10 µM ATP Transcreener™ KINASE Assay Tolerance Results

The highest concentration of additive that is within 3 standard deviations for both 0% and 10% conversion is listed below (5 hour read).

The tolerances listed below represent the concentration of additive in the final Transcreener™ Assay volume. (10 µL Kinase reaction + 10 µL Stop and Detect Buffer.)

solvents	
acetonitrile	10%
DMSO	20%
DMF	6%
ethanol	16%
methanol	6%
glycerol	6%

detergents	
Brij-35	0.3%
CHAPS	0.04%
NP40	0.04%
SDS	0.02%
Triton X-100	0.04%
sodium deoxycholate	0.04%
sodium lauroyl sarcosine	0.04%

metal chelates	
EDTA	30 mM
EGTA	125 mM

reductants	
beta mercaptoethanol	1.5%
dithiothreitol	250 mM

salts	
ammonium acetate	250 mM
ammonium sulfate	2 mM
calcium chloride (CaCl2)	8 mM
magnesium acetate (MgAc)	8 mM
magnesium chloride (MgCl2)	8 mM
magnesium sulfate (MgSO4)	2 mM
manganese chloride (MnCl2)	8 mM
potassium chloride (KCl)	125 mM
sodium azide (NaN3)	0.6%
sodium bromide (NaBr)	60 mM
sodium chloride (NaCl)	150 mM

Phosphatase inhibitors	
glycerol phosphate	2 mM
imidazole	60 mM
sodium fluoride (NaF)	500 mM
sodium molybdate (Na2Mo4)	15 mM
sodium tartrate	100 mM
sodium orthovanadate (NaVO4)	8 mM
sodium pyrophosphate	0.4 mM

carrier proteins/coactivators	
BSA	0.5 mg/mL
BGG	0.3 mg/mL
calmodulin	2.5 mg/mL

Compatible Kinase Buffers
50 mM HEPES, pH 6.5
50 mM HEPES, pH 7.0
50 mM HEPES, pH 7.5
50 mM HEPES, pH 8.0
50 mM HEPES, pH 8.5
50 mM Tris, pH 7.5
50 mM Tris, pH 8.0
50 mM Tris, pH 8.5
50 mM Tris, pH 9.0
50 mM MOPS, pH 7.0
50 mM Imidazole, pH 7.1

recommended buffer (in protocol)

This list shows that the kinase reaction can be performed with various buffers and at various pHs (without affecting the shift at 10% conversion). This is not a comprehensive list of compatible buffers, but rather a sample of common buffers which proved to be compatible.

These buffers give 10% conversion shift within 3 standard deviations relative to the recommended kinase buffer in the protocol (5 hour read).