg/ml CT-DNA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**DYRK1A**

The 2X DYRK1A / Ser/Thr 18 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.77 - 12.5 ng DYRK1A and 2 µM Ser/Thr 18 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**DYRK1B**

The 2X DYRK1B / Ser/Thr 18 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.47 - 5.84 ng DYRK1B and 2 µM Ser/Thr 18 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**DYRK3**

The 2X DYRK3 / Ser/Thr 09 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.93 - 5.38 ng DYRK3 and 2 µM Ser/Thr 09 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:512 dilution of Development Reagent A is added.

**DYRK4**

The 2X DYRK4 / Ser/Thr 09 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 9.4 - 74.1 ng DYRK4 and 2 µM Ser/Thr 09 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:512 dilution of Development Reagent A is added.

**EEF2K**

The 2X EEF2K / Ser/Thr 24 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 800 U/ml Calmodulin, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 11 - 47.7 ng EEF2K and 2 µM Ser/Thr 24 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 400 U/ml Calmodulin, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16384 dilution of Development Reagent A is added.

**EGFR (ErbB1)**

The 2X EGFR (ErbB1) / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 1.1 - 8 ng EGFR (ErbB1) and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**EGFR (ErbB1) C797S**

The 2X EGFR (ErbB1) C797S / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 1.5 - 10 ng EGFR (ErbB1) C797S and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**EGFR (ErbB1) G719C**

The 2X EGFR (ErbB1) G719C / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 3.75 - 22 ng EGFR (ErbB1) G719C and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**EGFR (ErbB1) G719S**

The 2X EGFR (ErbB1) G719S / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 1.75 - 60 ng EGFR (ErbB1) G719S and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**EGFR (ErbB1) L858R**

The 2X EGFR (ErbB1) L858R / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.2 - 3.36 ng EGFR (ErbB1) L858R and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**EGFR (ErbB1) L861Q**

The 2X EGFR (ErbB1) L861Q / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 2.31 - 11.2 ng EGFR (ErbB1) L861Q and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**EGFR (ErbB1) T790M**

The 2X EGFR (ErbB1) T790M / Tyr 04 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 3.9 - 40 ng EGFR (ErbB1) T790M and 2 µM Tyr 04 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**EGFR (ErbB1) T790M C797S L858R**

The 2X EGFR (ErbB1) T790M C797S L858R / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.5 - 6 ng EGFR (ErbB1) T790M C797S L858R and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**EGFR (ErbB1) T790M L858R**

The 2X EGFR (ErbB1) T790M L858R / Tyr 04 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.25 - 2.08 ng EGFR (ErbB1) T790M L858R and 2 µM Tyr 04 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**EPHA1**

The 2X EPHA1 / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3.12 - 45.2 ng EPHA1 and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**EPHA2**

The 2X EPHA2 / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.1 - 40 ng EPHA2 and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**EPHA4**

The 2X EPHA4 / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 5.42 - 21.7 ng EPHA4 and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**EPHA5**

The 2X EPHA5 / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.18 - 38.1 ng EPHA5 and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**EPHA8**

The 2X EPHA8 / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.2 - 24.6 ng EPHA8 and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**EPHB1**

The 2X EPHB1 / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.39 - 39.4 ng EPHB1 and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**EPHB2**

The 2X EPHB2 / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.55 - 36 ng EPHB2 and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**EPHB3**

The 2X EPHB3 / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.78 - 43.8 ng EPHB3 and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**EPHB4**

The 2X EPHB4 / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.68 - 18 ng EPHB4 and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**ERBB2 (HER2)**

The 2X ERBB2 (HER2) / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% Na<sub>3</sub>N. The final 10 µL Kinase Reaction consists of 1.78 - 30.4 ng ERBB2 (HER2) and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% Na<sub>3</sub>N. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**ERBB4 (HER4)**

The 2X ERBB4 (HER4) / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 2 - 20 ng ERBB4 (HER4) and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**FER**

The 2X FER / Tyr 05 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.9 - 6.82 ng FER and 2 µM Tyr 05 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**FES (FPS)**

The 2X FES (FPS) / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.65 - 5.21 ng FES (FPS) and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**FGFR1**

The 2X FGFR1 / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.3 - 2.45 ng FGFR1 and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**FGFR2**

The 2X FGFR2 / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.19 - 1.99 ng FGFR2 and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**FGFR2 N549H**

The 2X FGFR2 N549H / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.55 - 2.7 ng FGFR2 N549H and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**FGFR3**

The 2X FGFR3 / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.56 - 6 ng FGFR3 and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**FGFR3 K650E**

The 2X FGFR3 K650E / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.6 - 4.6 ng FGFR3 K650E and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**FGFR3 V555M**

The 2X FGFR3 V555M / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.8 - 8 ng FGFR3 V555M and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**FGFR4**

The 2X FGFR4 / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 1.94 - 84 ng FGFR4 and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**FGR**

The 2X FGR / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.75 - 5.9 ng FGR and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**FLT1 (VEGFR1)**

The 2X FLT1 (VEGFR1) / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 2.64 - 12.5 ng FLT1 (VEGFR1) and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**FLT3**

The 2X FLT3 / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.4 - 28 ng FLT3 and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**FLT3 D835Y**

The 2X FLT3 D835Y / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.03 - 1.48 ng FLT3 D835Y and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**FLT4 (VEGFR3)**

The 2X FLT4 (VEGFR3) / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 2 - 20 ng FLT4 (VEGFR3) and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.



**FRAP1 (mTOR)**

The 2X FRAP1 (mTOR) / Ser/Thr 11 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 11.1 - 56 ng FRAP1 (mTOR) and 2 µM Ser/Thr 11 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**FRK (PTK5)**

The 2X FRK (PTK5) / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.1 - 51.9 ng FRK (PTK5) and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**FYN**

The 2X FYN / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 4.44 - 45.6 ng FYN and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**GRK4**

The 2X GRK4 / Ser/Thr 16 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.79 - 3.76 ng GRK4 and 2 µM Ser/Thr 16 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**GRK5**

The 2X GRK5 / Ser/Thr 16 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 4.23 - 38.4 ng GRK5 and 2 µM Ser/Thr 16 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**GRK6**

The 2X GRK6 / Ser/Thr 16 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 11.2 - 61.8 ng GRK6 and 2 µM Ser/Thr 16 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**GRK7**

The 2X GRK7 / Ser/Thr 16 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 4.88 - 23 ng GRK7 and 2 µM Ser/Thr 16 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**GSK3A (GSK3 alpha)**

The 2X GSK3A (GSK3 alpha) / Ser/Thr 09 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.21 - 1.69 ng GSK3A (GSK3 alpha) and 2 µM Ser/Thr 09 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:512 dilution of Development Reagent A is added.

**GSK3B (GSK3 beta)**

The 2X GSK3B (GSK3 beta) / Ser/Thr 09 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.12 - 0.7 ng GSK3B (GSK3 beta) and 2 µM Ser/Thr 09 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:512 dilution of Development Reagent A is added.

**HCK**

The 2X HCK / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.09 - 1.5 ng HCK and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**HIPK1 (Myak)**

The 2X HIPK1 (Myak) / Ser/Thr 09 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.48 - 29.3 ng HIPK1 (Myak) and 2 µM Ser/Thr 09 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:512 dilution of Development Reagent A is added.

**HIPK2**

The 2X HIPK2 / Ser/Thr 09 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3.53 - 19 ng HIPK2 and 2 µM Ser/Thr 09 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:512 dilution of Development Reagent A is added.

**HIPK3 (YAK1)**

The 2X HIPK3 (YAK1) / Ser/Thr 18 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 8.75 - 40 ng HIPK3 (YAK1) and 2 µM Ser/Thr 18 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**HIPK4**

The 2X HIPK4 / Ser/Thr 18 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3 - 35.5 ng HIPK4 and 2 µM Ser/Thr 18 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**IGF1R**

The 2X IGF1R / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 10 - 160 ng IGF1R and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**IKBKB (IKK beta)**

The 2X IKBKB (IKK beta) / Ser/Thr 05 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.93 - 12 ng IKBKB (IKK beta) and 2 µM Ser/Thr 05 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**IKBKE (IKK epsilon)**

The 2X IKBKE (IKK epsilon) / Ser/Thr 11 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.65 - 10.6 ng IKBKE (IKK epsilon) and 2 µM Ser/Thr 11 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**INSR**

The 2X INSR / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 1.94 - 50 ng INSR and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**INSRR (IRR)**

The 2X INSRR (IRR) / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 2.19 - 13.9 ng INSRR (IRR) and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**IRAK4**

The 2X IRAK4 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% Na<sub>3</sub>. The final 10 µL Kinase Reaction consists of 3.45 - 63.6 ng IRAK4 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM DTT, 0.01% Na<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**ITK**

The 2X ITK / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 4.69 - 144 ng ITK and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**JAK1**

The 2X JAK1 / Tyr 06 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% Na<sub>3</sub>. The final 10 µL Kinase Reaction consists of 21.2 - 91.5 ng JAK1 and 2 µM Tyr 06 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% Na<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**JAK2**

The 2X JAK2 / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.05 - 0.42 ng JAK2 and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**JAK2 JH1 JH2**

The 2X JAK2 JH1 JH2 / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.64 - 2.98 ng JAK2 JH1 JH2 and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**JAK2 JH1 JH2 V617F**

The 2X JAK2 JH1 JH2 V617F / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 7.38 - 100 ng JAK2 JH1 JH2 V617F and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**JAK3**

The 2X JAK3 / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.5 - 3.58 ng JAK3 and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**KDR (VEGFR2)**

The 2X KDR (VEGFR2) / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1 - 30 ng KDR (VEGFR2) and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**KIT**

The 2X KIT / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 3.17 - 16 ng KIT and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**KIT T670I**

The 2X KIT T670I / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 6 - 113 ng KIT T670I and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**KIT V559D**

The 2X KIT V559D / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 3.64 - 30 ng KIT V559D and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**KIT V559D V654A**

The 2X KIT V559D V654A / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 8.75 - 50 ng KIT V559D V654A and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**KIT V560G**

The 2X KIT V560G / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 5 - 40 ng KIT V560G and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**KSR2**

The 2X KSR2 / Ser/Thr 17 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.06 - 9.18 ng KSR2 and 2 µM Ser/Thr 17 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**LCK**

The 2X LCK / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.25 - 100 ng LCK and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**LTK (TYK1)**

The 2X LTK (TYK1) / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.78 - 55.5 ng LTK (TYK1) and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**LYN A**

The 2X LYN A / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.75 - 20 ng LYN A and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**LYN B**

The 2X LYN B / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.85 - 14.8 ng LYN B and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**MAP3K19 (YSK4)**

The 2X MAP3K19 (YSK4) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.06 - 9.5 ng MAP3K19 (YSK4) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**MAP3K9 (MLK1)**

The 2X MAP3K9 (MLK1) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% Na<sub>3</sub>N. The final 10 µL Kinase Reaction consists of 5 - 27 ng MAP3K9 (MLK1) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% Na<sub>3</sub>N. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**MAP4K2 (GCK)**

The 2X MAP4K2 (GCK) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.11 - 3 ng MAP4K2 (GCK) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**MAP4K4 (HGK)**

The 2X MAP4K4 (HGK) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.36 - 2 ng MAP4K4 (HGK) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**MAP4K5 (KHS1)**

The 2X MAP4K5 (KHS1) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.25 - 4.5 ng MAP4K5 (KHS1) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**MAPK1 (ERK2)**

The 2X MAPK1 (ERK2) / Ser/Thr 03 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2 - 45.5 ng MAPK1 (ERK2) and 2 µM Ser/Thr 03 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**MAPK11 (p38 beta)**

The 2X MAPK11 (p38 beta) / Ser/Thr 15 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 7.1 - 42.6 ng MAPK11 (p38 beta) and 2 µM Ser/Thr 15 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**MAPK12 (p38 gamma)**

The 2X MAPK12 (p38 gamma) / Ser/Thr 03 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.47 - 3.26 ng MAPK12 (p38 gamma) and 2 µM Ser/Thr 03 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**MAPK13 (p38 delta)**

The 2X MAPK13 (p38 delta) / Ser/Thr 03 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3.22 - 36.6 ng MAPK13 (p38 delta) and 2 µM Ser/Thr 03 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**MAPK14 (p38 alpha) Direct**

The 2X MAPK14 (p38 alpha) / Ser/Thr 15 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 7.07 - 51.4 ng MAPK14 (p38 alpha) and 2 µM Ser/Thr 15 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**MAPK3 (ERK1)**

The 2X MAPK3 (ERK1) / Ser/Thr 03 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 5.94 - 94.5 ng MAPK3 (ERK1) and 2 µM Ser/Thr 03 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**MAPK7 (ERK5)**

The 2X MAPK7 (ERK5) / Ser/Thr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.51 - 12.7 ng MAPK7 (ERK5) and 2 µM Ser/Thr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**MAPKAPK2**

The 2X MAPKAPK2 / Ser/Thr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.04 - 0.18 ng MAPKAPK2 and 2 µM Ser/Thr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**MAPKAPK3**

The 2X MAPKAPK3 / Ser/Thr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.09 - 2.05 ng MAPKAPK3 and 2 µM Ser/Thr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**MAPKAPK5 (PRAK)**

The 2X MAPKAPK5 (PRAK) / Ser/Thr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.29 - 8.32 ng MAPKAPK5 (PRAK) and 2 µM Ser/Thr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**MARK1 (MARK)**

The 2X MARK1 (MARK) / Ser/Thr 21 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 14.9 - 168 ng MARK1 (MARK) and 2 µM Ser/Thr 21 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**MARK2**

The 2X MARK2 / Ser/Thr 21 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 37.2 - 149 ng MARK2 and 2 µM Ser/Thr 21 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**MARK3**

The 2X MARK3 / Ser/Thr 25 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.46 - 2.59 ng MARK3 and 2 µM Ser/Thr 25 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**MARK4**

The 2X MARK4 / Ser/Thr 25 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.54 - 2.36 ng MARK4 and 2 µM Ser/Thr 25 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**MATK (HYL)**

The 2X MATK (HYL) / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 4.06 - 117 ng MATK (HYL) and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**MELK**

The 2X MELK / Ser/Thr 17 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.76 - 6.48 ng MELK and 2 µM Ser/Thr 17 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**MERTK (cMER)**

The 2X MERTK (cMER) / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.68 - 6.22 ng MERTK (cMER) and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**MET (cMet)**

The 2X MET (cMet) / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.49 - 7.84 ng MET (cMet) and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**MET (cMet) Y1235D**

The 2X MET (cMet) Y1235D / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.55 - 6 ng MET (cMet) Y1235D and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**MET M1250T**

The 2X MET M1250T / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3 - 16.2 ng MET M1250T and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**MINK1**

The 2X MINK1 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.9 - 10 ng MINK1 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**MKNK1 (MNK1)**

The 2X MKNK1 (MNK1) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 16 - 63.8 ng MKNK1 (MNK1) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**MST1R (RON)**

The 2X MST1R (RON) / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.25 - 1.18 ng MST1R (RON) and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**MST4**

The 2X MST4 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 6 - 50 ng MST4 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**MUSK**

The 2X MUSK / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 21.3 - 134 ng MUSK and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**MYLK2 (skMLCK)**

The 2X MYLK2 (skMLCK) / Ser/Thr 13 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 800 U/ml Calmodulin, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 3.13 - 15 ng MYLK2 (skMLCK) and 2 µM Ser/Thr 13 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 400 U/ml Calmodulin, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**NEK1**

The 2X NEK1 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.3 - 80 ng NEK1 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**NEK2**

The 2X NEK2 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.75 - 5.4 ng NEK2 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**NEK4**

The 2X NEK4 / Ser/Thr 17 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 4 - 18.1 ng NEK4 and 2 µM Ser/Thr 17 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**NEK6**

The 2X NEK6 / Ser/Thr 22 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.13 - 22 ng NEK6 and 2 µM Ser/Thr 22 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:8192 dilution of Development Reagent A is added.

**NEK7**

The 2X NEK7 / Ser/Thr 22 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 7.5 - 103 ng NEK7 and 2 µM Ser/Thr 22 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:8192 dilution of Development Reagent A is added.

**NEK9**

The 2X NEK9 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 1.44 - 22.4 ng NEK9 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**NIM1K**

The 2X NIM1K / Ser/Thr 25 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.28 - 30 ng NIM1K and 2 µM Ser/Thr 25 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**NTRK1 (TRKA)**

The 2X NTRK1 (TRKA) / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 6 - 88.7 ng NTRK1 (TRKA) and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**NTRK2 (TRKB)**

The 2X NTRK2 (TRKB) / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.34 - 5.46 ng NTRK2 (TRKB) and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**NTRK3 (TRKC)**

The 2X NTRK3 (TRKC) / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.33 - 45.1 ng NTRK3 (TRKC) and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**PAK1**

The 2X PAK1 / Ser/Thr 19 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.71 - 30.8 ng PAK1 and 2 µM Ser/Thr 19 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**PAK2 (PAK65)**

The 2X PAK2 (PAK65) / Ser/Thr 20 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.29 - 6 ng PAK2 (PAK65) and 2 µM Ser/Thr 20 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**PAK3**

The 2X PAK3 / Ser/Thr 20 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.38 - 22 ng PAK3 and 2 µM Ser/Thr 20 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**PAK4**

The 2X PAK4 / Ser/Thr 20 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.1 - 0.75 ng PAK4 and 2 µM Ser/Thr 20 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**PAK6**

The 2X PAK6 / Ser/Thr 20 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.38 - 7.38 ng PAK6 and 2 µM Ser/Thr 20 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**PAK7 (KIAA1264)**

The 2X PAK7 (KIAA1264) / Ser/Thr 20 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.05 - 0.27 ng PAK7 (KIAA1264) and 2 µM Ser/Thr 20 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**PASK**

The 2X PASK / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3.6 - 15.4 ng PASK and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PDGFRA (PDGFR alpha)**

The 2X PDGFRA (PDGFR alpha) / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 1.54 - 22.6 ng PDGFRA (PDGFR alpha) and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**PDGFRA D842V**

The 2X PDGFRA D842V / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 3.49 - 22.4 ng PDGFRA D842V and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**PDGFRA T674I**

The 2X PDGFRA T674I / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 11.4 - 45.4 ng PDGFRA T674I and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.



**PDGFRA V561D**

The 2X PDGFRA V561D / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.5 - 12 ng PDGFRA V561D and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**PDGFRB (PDGFR beta)**

The 2X PDGFRB (PDGFR beta) / Tyr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 7 - 50 ng PDGFRB (PDGFR beta) and 2 µM Tyr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**PDK1 Direct**

The 2X PDK1 / Ser/Thr 07 mixture is prepared in 50 mM Tris pH 8.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 5.15 - 44 ng PDK1 and 2 µM Ser/Thr 07 in 50 mM Tris / HEPES pH 8.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PEAK1**

The 2X PEAK1 / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 2.73 - 40 ng PEAK1 and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**PHKG1**

The 2X PHKG1 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 800 U/ml Calmodulin, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 2.13 - 15 ng PHKG1 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 400 U/ml Calmodulin, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PHKG2**

The 2X PHKG2 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.3 - 10.1 ng PHKG2 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PIM1**

The 2X PIM1 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.3 - 13.9 ng PIM1 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PIM2**

The 2X PIM2 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.44 - 6.86 ng PIM2 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PIM3**

The 2X PIM3 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.38 - 50 ng PIM3 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PKN1 (PRK1)**

The 2X PKN1 (PRK1) / Ser/Thr 07 mixture is prepared in 50 mM Tris pH 8.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 3 - 60 ng PKN1 (PRK1) and 2 µM Ser/Thr 07 in 50 mM Tris / HEPES pH 8.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PLK1**

The 2X PLK1 / Ser/Thr 16 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.98 - 26 ng PLK1 and 2 µM Ser/Thr 16 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**PLK2**

The 2X PLK2 / Ser/Thr 16 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 9.59 - 160 ng PLK2 and 2 µM Ser/Thr 16 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**PLK3**

The 2X PLK3 / Ser/Thr 16 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.27 - 1.8 ng PLK3 and 2 µM Ser/Thr 16 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**PRKACA (PKA)**

The 2X PRKACA (PKA) / Ser/Thr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.01 - 0.14 ng PRKACA (PKA) and 2 µM Ser/Thr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**PRKCA (PKC alpha)**

The 2X PRKCA (PKC alpha) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 2X Novel Lipid Mix, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.02 - 0.16 ng PRKCA (PKC alpha) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 1X Novel Lipid Mix, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PRKCB1 (PKC beta I)**

The 2X PRKCB1 (PKC beta I) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 2X Novel Lipid Mix, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.02 - 0.58 ng PRKCB1 (PKC beta I) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 1X Novel Lipid Mix, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PRKCB2 (PKC beta II)**

The 2X PRKCB2 (PKC beta II) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 2X Novel Lipid Mix, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.04 - 0.56 ng PRKCB2 (PKC beta II) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 1X Novel Lipid Mix, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PRKCD (PKC delta)**

The 2X PRKCD (PKC delta) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 2X Novel Lipid Mix, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.23 - 1.32 ng PRKCD (PKC delta) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 1X Novel Lipid Mix, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PRKCE (PKC epsilon)**

The 2X PRKCE (PKC epsilon) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 2X Novel Lipid Mix, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.14 - 3 ng PRKCE (PKC epsilon) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 1X Novel Lipid Mix, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PRKCG (PKC gamma)**

The 2X PRKCG (PKC gamma) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 2X Novel Lipid Mix, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.03 - 0.24 ng PRKCG (PKC gamma) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 1X Novel Lipid Mix, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PRKCH (PKC eta)**

The 2X PRKCH (PKC eta) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 2X Novel Lipid Mix, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.29 - 6 ng PRKCH (PKC eta) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 1X Novel Lipid Mix, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PRKCI (PKC iota)**

The 2X PRKCI (PKC iota) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 2X Novel Lipid Mix, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 1.25 - 6 ng PRKCI (PKC iota) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 1X Novel Lipid Mix, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PRKCN (PKD3)**

The 2X PRKCN (PKD3) / Ser/Thr 17 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.25 - 40 ng PRKCN (PKD3) and 2 µM Ser/Thr 17 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**PRKCQ (PKC theta)**

The 2X PRKCQ (PKC theta) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 2X Novel Lipid Mix, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 0.02 - 0.36 ng PRKCQ (PKC theta) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 1X Novel Lipid Mix, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PRKCZ (PKC zeta)**

The 2X PRKCZ (PKC zeta) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 4 mM CaCl<sub>2</sub>, 2X Novel Lipid Mix, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 2.8 - 12.7 ng PRKCZ (PKC zeta) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM CaCl<sub>2</sub>, 1X Novel Lipid Mix, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**PRKD1 (PKC mu)**

The 2X PRKD1 (PKC mu) / Ser/Thr 17 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.56 - 7.8 ng PRKD1 (PKC mu) and 2 µM Ser/Thr 17 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**PRKD2 (PKD2)**

The 2X PRKD2 (PKD2) / Ser/Thr 17 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.64 - 5.84 ng PRKD2 (PKD2) and 2 µM Ser/Thr 17 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**PRKG1**

The 2X PRKG1 / Ser/Thr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 20 µM cGMP. The final 10 µL Kinase Reaction consists of 0.17 - 1 ng PRKG1 and 2 µM Ser/Thr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 10 µM cGMP. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**PRKG2 (PKG2)**

The 2X PRKG2 (PKG2) / Ser/Thr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 20 µM cGMP. The final 10 µL Kinase Reaction consists of 0.1 - 1.4 ng PRKG2 (PKG2) and 2 µM Ser/Thr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 10 µM cGMP. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**PRKX**

The 2X PRKX / Ser/Thr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.75 - 4.98 ng PRKX and 2 µM Ser/Thr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**PTK2 (FAK)**

The 2X PTK2 (FAK) / Tyr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 12.5 - 100 ng PTK2 (FAK) and 2 µM Tyr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.

**PTK2B (FAK2)**

The 2X PTK2B (FAK2) / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 5.26 - 45.9 ng PTK2B (FAK2) and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**PTK6 (Brk)**

The 2X PTK6 (Brk) / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 11.8 - 65 ng PTK6 (Brk) and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**RET**

The 2X RET / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.49 - 3.64 ng RET and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**RET A883F**

The 2X RET A883F / Tyr 04 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 1.02 - 6.74 ng RET A883F and 2 µM Tyr 04 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**RET S891A**

The 2X RET S891A / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.15 - 1.2 ng RET S891A and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**RET V804E**

The 2X RET V804E / Tyr 04 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 1.5 - 20 ng RET V804E and 2 µM Tyr 04 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**RET V804L**

The 2X RET V804L / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.52 - 4.74 ng RET V804L and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**RET Y791F**

The 2X RET Y791F / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.86 - 6.16 ng RET Y791F and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**ROCK1**

The 2X ROCK1 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.1 - 8 ng ROCK1 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**ROCK2**

The 2X ROCK2 / Ser/Thr 13 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.58 - 4.13 ng ROCK2 and 2 µM Ser/Thr 13 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**ROS1**

The 2X ROS1 / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.63 - 11.6 ng ROS1 and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**RPS6KA1 (RSK1)**

The 2X RPS6KA1 (RSK1) / Ser/Thr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3.51 - 22.3 ng RPS6KA1 (RSK1) and 2 µM Ser/Thr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**RPS6KA2 (RSK3)**

The 2X RPS6KA2 (RSK3) / Ser/Thr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.5 - 7 ng RPS6KA2 (RSK3) and 2 µM Ser/Thr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**RPS6KA3 (RSK2)**

The 2X RPS6KA3 (RSK2) / Ser/Thr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.1 - 0.48 ng RPS6KA3 (RSK2) and 2 µM Ser/Thr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**RPS6KA4 (MSK2)**

The 2X RPS6KA4 (MSK2) / Ser/Thr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3.1 - 23.4 ng RPS6KA4 (MSK2) and 2 µM Ser/Thr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**RPS6KA5 (MSK1)**

The 2X RPS6KA5 (MSK1) / Ser/Thr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.23 - 2.84 ng RPS6KA5 (MSK1) and 2 µM Ser/Thr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**RPS6KA6 (RSK4)**

The 2X RPS6KA6 (RSK4) / Ser/Thr 20 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.19 - 2.5 ng RPS6KA6 (RSK4) and 2 µM Ser/Thr 20 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**RPS6KB1 (p70S6K)**

The 2X RPS6KB1 (p70S6K) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.87 - 17.7 ng RPS6KB1 (p70S6K) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**RPS6KB2 (p70S6Kb)**

The 2X RPS6KB2 (p70S6Kb) / Ser/Thr 19 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 10 - 70 ng RPS6KB2 (p70S6Kb) and 2 µM Ser/Thr 19 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:256 dilution of Development Reagent A is added.

**SBK1**

The 2X SBK1 / Ser/Thr 25 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 2.79 - 18.5 ng SBK1 and 2 µM Ser/Thr 25 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**SGK (SGK1)**

The 2X SGK (SGK1) / Ser/Thr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.09 - 1 ng SGK (SGK1) and 2 µM Ser/Thr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**SGK2**

The 2X SGK2 / Ser/Thr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.21 - 3 ng SGK2 and 2 µM Ser/Thr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**SGKL (SGK3)**

The 2X SGKL (SGK3) / Ser/Thr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.19 - 1.26 ng SGKL (SGK3) and 2 µM Ser/Thr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**SNF1LK2**

The 2X SNF1LK2 / Ser/Thr 25 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.62 - 9.94 ng SNF1LK2 and 2 µM Ser/Thr 25 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**SRC**

The 2X SRC / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.54 - 36 ng SRC and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**SRC N1**

The 2X SRC N1 / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1 - 15.5 ng SRC N1 and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**SRMS (Srm)**

The 2X SRMS (Srm) / Tyr 01 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2 - 8.97 ng SRMS (Srm) and 2 µM Tyr 01 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent B is added.

**SRPK1**

The 2X SRPK1 / Ser/Thr 18 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3.74 - 37 ng SRPK1 and 2 µM Ser/Thr 18 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**SRPK2**

The 2X SRPK2 / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 3.76 - 46.8 ng SRPK2 and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**STK22B (TSSK2)**

The 2X STK22B (TSSK2) / Ser/Thr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 5.55 - 47.2 ng STK22B (TSSK2) and 2 µM Ser/Thr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**STK22D (TSSK1)**

The 2X STK22D (TSSK1) / Ser/Thr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.69 - 12.2 ng STK22D (TSSK1) and 2 µM Ser/Thr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**STK23 (MSSK1)**

The 2X STK23 (MSSK1) / Ser/Thr 18 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 5.69 - 26.2 ng STK23 (MSSK1) and 2 µM Ser/Thr 18 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**STK24 (MST3)**

The 2X STK24 (MST3) / Ser/Thr 07 mixture is prepared in 50 mM Tris pH 8.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 14.7 - 59.4 ng STK24 (MST3) and 2 µM Ser/Thr 07 in 50 mM Tris / HEPES pH 8.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**STK25 (YSK1)**

The 2X STK25 (YSK1) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.34 - 37.5 ng STK25 (YSK1) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**STK3 (MST2)**

The 2X STK3 (MST2) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 6.25 - 38.7 ng STK3 (MST2) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**STK4 (MST1)**

The 2X STK4 (MST1) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 6.25 - 160 ng STK4 (MST1) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**SYK**

The 2X SYK / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.69 - 5.35 ng SYK and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**TAOK2 (TAO1)**

The 2X TAOK2 (TAO1) / Ser/Thr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.4 - 11 ng TAOK2 (TAO1) and 2 µM Ser/Thr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:45000 dilution of Development Reagent A is added.

**TBK1**

The 2X TBK1 / Ser/Thr 05 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.93 - 13 ng TBK1 and 2 µM Ser/Thr 05 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:64 dilution of Development Reagent B is added.

**TEK (Tie2)**

The 2X TEK (Tie2) / Tyr 05 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 4 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 1 - 4.6 ng TEK (Tie2) and 2 µM Tyr 05 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 2 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**TEK (TIE2) Y897S**

The 2X TEK (TIE2) Y897S / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.25 - 16 ng TEK (TIE2) Y897S and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**TNK1**

The 2X TNK1 / Ser/Thr 13 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 10 - 40 ng TNK1 and 2 µM Ser/Thr 13 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**TXK**

The 2X TXK / Tyr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.38 - 93.6 ng TXK and 2 µM Tyr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**TYK2**

The 2X TYK2 / Tyr 03 mixture is prepared in 50 mM HEPES pH 6.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 20 - 120 ng TYK2 and 2 µM Tyr 03 in 50 mM HEPES pH 7.0, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**TYRO3 (RSE)**

The 2X TYRO3 (RSE) / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 2.15 - 25.8 ng TYRO3 (RSE) and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**YES1**

The 2X YES1 / Tyr 02 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.75 - 4.5 ng YES1 and 2 µM Tyr 02 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:128 dilution of Development Reagent A is added.

**ZAP70**

The 2X ZAP70 / Tyr 07 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MnCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT, 0.02% NaN<sub>3</sub>. The final 10 µL Kinase Reaction consists of 20.3 - 126 ng ZAP70 and 2 µM Tyr 07 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 5 mM MgCl<sub>2</sub>, 5 mM MnCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT, 0.01% NaN<sub>3</sub>. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:16 dilution of Development Reagent B is added.



## ***Kinase-Specific Assay Conditions - Cascade Format***

### **BRAF**

The 2X BRAF / inactive MAP2K1 (MEK1) / inactive MAPK1 (ERK2) / Ser/Thr 03 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.04 - 0.15 ng BRAF, 1X inactive MAP2K1 (MEK1) / inactive MAPK1 (ERK2), and 2 µM Ser/Thr 03 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

### **BRAF V599E**

The 2X BRAF V599E / inactive MAP2K1 (MEK1) / inactive MAPK1 (ERK2) / Ser/Thr 03 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.003 - 0.01 ng BRAF V599E, 1X inactive MAP2K1 (MEK1) / inactive MAPK1 (ERK2), and 2 µM Ser/Thr 03 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

### **MAP2K1 (MEK1)**

The 2X MAP2K1 (MEK1) / inactive MAPK1 (ERK2) / Ser/Thr 03 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.06 - 0.25 ng MAP2K1 (MEK1), 105 ng inactive MAPK1 (ERK2), and 2 µM Ser/Thr 03 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

### **MAP2K2 (MEK2)**

The 2X MAP2K2 (MEK2) / inactive MAPK1 (ERK2) / Ser/Thr 03 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.15 - 0.6 ng MAP2K2 (MEK2), 105 ng inactive MAPK1 (ERK2), and 2 µM Ser/Thr 03 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

### **MAP2K6 (MKK6)**

The 2X MAP2K6 (MKK6) / inactive MAPK12 (p38 gamma) / Ser/Thr 03 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1.92 - 7.67 ng MAP2K6 (MKK6), 100 ng inactive MAPK12 (p38 gamma), and 2 µM Ser/Thr 03 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

### **MAP3K8 (COT)**

The 2X MAP3K8 (COT) / inactive MAP2K1 (MEK1) / inactive MAPK1 (ERK2) / Ser/Thr 03 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 1 - 4 ng MAP3K8 (COT), 1X inactive MAP2K1 (MEK1) / inactive MAPK1 (ERK2), and 2 µM Ser/Thr 03 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

### **MAPK10 (JNK3)**

The 2X MAPK10 (JNK3) / inactive MAPKAPK2 / Ser/Thr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.96 - 3.86 ng MAPK10 (JNK3), 12.5 ng inactive MAPKAPK2, and 2 µM Ser/Thr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

### **MAPK14 (p38 alpha)**

The 2X MAPK14 (p38 alpha) / inactive MAPKAPK2 / Ser/Thr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.003 - 0.01 ng MAPK14 (p38 alpha), 6.5 ng inactive MAPKAPK2, and 2 µM Ser/Thr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

### **MAPK8 (JNK1)**

The 2X MAPK8 (JNK1) / inactive MAPKAPK2 / Ser/Thr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 1.5 - 6 ng MAPK8 (JNK1), 25 ng inactive MAPKAPK2, and 2 µM Ser/Thr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**MAPK9 (JNK2)**

The 2X MAPK9 (JNK2) / inactive MAPKAPK2 / Ser/Thr 04 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 2 mM DTT. The final 10 µL Kinase Reaction consists of 0.9 - 3.6 ng MAPK9 (JNK2), 12.5 ng inactive MAPKAPK2, and 2 µM Ser/Thr 04 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA, 1 mM DTT. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**PDK1**

The 2X PDK1 / inactive AKT2 (PKB beta) / Ser/Thr 06 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 5 - 20 ng PDK1, 150 ng inactive AKT2 (PKB beta), and 2 µM Ser/Thr 06 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:4096 dilution of Development Reagent A is added.

**RAF1 (cRAF) Y340D Y341D**

The 2X RAF1 (cRAF) Y340D Y341D / inactive MAP2K1 (MEK1) / inactive MAPK1 (ERK2) / Ser/Thr 03 mixture is prepared in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. The final 10 µL Kinase Reaction consists of 0.006 - 0.02 ng RAF1 (cRAF) Y340D Y341D, 1X inactive MAP2K1 (MEK1) / inactive MAPK1 (ERK2), and 2 µM Ser/Thr 03 in 50 mM HEPES pH 7.5, 0.01% BRIJ-35, 10 mM MgCl<sub>2</sub>, 1 mM EGTA. After the 1 hour Kinase Reaction incubation, 5 µL of a 1:1024 dilution of Development Reagent A is added.

**Table of Kinase ATP Km Bins and Inhibitor Validation**

The table below provides specifications and data around each kinase. The representative IC50 value with a known inhibitor for each kinase was determined at the ATP bin nearest to the ATP Km app, unless indicated with an asterisk (\*) in which case the IC50 value was determined at 100 μM ATP.

| Assay                 | Z'-LYTE Substrate | ATP Km app (μM) | ATP Bin (μM) | Inhibitor         | IC50 (nM) |
|-----------------------|-------------------|-----------------|--------------|-------------------|-----------|
| ABL1                  | Tyr 02            | 12              | 10           | Tyrphostin AG1478 | 357       |
| ABL1 E255K            | Tyr 02            | 4.2             | 5            | Staurosporine     | 79.8      |
| ABL1 F317I            | Tyr 02            | 15              | 10           | Staurosporine     | 218       |
| ABL1 F317L            | Tyr 02            | 14              | 10           | Staurosporine     | 198       |
| ABL1 G250E            | Tyr 02            | 5.4             | 5            | Staurosporine     | 38.8      |
| ABL1 T315I            | Tyr 02            | 2.8             | 5            | Staurosporine     | 13.6      |
| ABL1 Y253F            | Tyr 02            | 11.8            | 10           | Staurosporine     | 50.8      |
| ABL2 (Arg)            | Tyr 02            | 30              | 25           | Tyrphostin AG1478 | 907       |
| ACVR1B (ALK4)         | Ser/Thr 16        | 1.6             | 5            | SB 431542         | 646       |
| ADRBK1 (GRK2)         | Ser/Thr 16        | 13.3            | 10           | Staurosporine     | 1280      |
| ADRBK2 (GRK3)         | Ser/Thr 16        | 11.7            | 10           | Staurosporine     | 1540      |
| AKT1 (PKB alpha)      | Ser/Thr 06        | 75              | 75           | Staurosporine     | 12.5      |
| AKT2 (PKB beta)       | Ser/Thr 06        | 200             | 200          | Staurosporine     | 51.9      |
| AKT3 (PKB gamma)      | Ser/Thr 06        | 100             | 100          | Staurosporine     | 10.9      |
| ALK                   | Tyr 01            | 30              | 25           | Staurosporine     | 17.1      |
| AMPK (A1/B2/G2)       | Ser/Thr 25        | 9               | 10           | Staurosporine     | 0.242     |
| AMPK (A1/B2/G3)       | Ser/Thr 25        | 13              | 10           | Staurosporine     | 0.791     |
| AMPK (A2/B1/G2)       | Ser/Thr 25        | 11              | 10           | Staurosporine     | 0.499     |
| AMPK (A2/B1/G3)       | Ser/Thr 25        | 24              | 25           | Staurosporine     | 0.845     |
| AMPK (A2/B2/G3)       | Ser/Thr 25        | 40              | 50           | Staurosporine     | 0.359     |
| AMPK A1/B1/G1         | Ser/Thr 23        | 43              | 50           | Staurosporine     | 0.245     |
| AMPK A2/B1/G1         | Ser/Thr 23        | 148             | 150          | Staurosporine     | 0.722     |
| AURKA (Aurora A)      | Ser/Thr 01        | 10              | 10           | Staurosporine     | 3.99      |
| AURKB (Aurora B)      | Ser/Thr 01        | 81              | 75           | Staurosporine     | 5.80      |
| AURKC (Aurora C)      | Ser/Thr 19        | 26              | 25           | Staurosporine     | 6.05      |
| AXL                   | Tyr 06            | 71              | 75           | Staurosporine     | 3.99      |
| BLK                   | Tyr 01            | 30              | 25           | Tyrphostin AG1478 | 972       |
| BMX                   | Tyr 01            | 107             | 100          | Staurosporine     | 16.1      |
| BRAF                  | Ser/Thr 03        | Cascade         | 100          | Staurosporine     | 28.7 *    |
| BRAF V599E            | Ser/Thr 03        | Cascade         | 100          | Staurosporine     | 37.7 *    |
| BRSK1 (SAD1)          | Ser/Thr 21        | 32              | 25           | Staurosporine     | 2.27      |
| BTK                   | Tyr 01            | 36              | 25           | Tyrphostin AG1478 | 2370      |
| CAMK1D (CaMKI delta)  | Ser/Thr 25        | 28              | 25           | Staurosporine     | 19.6      |
| CAMK1G (CaMKI gamma)  | Ser/Thr 23        | 294             | 300          | Staurosporine     | 14.3      |
| CAMK2A (CaMKII alpha) | Ser/Thr 04        | 10              | 10           | Staurosporine     | 0.709     |
| CAMK2B (CaMKII beta)  | Ser/Thr 17        | 76              | 75           | Staurosporine     | 0.740     |
| CAMK2D (CaMKII delta) | Ser/Thr 04        | 6               | 5            | Staurosporine     | 0.356     |
| CAMK4 (CaMKIV)        | Ser/Thr 13        | 18              | 25           | Staurosporine     | 93.1      |
| CDC42 BPA (MRCKA)     | Ser/Thr 13        | 1               | 5            | Staurosporine     | 13.6      |
| CDC42 BPB (MRCKB)     | Ser/Thr 13        | 1               | 5            | Staurosporine     | 15.7      |
| CDC42 BPG (MRCKG)     | Ser/Thr 13        | 2               | 5            | Staurosporine     | 5.22      |
| CDK1/cyclin B         | Ser/Thr 18        | 34              | 25           | Staurosporine     | 7.02      |
| CDK17/cyclin Y        | Ser/Thr 12        | 47              | 50           | Staurosporine     | 22.7      |
| CDK18/cyclin Y        | Ser/Thr 12        | 74              | 75           | Staurosporine     | 47.6      |
| CDK2/cyclin A         | Ser/Thr 12        | 31              | 25           | Staurosporine     | 3.53      |

| Assay                          | Z'-LYTE<br>Substrate | ATP<br>Km app (μM) | ATP<br>Bin (μM) | Inhibitor         | IC50 (nM) |
|--------------------------------|----------------------|--------------------|-----------------|-------------------|-----------|
| CDK5/p25                       | Ser/Thr 12           | 17                 | 10              | Staurosporine     | 4.24      |
| CDK5/p35                       | Ser/Thr 12           | 8                  | 10              | Staurosporine     | 4.70      |
| CDKL5                          | Ser/Thr 18           | 516                | 500             | Staurosporine     | 408       |
| CHEK1 (CHK1)                   | Ser/Thr 19           | 53                 | 50              | Staurosporine     | 1.84      |
| CHEK2 (CHK2)                   | Ser/Thr 07           | 84                 | 75              | Staurosporine     | 18.2      |
| CLK1                           | Ser/Thr 09           | 24                 | 25              | Staurosporine     | 72.0      |
| CLK2                           | Ser/Thr 06           | 30                 | 25              | Staurosporine     | 9.62      |
| CLK3                           | Ser/Thr 18           | 128                | 150             | Staurosporine     | 1390      |
| CSF1R (FMS)                    | Tyr 01               | 450                | 500             | Tyrphostin AG1478 | 4110      |
| CSK                            | Tyr 02               | 15                 | 10              | Staurosporine     | 61.9      |
| CSNK1A1 (CK1 alpha 1)          | Ser/Thr 11           | 2                  | 5               | TBB               | 10800     |
| CSNK1A1L                       | Ser/Thr 11           | 2                  | 5               | TBB               | 14100     |
| CSNK1D (CK1 delta)             | Ser/Thr 11           | 4                  | 5               | TBB               | 2890      |
| CSNK1E (CK1 epsilon)           | Ser/Thr 11           | 2                  | 5               | TBB               | 2870      |
| CSNK1E (CK1 epsilon) R178C     | Ser/Thr 11           | 20                 | 25              | TBB               | 2340      |
| CSNK1G1 (CK1 gamma 1)          | Ser/Thr 05           | 2                  | 5               | Chetomin          | 82.6      |
| CSNK1G2 (CK1 gamma 2)          | Ser/Thr 05           | 5                  | 5               | TBB               | 21200     |
| CSNK1G3 (CK1 gamma 3)          | Ser/Thr 05           | 4.4                | 5               | TBB               | 48400     |
| CSNK2A1 (CK2 alpha 1)          | Ser/Thr 11           | 4                  | 5               | TBB               | 2290      |
| CSNK2A2 (CK2 alpha 2)          | Ser/Thr 11           | 46                 | 50              | TBB               | 2150      |
| DAPK3 (ZIPK)                   | Ser/Thr 13           | 2.5                | 5               | Staurosporine     | 16.8      |
| DCAMKL1 (DCLK1)                | Ser/Thr 23           | 261                | 300             | Staurosporine     | 244       |
| DCAMKL2 (DCK2)                 | Ser/Thr 17           | 149.5              | 150             | Staurosporine     | 36.7      |
| DNA-PK                         | Ser/Thr 26           | 17                 | 25              | PI-103            | 9.87      |
| DYRK1A                         | Ser/Thr 18           | 99.7               | 100             | Staurosporine     | 32.5      |
| DYRK1B                         | Ser/Thr 18           | 86                 | 75              | Staurosporine     | 5.81      |
| DYRK3                          | Ser/Thr 09           | 4.7                | 5               | Staurosporine     | 96.8      |
| DYRK4                          | Ser/Thr 09           | 2.2                | 5               | TBB               | 5970      |
| EEF2K                          | Ser/Thr 24           | 15                 | 10              | NH125             | 10300     |
| EGFR (ErbB1)                   | Tyr 04               | 11.5               | 10              | Staurosporine     | 60.4      |
| EGFR (ErbB1) C797S             | Tyr 04               | 2                  | 5               | Staurosporine     | 764       |
| EGFR (ErbB1) G719C             | Tyr 04               | 142                | 150             | Staurosporine     | 3360      |
| EGFR (ErbB1) G719S             | Tyr 04               | 94                 | 100             | Staurosporine     | 3900      |
| EGFR (ErbB1) L858R             | Tyr 04               | 44.2               | 50              | Staurosporine     | 66.7      |
| EGFR (ErbB1) L861Q             | Tyr 04               | 12.7               | 10              | Staurosporine     | 62.8      |
| EGFR (ErbB1) T790M             | Tyr 04               | 10                 | 10              | Staurosporine     | 0.823     |
| EGFR (ErbB1) T790M C797S L858R | Tyr 04               | 6                  | 5               | Staurosporine     | 1.07      |
| EGFR (ErbB1) T790M L858R       | Tyr 04               | 26                 | 25              | Staurosporine     | 0.473     |
| EPHA1                          | Tyr 02               | 18.5               | 25              | Staurosporine     | 67.5      |
| EPHA2                          | Tyr 01               | 65.4               | 75              | Staurosporine     | 256       |
| EPHA4                          | Tyr 02               | 106                | 100             | Staurosporine     | 81.6      |
| EPHA5                          | Tyr 01               | 129                | 150             | Staurosporine     | 74.9      |
| EPHA8                          | Tyr 02               | 123                | 100             | Staurosporine     | 122       |
| EPHB1                          | Tyr 02               | 61.4               | 50              | Staurosporine     | 100       |
| EPHB2                          | Tyr 02               | 66                 | 75              | Staurosporine     | 104       |
| EPHB3                          | Tyr 02               | 70                 | 75              | PP2               | 372       |
| EPHB4                          | Tyr 01               | 115                | 100             | Tyrphostin AG1478 | 1350      |
| ERBB2 (HER2)                   | Tyr 06               | 14.1               | 10              | Tyrphostin AG1478 | 100       |
| ERBB4 (HER4)                   | Tyr 01               | 5                  | 5               | Tyrphostin AG1478 | 124       |
| FER                            | Tyr 05               | 18.5               | 25              | Staurosporine     | 2.21      |
| FES (FPS)                      | Tyr 01               | 30                 | 25              | Staurosporine     | 2.72      |
| FGFR1                          | Tyr 04               | 20                 | 25              | Staurosporine     | 7.78      |
| FGFR2                          | Tyr 04               | 1                  | 5               | Staurosporine     | 2.89      |
| FGFR2 N549H                    | Tyr 04               | 1                  | 5               | Staurosporine     | 13.9      |

| Assay               | Z'-LYTE Substrate | ATP Km app (μM) | ATP Bin (μM) | Inhibitor         | IC50 (nM) |
|---------------------|-------------------|-----------------|--------------|-------------------|-----------|
| FGFR3               | Tyr 04            | 80              | 75           | Staurosporine     | 33.0      |
| FGFR3 K650E         | Tyr 04            | 6               | 5            | Staurosporine     | 7.35      |
| FGFR3 V555M         | Tyr 04            | 2               | 5            | Staurosporine     | 1.14      |
| FGFR4               | Tyr 04            | 162             | 150          | Staurosporine     | 185       |
| FGR                 | Tyr 02            | 10              | 10           | Tyrphostin AG1478 | 664       |
| FLT1 (VEGFR1)       | Tyr 04            | 158             | 150          | Staurosporine     | 24.6      |
| FLT3                | Tyr 02            | 470             | 500          | Tyrphostin AG1478 | 1870      |
| FLT3 D835Y          | Tyr 02            | 18.8            | 25           | Staurosporine     | 0.400     |
| FLT4 (VEGFR3)       | Tyr 04            | 3.8             | 5            | Staurosporine     | 4.22      |
| FRAP1 (mTOR)        | Ser/Thr 11        | 11              | 10           | PI-103            | 30.0      |
| FRK (PTK5)          | Tyr 01            | 51.7            | 50           | Staurosporine     | 10.5      |
| FYN                 | Tyr 02            | 85              | 75           | Tyrphostin AG1478 | 5740      |
| GRK4                | Ser/Thr 16        | 12              | 10           | Staurosporine     | 120       |
| GRK5                | Ser/Thr 16        | 3               | 5            | Staurosporine     | 171       |
| GRK6                | Ser/Thr 16        | 11.7            | 10           | Staurosporine     | 61.2      |
| GRK7                | Ser/Thr 16        | 10.5            | 10           | Staurosporine     | 3.92      |
| GSK3A (GSK3 alpha)  | Ser/Thr 09        | 9               | 10           | Staurosporine     | 14.3      |
| GSK3B (GSK3 beta)   | Ser/Thr 09        | 7               | 10           | Staurosporine     | 12.6      |
| HCK                 | Tyr 02            | 24              | 25           | Tyrphostin AG1478 | 792       |
| HIPK1 (Myak)        | Ser/Thr 09        | 5               | 5            | TBB               | 2290      |
| HIPK2               | Ser/Thr 09        | 17              | 10           | Staurosporine     | 645       |
| HIPK3 (YAK1)        | Ser/Thr 18        | 22              | 25           | TBB               | 3530      |
| HIPK4               | Ser/Thr 18        | 39              | 50           | Staurosporine     | 782       |
| IGF1R               | Tyr 01            | 140             | 150          | Staurosporine     | 170       |
| IKBKB (IKK beta)    | Ser/Thr 05        | 5               | 5            | Staurosporine     | 496       |
| IKBKE (IKK epsilon) | Ser/Thr 11        | 16              | 10           | Staurosporine     | 5.19      |
| INSR                | Tyr 01            | 20              | 25           | Staurosporine     | 77.9      |
| INSRR (IRR)         | Tyr 04            | 55              | 50           | Staurosporine     | 121       |
| IRAK4               | Ser/Thr 07        | 34              | 25           | Staurosporine     | 22.8      |
| ITK                 | Tyr 01            | 5.6             | 5            | Staurosporine     | 44.4      |
| JAK1                | Tyr 06            | 87              | 75           | Staurosporine     | 1.62      |
| JAK2                | Tyr 06            | 31              | 25           | Staurosporine     | 0.555     |
| JAK2 JH1 JH2        | Tyr 06            | 46              | 50           | Staurosporine     | 0.866     |
| JAK2 JH1 JH2 V617F  | Tyr 06            | 49              | 50           | Staurosporine     | 0.835     |
| JAK3                | Tyr 06            | 14              | 10           | Staurosporine     | 0.952     |
| KDR (VEGFR2)        | Tyr 01            | 78              | 75           | Staurosporine     | 6.52      |
| KIT                 | Tyr 06            | 284             | 300          | Staurosporine     | 614       |
| KIT T670I           | Tyr 06            | 220             | 200          | Staurosporine     | 141       |
| KIT V559D           | Tyr 06            | 6               | 5            | Staurosporine     | 7.59      |
| KIT V559D V654A     | Tyr 06            | 2               | 5            | Staurosporine     | 9.50      |
| KIT V560G           | Tyr 06            | 5               | 5            | Staurosporine     | 8.43      |
| KSR2                | Ser/Thr 17        | 15              | 10           | NH125             | 9280      |
| LCK                 | Tyr 02            | 45              | 50           | Tyrphostin AG1478 | 1140      |
| LTK (TYK1)          | Tyr 06            | 82              | 75           | Staurosporine     | 6.97      |
| LYN A               | Tyr 02            | 26              | 25           | Tyrphostin AG1478 | 334       |
| LYN B               | Tyr 02            | 25              | 25           | Tyrphostin AG1478 | 221       |
| MAP2K1 (MEK1)       | Ser/Thr 03        | Cascade         | 100          | Staurosporine     | 2.77 *    |
| MAP2K2 (MEK2)       | Ser/Thr 03        | Cascade         | 100          | Staurosporine     | 3.74 *    |
| MAP2K6 (MKK6)       | Ser/Thr 03        | Cascade         | 100          | Staurosporine     | 4.32 *    |
| MAP3K19 (YSK4)      | Ser/Thr 07        | 5               | 5            | Staurosporine     | 17.1      |
| MAP3K8 (COT)        | Ser/Thr 03        | Cascade         | 100          | Staurosporine     | 28.3 *    |
| MAP3K9 (MLK1)       | Ser/Thr 07        | 73              | 75           | Staurosporine     | 2.89      |
| MAP4K2 (GCK)        | Ser/Thr 07        | 109             | 100          | Staurosporine     | 0.680     |
| MAP4K4 (HGK)        | Ser/Thr 07        | 12.7            | 10           | Staurosporine     | 1.20      |

| Assay                     | Z'-LYTE<br>Substrate | ATP<br>Km app (µM) | ATP<br>Bin (µM) | Inhibitor     | IC50 (nM) |
|---------------------------|----------------------|--------------------|-----------------|---------------|-----------|
| MAP4K5 (KHS1)             | Ser/Thr 07           | 55                 | 50              | Staurosporine | 0.864     |
| MAPK1 (ERK2)              | Ser/Thr 03           | 100                | 100             | Staurosporine | 1970      |
| MAPK10 (JNK3)             | Ser/Thr 04           | Cascade            | 100             | Staurosporine | 1400 *    |
| MAPK11 (p38 beta)         | Ser/Thr 15           | 39                 | 50              | PP2           | 2070      |
| MAPK12 (p38 gamma)        | Ser/Thr 03           | 16                 | 10              | Staurosporine | 150       |
| MAPK13 (p38 delta)        | Ser/Thr 03           | 13                 | 10              | Staurosporine | 285       |
| MAPK14 (p38 alpha)        | Ser/Thr 04           | Cascade            | 100             | Staurosporine | 3140 *    |
| MAPK14 (p38 alpha) Direct | Ser/Thr 15           | 497                | 500             | PP2           | 4610      |
| MAPK3 (ERK1)              | Ser/Thr 03           | 45                 | 50              | Staurosporine | 2130      |
| MAPK7 (ERK5)              | Ser/Thr 04           | 17                 | 10              | NH125         | 8470      |
| MAPK8 (JNK1)              | Ser/Thr 04           | Cascade            | 100             | Staurosporine | 1520 *    |
| MAPK9 (JNK2)              | Ser/Thr 04           | Cascade            | 100             | Staurosporine | 2610 *    |
| MAPKAPK2                  | Ser/Thr 04           | 2.5                | 5               | Staurosporine | 542       |
| MAPKAPK3                  | Ser/Thr 04           | 200                | 200             | Staurosporine | 82400     |
| MAPKAPK5 (PRAK)           | Ser/Thr 04           | 14                 | 10              | Staurosporine | 623       |
| MARK1 (MARK)              | Ser/Thr 21           | 7                  | 5               | Staurosporine | 26.1      |
| MARK2                     | Ser/Thr 21           | 12                 | 10              | Staurosporine | 28.8      |
| MARK3                     | Ser/Thr 25           | 7                  | 5               | Staurosporine | 0.750     |
| MARK4                     | Ser/Thr 25           | 16                 | 10              | Staurosporine | 0.828     |
| MATK (HYL)                | Tyr 01               | 350                | 300             | Staurosporine | 708       |
| MELK                      | Ser/Thr 17           | 30                 | 25              | Staurosporine | 2.01      |
| MERTK (cMER)              | Tyr 02               | 15                 | 10              | Staurosporine | 19.3      |
| MET (cMet)                | Tyr 06               | 64                 | 50              | Staurosporine | 104       |
| MET (cMet) Y1235D         | Tyr 06               | 31                 | 25              | Staurosporine | 197       |
| MET M1250T                | Tyr 06               | 14.1               | 10              | Staurosporine | 144       |
| MINK1                     | Ser/Thr 07           | 26.5               | 25              | Staurosporine | 1.73      |
| MKNK1 (MNK1)              | Ser/Thr 07           | 103                | 100             | Staurosporine | 214       |
| MST1R (RON)               | Tyr 06               | 10.7               | 10              | Staurosporine | 78.7      |
| MST4                      | Ser/Thr 07           | 28                 | 25              | Staurosporine | 4.70      |
| MUSK                      | Tyr 04               | 49.5               | 50              | Staurosporine | 8.21      |
| MYLK2 (skMLCK)            | Ser/Thr 13           | 310                | 300             | Staurosporine | 109       |
| NEK1                      | Ser/Thr 07           | 118.7              | 100             | Staurosporine | 88.1      |
| NEK2                      | Ser/Thr 07           | 150                | 150             | Staurosporine | 5150      |
| NEK4                      | Ser/Thr 17           | 56.2               | 50              | Staurosporine | 413       |
| NEK6                      | Ser/Thr 22           | 108                | 100             | Chetomin      | 182.3     |
| NEK7                      | Ser/Thr 22           | 62                 | 50              | Chetomin      | 233       |
| NEK9                      | Ser/Thr 07           | 90                 | 100             | Staurosporine | 1540      |
| NIM1K                     | Ser/Thr 25           | 11                 | 10              | Staurosporine | 426       |
| NTRK1 (TRKA)              | Tyr 01               | 425                | 400             | Staurosporine | 4.51      |
| NTRK2 (TRKB)              | Tyr 01               | 22                 | 25              | Staurosporine | 0.627     |
| NTRK3 (TRKC)              | Tyr 01               | 54                 | 50              | Staurosporine | 1.74      |
| PAK1                      | Ser/Thr 19           | 48.5               | 50              | Staurosporine | 1.25      |
| PAK2 (PAK65)              | Ser/Thr 20           | 89                 | 75              | Staurosporine | 6.50      |
| PAK3                      | Ser/Thr 20           | 101                | 100             | Staurosporine | 6.94      |
| PAK4                      | Ser/Thr 20           | 3                  | 5               | Staurosporine | 5.02      |
| PAK6                      | Ser/Thr 20           | 8                  | 10              | Staurosporine | 4.66      |
| PAK7 (KIAA1264)           | Ser/Thr 20           | 4                  | 5               | Staurosporine | 6.71      |
| PASK                      | Ser/Thr 07           | 53.3               | 50              | Staurosporine | 62.2      |
| PDGFRA (PDGFR alpha)      | Tyr 04               | 9                  | 10              | Staurosporine | 5.16      |
| PDGFRA D842V              | Tyr 04               | 5.3                | 5               | Staurosporine | 5.58      |
| PDGFRA T674I              | Tyr 04               | 93                 | 100             | Staurosporine | 4.01      |
| PDGFRA V561D              | Tyr 04               | 48                 | 50              | Staurosporine | 1.30      |
| PDGFRB (PDGFR beta)       | Tyr 04               | 100                | 100             | Staurosporine | 5.68      |
| PDK1                      | Ser/Thr 06           | Cascade            | 100             | Staurosporine | 8.75 *    |

| Assay                   | Z'-LYTE Substrate | ATP Km app (μM) | ATP Bin (μM) | Inhibitor     | IC50 (nM) |
|-------------------------|-------------------|-----------------|--------------|---------------|-----------|
| PDK1 Direct             | Ser/Thr 07        | 27              | 25           | Staurosporine | 7.57      |
| PEAK1                   | Tyr 01            | 56              | 50           | Staurosporine | 11.4      |
| PHKG1                   | Ser/Thr 07        | 63.3            | 75           | Staurosporine | 0.689     |
| PHKG2                   | Ser/Thr 07        | 10              | 10           | Staurosporine | 1.52      |
| PIM1                    | Ser/Thr 07        | 407             | 400          | TBB           | 1320      |
| PIM2                    | Ser/Thr 07        | 3               | 5            | Staurosporine | 25.3      |
| PIM3                    | Ser/Thr 07        | 818             | 1000         | Staurosporine | 3.62      |
| PKN1 (PRK1)             | Ser/Thr 07        | 40              | 50           | Staurosporine | 3.62      |
| PLK1                    | Ser/Thr 16        | 12.8            | 10           | Staurosporine | 636       |
| PLK2                    | Ser/Thr 16        | 29.6            | 25           | Staurosporine | 361       |
| PLK3                    | Ser/Thr 16        | 47.8            | 50           | TBB           | 1950      |
| PRKACA (PKA)            | Ser/Thr 01        | 4               | 5            | Staurosporine | 1.98      |
| PRKCA (PKC alpha)       | Ser/Thr 07        | 37              | 25           | Staurosporine | 1.23      |
| PRKCB1 (PKC beta I)     | Ser/Thr 07        | 250             | 200          | Staurosporine | 1.77      |
| PRKCB2 (PKC beta II)    | Ser/Thr 07        | 225             | 200          | Staurosporine | 0.971     |
| PRKCD (PKC delta)       | Ser/Thr 07        | 30              | 25           | Staurosporine | 0.743     |
| PRKCE (PKC epsilon)     | Ser/Thr 07        | 35              | 25           | Staurosporine | 4.95      |
| PRKCG (PKC gamma)       | Ser/Thr 07        | 25              | 25           | Staurosporine | 1.14      |
| PRKCH (PKC eta)         | Ser/Thr 07        | 35              | 25           | Staurosporine | 2.34      |
| PRKCI (PKC iota)        | Ser/Thr 07        | 25              | 25           | Staurosporine | 169       |
| PRKCN (PKD3)            | Ser/Thr 17        | 26              | 25           | Staurosporine | 1.68      |
| PRKCQ (PKC theta)       | Ser/Thr 07        | 100             | 100          | Staurosporine | 2.43      |
| PRKCZ (PKC zeta)        | Ser/Thr 07        | 4               | 5            | Staurosporine | 546       |
| PRKD1 (PKC mu)          | Ser/Thr 17        | 15              | 10           | Staurosporine | 3.97      |
| PRKD2 (PKD2)            | Ser/Thr 17        | 27              | 25           | Staurosporine | 2.37      |
| PRKG1                   | Ser/Thr 01        | 20              | 25           | Staurosporine | 4.25      |
| PRKG2 (PKG2)            | Ser/Thr 01        | 177             | 150          | Staurosporine | 0.871     |
| PRKX                    | Ser/Thr 01        | 17              | 10           | Staurosporine | 1.87      |
| PTK2 (FAK)              | Tyr 07            | 45              | 50           | Staurosporine | 53.3      |
| PTK2B (FAK2)            | Tyr 01            | 5               | 5            | Staurosporine | 13.0      |
| PTK6 (Brk)              | Tyr 01            | 82              | 75           | Staurosporine | 974       |
| RAF1 (cRAF) Y340D Y341D | Ser/Thr 03        | Cascade         | 100          | Staurosporine | 45.8 *    |
| RET                     | Tyr 02            | 11              | 10           | Staurosporine | 3.10      |
| RET A883F               | Tyr 04            | 10              | 10           | Staurosporine | 12.5      |
| RET S891A               | Tyr 06            | 8               | 10           | Staurosporine | 0.856     |
| RET V804E               | Tyr 04            | 50              | 50           | Staurosporine | 2.25      |
| RET V804L               | Tyr 02            | 5               | 5            | Staurosporine | 2.07      |
| RET Y791F               | Tyr 02            | 12              | 10           | Staurosporine | 3.22      |
| ROCK1                   | Ser/Thr 07        | 3.1             | 5            | Staurosporine | 3.36      |
| ROCK2                   | Ser/Thr 13        | 39.5            | 50           | Staurosporine | 3.07      |
| ROS1                    | Tyr 01            | 61.4            | 50           | Staurosporine | 2.36      |
| RPS6KA1 (RSK1)          | Ser/Thr 06        | 6.5             | 5            | Staurosporine | 0.778     |
| RPS6KA2 (RSK3)          | Ser/Thr 06        | 8               | 10           | Staurosporine | 0.717     |
| RPS6KA3 (RSK2)          | Ser/Thr 06        | 18              | 10           | Staurosporine | 0.339     |
| RPS6KA4 (MSK2)          | Ser/Thr 01        | 14.1            | 10           | Staurosporine | 4.21      |
| RPS6KA5 (MSK1)          | Ser/Thr 01        | 39              | 50           | Staurosporine | 2.66      |
| RPS6KA6 (RSK4)          | Ser/Thr 20        | 30              | 25           | Staurosporine | 0.378     |
| RPS6KB1 (p70S6K)        | Ser/Thr 07        | 17              | 10           | Staurosporine | 2.87      |
| RPS6KB2 (p70S6Kb)       | Ser/Thr 19        | 44              | 50           | Staurosporine | 5.91      |
| SBK1                    | Ser/Thr 25        | 3               | 5            | Staurosporine | 45.2      |
| SGK (SGK1)              | Ser/Thr 06        | 36              | 25           | Staurosporine | 11.8      |
| SGK2                    | Ser/Thr 06        | 50              | 50           | Staurosporine | 62.8      |
| SGKL (SGK3)             | Ser/Thr 06        | 12              | 10           | Staurosporine | 69.9      |
| SNF1LK2                 | Ser/Thr 25        | 59              | 50           | Staurosporine | 0.745     |

| Assay            | Z'-LYTE<br>Substrate | ATP<br>Km app (μM) | ATP<br>Bin (μM) | Inhibitor         | IC50 (nM) |
|------------------|----------------------|--------------------|-----------------|-------------------|-----------|
| SRC              | Tyr 02               | 50                 | 50              | Tyrphostin AG1478 | 2100      |
| SRC N1           | Tyr 02               | 50                 | 50              | Tyrphostin AG1478 | 1370      |
| SRMS (Srm)       | Tyr 01               | 126.9              | 150             | Staurosporine     | 339       |
| SRPK1            | Ser/Thr 18           | 32                 | 25              | Staurosporine     | 522       |
| SRPK2            | Ser/Thr 07           | 17.6               | 25              | Staurosporine     | 1780      |
| STK22B (TSSK2)   | Ser/Thr 04           | 3                  | 5               | Staurosporine     | 14.6      |
| STK22D (TSSK1)   | Ser/Thr 04           | 9                  | 10              | Staurosporine     | 0.541     |
| STK23 (MSSK1)    | Ser/Thr 18           | 69                 | 75              | Staurosporine     | 4020      |
| STK24 (MST3)     | Ser/Thr 07           | 56                 | 50              | Staurosporine     | 7.55      |
| STK25 (YSK1)     | Ser/Thr 07           | 83                 | 75              | Staurosporine     | 4.01      |
| STK3 (MST2)      | Ser/Thr 07           | 50                 | 50              | Staurosporine     | 5.40      |
| STK4 (MST1)      | Ser/Thr 07           | 48.9               | 50              | Staurosporine     | 7.35      |
| SYK              | Tyr 02               | 24.8               | 25              | Staurosporine     | 0.576     |
| TAOK2 (TAO1)     | Ser/Thr 07           | 322                | 300             | Staurosporine     | 17.4      |
| TBK1             | Ser/Thr 05           | 35                 | 25              | Staurosporine     | 1.77      |
| TEK (Tie2)       | Tyr 05               | 16.7               | 10              | Staurosporine     | 79.9      |
| TEK (TIE2) Y897S | Tyr 06               | 207                | 300             | Staurosporine     | 268       |
| TNK1             | Ser/Thr 13           | 237                | 200             | Staurosporine     | 4.16      |
| TXK              | Tyr 06               | 96                 | 100             | Staurosporine     | 51.6      |
| TYK2             | Tyr 03               | 23                 | 25              | Staurosporine     | 1.09      |
| TYRO3 (RSE)      | Tyr 02               | 27.7               | 25              | Staurosporine     | 7.78      |
| YES1             | Tyr 02               | 20.3               | 25              | Tyrphostin AG1478 | 963       |
| ZAP70            | Tyr 07               | 2                  | 5               | Staurosporine     | 457       |