

## SUPPORTING INFORMATION

### Global analysis of human non-receptor tyrosine kinase specificity using high-density peptide microarrays

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**Supplementary Dataset 1. Quantified microarray data for all kinases analyzed.** Spot intensities were quantified from phosphor imager scans of microarray slides using QuantityOne software (BioRad). Data were normalized so that the average value of all amino acids within a single position was assigned a value of 1. Normalized data for two separate microarray runs were averaged.

**Table S1. Expression systems used to generate kinases for microarray analysis.** Kinases produced in HEK293T cells were expressed as N-terminal GST fusion proteins and purified by glutathione affinity chromatography as described in the main text. Kinases produced in insect cells or bacteria were expressed as hexahistidine-tagged proteins and purified by immobilized metal affinity chromatography. Where indicated, kinases were activated by treating transfected HEK293T cells with the general tyrosine phosphatase inhibitor pervanadate prior to lysis.

Kinase	Expression system	Notes
ABL	Insect cells	
ACK	HEK293T cells	
ARG	HEK293T cells	
BLK	HEK293T cells	
BMX	HEK293T cells	Activated with pervanadate
BRK	HEK293T cells	Activated with pervanadate
BTK	HEK293T cells	
CSK	Insect cells	
FER	HEK293T cells	
FES	Bacteria	
FGR	HEK293T cells	
FRK	HEK293T cells	
FYN	HEK293T cells	
HCK	HEK293T cells	
JAK1	Insect cells	
JAK2	Insect cells	Commercial source
JAK3	Insect cells	
LCK	Insect cells	Commercial source
LYN	HEK293T cells	
PYK2	HEK293T cells	
SRC	HEK293T cells	
SRM	HEK293T cells	
SYK	HEK293T cells	
TEC	HEK293T cells	Activated with pervanadate
TXK	Insect cells	Commercial source
YES	HEK293T cells	