

Technology Spotlight and Applications

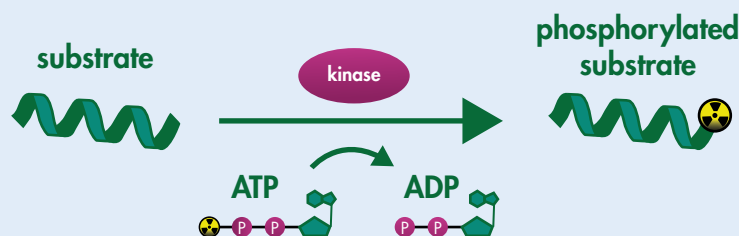
Radiometric Filter Binding Assay

The gold standard for kinase profiling just got better

Background Info

The radiometric filter binding assay is **the gold standard method** to study the activity of your target kinase by directly measuring its phosphorylated substrate products, without the use of modified substrates, coupling enzymes, or detection antibodies.

Radiometric assays are not kinase dependent and **can be applied to any kinase inhibitor** thus representing a truly universal assay that offers **consistent and robust results** with **low background signal**.



**1mM ATP now
available on 340
wild type kinase
targets**

Reaction Biology's radiometric assay protocols are further optimised to deliver best-in-class quality data, minimise error rates and avoid false positive and negatives caused by other assay formats.

Our industry-leading radiometric assay can be run using **physiologically relevant** concentrations of ATP (1mM) in addition to previously available concentrations of 1µM, 10µM, or apparent ATP-K_m up to 250µM.

Why Choose a Radiometric Assay

	Measures Kinase Activity	Detects all types of inhibitors, including substrate-specific inhibitors	Accommodates both peptide and protein substrates	No modified substrates/Additional detection reagents	Universally applicable to all kinases
Radiometric Filter Binding Assay	✓	✓	✓	✓	✓
FRET Peptide	✓	✓	✗	✗	✓
Luminescence	✓	✓	✓	✗	✓
Mobility Shift	✓	✓	✗	✗	✗
Competition Binding	✗	Suboptimal	✗	✗	✗

Why 1mM ATP?

Relevant and meaningful results on cellular signaling and regulatory mechanisms

Selecting 1mM ATP concentration in radiometric kinase assays can provide a more accurate representation of physiological conditions, ensuring that the obtained results are more relevant and meaningful for understanding your compound's kinase inhibitory activity.

Here's why our 1mM ATP kinase screening solution stands out:

- 1 True-to-Life Conditions:** our assays mimic typical intracellular ATP concentrations in mammalian cells, which are 1mM or higher
- 2 Maximize Your Kinase Inhibitor Potential:** our conditions guarantee an accurate estimation of your target's kinase inhibitory activity and kinetics.
- 3 Stability Guaranteed:** physiological ATP concentrations are required to maintain some kinase's stability, leading to accurate assay results.
- 4 Relevance to Cell and *In Vivo* Data:** obtain quick biochemical data applicable to follow up cell-based testing and *in vivo* studies

Our Solution

HotSpot™ Kinase Screening

"The New Gold Standard, Only at Reaction Biology"

Gain an in-depth understanding of your compound's specific activity and selectivity

The largest commercially available human kinase panel of over 700 targets

Using a proprietary radiometric assay optimised for maximal sensitivity and accuracy

Now available at physiologically relevant 1mM ATP for 340 wild type kinases

Discover the Benefits



Flexible

Choose any screening design

- IC₅₀ value determination with 5 or 10 concentrations
- 1 or 2 doses per compound



Fast

- Results in as fast as 10 business days for standard projects
- Run bi-weekly



Quality data

- A kinase-specific control compound included for each in 10-dose IC₅₀



Physiologically relevant

- Now available using 1mM ³³P-γ-ATP on 340 wild type kinase targets

ATP-Max KinomeScreen

A full panel of 340 kinase targets available at 1mM ATP, for comprehensive and deep interrogation of your compound's selectivity against the whole human kinome
Available as a full panel, or select targets to create your own custom panel.

Kinase Family	ATP-MAX Kinome Targets
AG	MSK2/RPS6KA4
AGC	AKT1, AKT2, AKT3, DMPK2, GRK2, GRK3, GRK4, GRK5, GRK6, GRK7, LATS1, LATS2, LYN-B, MAST3, MASTL, MRCKα/CDC42BPA, MRCKβ/CDC42BPB, MSK1/RPS6KA5, P70S6K/RPS6KB1, P70S6KB/RPS6KB2, PDK1/PDPK1, PKA, PKACB, PKACG, PKCα, PKCβ1, PKCβ2, PKCδ, PKCε, PKCη, PKCγ, PKCi, PKCθ, PKCζ, PKG1α, PKG1b, PKG2/PRKG2, PKN1/PRK1, STK32B/YANK2, PRKX, ROCK1, ROCK2, RSK1, RSK2, RSK3, RSK4, SGK1, SGK2, SGK3/SGKL, STK21/CIT, STK38/NDR1, STK38L/NDR2
CAMK	ARK5/NUAK1, BRSK1, BRSK2, CAMK1A, CAMK1B, CAMK1D, CAMK1g, CAMK2a, CAMK2b, CAMK2D, CAMK2G, CAMK4, CHK1, CHK2, DAPK1, DAPK2, DCAMKL1, DCAMKL2, LKB1, MAPKAPK2, MAPKAPK3, MAPKAPK5/PRAK, MARK1, MARK2/PAR-1BA, MARK3, MARK4, MELK, MLCK/MYLK, MLCK2/MYLK2, MNK1, MNK2, MYLK3, MYLK4, NIM1, PASK, PHKG1, PHKG2, PIM1, PIM2, PIM3, PKCMU/PRKD1, PKCNU/PRKD3, PKD2/PRKD2, SIK1, SIK2, SIK3, SNARK/NUAK2, SNRK, STK22D/TSSK1, TSSK2, TSSK3/STK22C, ZIPK/DAPK3
CK1	CK1α1, CK1α1L, CK1δ, CK1ε, CK1γ1, CK1γ2, CK1γ3, TTBK1, TTBK2, VRK2
CMGC	CDK1/CYCLIN A, CDK1/CYCLIN B, CDK1/CYCLIN E, CDK14/CYCLIN Y (PFTK1), CDK15/CYCLIN A2, CDK15/CYCLIN B1, CDK16/CYCLIN Y (PCTAIRE), CDK17/CYCLIN Y (PCTK2), CDK18/CYCLIN Y (PCTK3), CDK2/CYCLIN A, CDK2/CYCLIN A1, CDK2/CYCLIN E, CDK2/CYCLIN E2, CDK2/CYCLIN O, CDK3/CYCLIN C, CDK3/CYCLIN E, CDK3/CYCLIN E2, CDK4/CYCLIN D1, CDK4/CYCLIN D2, CDK4/CYCLIN D3, CDK5/P25, CDK5/P35, CDK6/CYCLIN D1, CDK6/CYCLIN D2, CDK6/CYCLIN D3, CDK7/CYCLIN H, CDK9/CYCLIN K, CDK9/CYCLIN T1, CDK9/CYCLIN T2, CK2A, CK2A2, CLK1, CLK2, CLK3, CLK4, DYRK1/DYRK1A, DYRK1B, DYRK2, DYRK3, ERK1, ERK2/MAPK1, ERK5/MAPK7, ERK7/MAPK15, GSK3A, GSK3B, HIPK1, HIPK2, HIPK3, HIPK4, JNK1, JNK2, JNK3, LCK2/ICK, MAK, MSSK1/STK23, P38A/MAPK14, P38B/MAPK11, P38d/MAPK13, P38G, SRPK1, SRPK2
STE	ASK1/MAP3K5, COT1/MAP3K8, GCK/MAP4K2, GLK/MAP4K3, HGK/MAP4K4, HPK1/MAP4K1, KHS/MAP4K5, LOK/STK10, MEK1, MEK3, MEK5, MEKK1, MEKK2, MEKK3, MINK/MINK1, MKK4, MKK6, MKK7, MST1/STK4, MST2/STK3, MST3/STK24, MYO3A, MYO3B, OSR1/OXSR1, PAK1, PAK2, PAK3, PAK4, PAK5, PAK6, SLK/STK2, STK25/YSK1, STK39/STLK3, TAOK1, TAOK2/TAO1, TAOK3/JIK, TNIK, YSK4/MAP3K19
TK	ABL1, ABL2/ARG, ACK1, ALK, AXL, BLK, BMX/ETK, BRK, BTK, C-KIT, C-MER, C-MET, CSK, C-SRC, CTK/MATK, DDR2, EGFR, EPHA1, EPHA2, EPHA3, EPHA4, EPHA5, EPHA6, EPHA7, EPHA8, EPHB1, EPHB2, EPHB3, EPHB4, ERBB2/HER2, ERBB4/HER4, FAK/PTK2, FER, FES/FPS, FGFR1, FGFR2, FGFR3, FGFR4, FGR, FLT1/VEGFR1, FLT3, FLT4/VEGFR3, FMS, FRK/PTK5, FYN, HCK, IGF1R, IR, IRR/INSRR, ITK, JAK1, JAK2, JAK3, KDR/VEGFR2, LCK, LYN, MUSK, PDGFRA, PDGFRB, PYK2, RET, RON/MST1R, ROS/ROS1, SRMS, SYK, TEC, TIE2/TEK, TRKA, TRKB, TRKC, TXK, TYK1/LTK, TYK2, TYRO3/SKY, YES/YES1, ZAP70
TKL	ALK4/ACVR1B, ALK5/TGFBF1, ALK6/BMPR1B, ARAF, BMPR2, BRAF, IRAK1, IRAK4, LIMK1, LIMK2, LRRK2, MLK1/MAP3K9, MLK2/MAP3K10, MLK3/MAP3K11, MLK4, PBK/TOPK, RAF1, RIPK2, RIPK4, RIPK5, TESK1, TESK2, TGFβR2, ZAK/MLTK
Other	AUR-A, AUR-B, AUR-C, CAMKK2, ERN1/IRE1, ERN2/IRE2, HASPIN, IKKβ/IKBKB, IKKε/IKBKE, NEK1, NEK2, NEK3, NEK4, NEK5, NEK6, NEK7, NEK9, PLK1, PLK2, PLK3, TBK1, TLK1, TLK2, ULK1, ULK2, ULK3, WNK3

ATP-Max Diversify

A tailored panel of representative isoforms from each major kinase family available at 1mM ATP, for quick understanding of your compound's selectivity

Kinase Family	ATP-Max Diversify targets
AGC	MAST3, ROCK1, PKA α , PKC α , PKC θ , Akt1/PKB α , RSK1, p70S6K, MSK2
CAMK	caMLCK, DAPK2, TSSK1, Pim3, Chk1, ARK5, MARK2, MNK2, CAMK1 α , DCAMKL1
CK1	CK1 α , CK2 α , CK1 γ 1
CMGC	ERK1, p38 α , CDK4, JNK3, CDK7, CDK9, CDK1, GSK3 β , CLK2, HIPK2, DYRK1A
TKL	Mlk1, ALK4, BRAF, TGF β R2, LIMk1, IRAK4
TK	Abl1, BTK, TEC, Lyn, Src, EphA5, EphB2, IGF1R, TrkA, Met, Ret, FGFR2, FLT1/VEGFR1, Kit, FLT3, EGFR, Jak2
STE	MST1, LOK, PAK4, MEK1/MAP2K1, MEKK1
Lipid Kinases	PI3K α , PI3K β , PI3K δ , PI3K γ
Atypical Kinases	AurA, Nek1, ULK1, IKK ϵ , PLK1

Discuss your project directly with our kinase experts



v1.0 | Feb 2024

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