

Supplementary Information for

PTMNavigator: Interactive Visualization of Differentially Regulated Post-Translational Modifications in Cellular Signaling Pathways

Julian Müller¹, Florian P. Bayer¹, Mathias Wilhelm², Maximilian G. Schuh^{1†}, Bernhard Kuster^{1,3}, and Matthew The^{1*}

¹Proteomics and Bioanalytics, School of Life Sciences, Technical University of Munich; 85354 Freising, Germany.

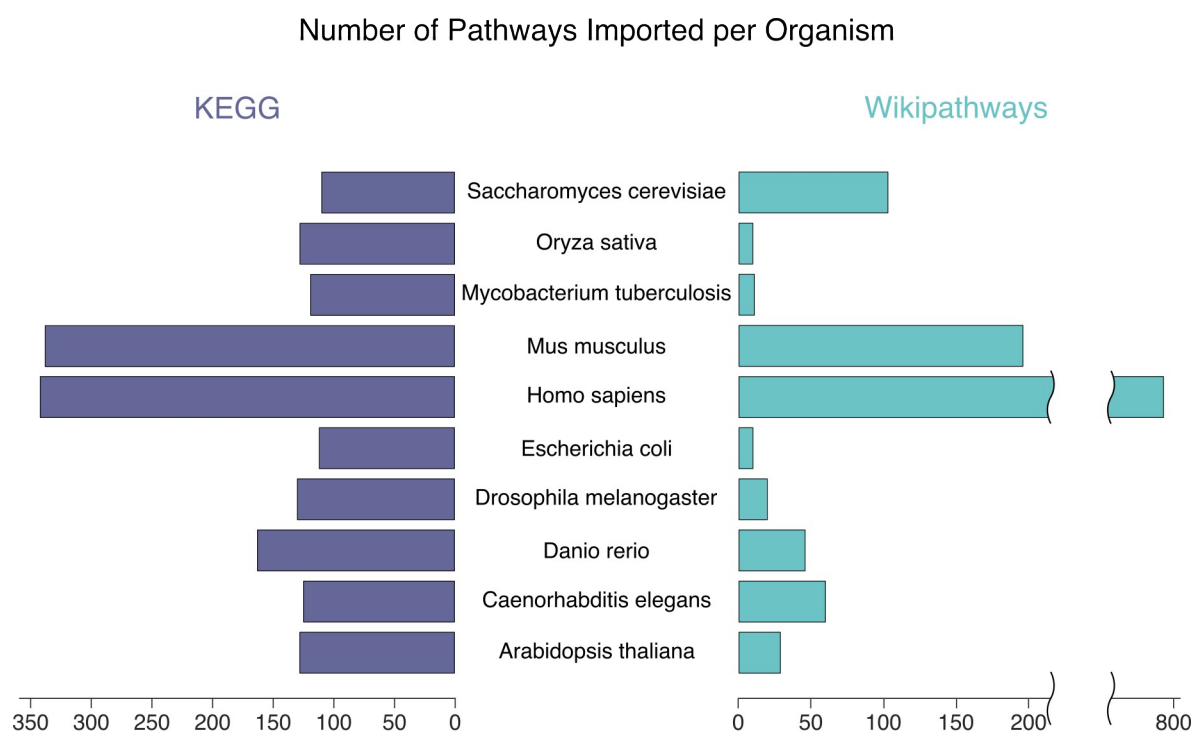
²Computational Mass Spectrometry, School of Life Sciences, Technical University of Munich; 85354 Freising, Germany.

³German Cancer Consortium (DKTK), Partner Site Munich, 80336 Munich, Germany.

[†]Present Address: Organic Chemistry II, School of Natural Sciences, Technical University of Munich; 85748 Garching, Germany.

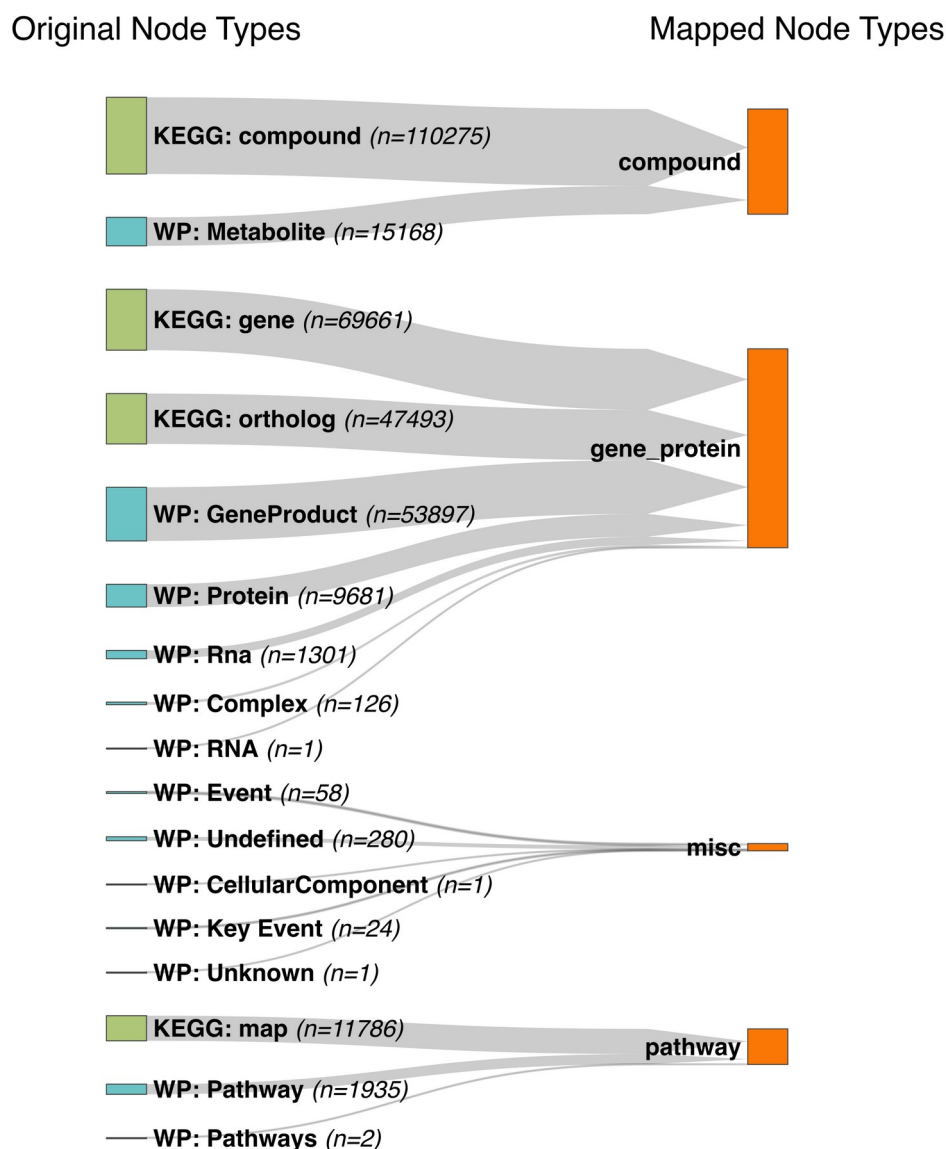
*Corresponding author.

This PDF contains Supplementary Figures 1 to 10.



Supplementary Figure 1. Number of pathways per organism and reference database that were imported into ProteomicsDB and are available in PTMNavigator.

Source data are provided as a Source Data file.

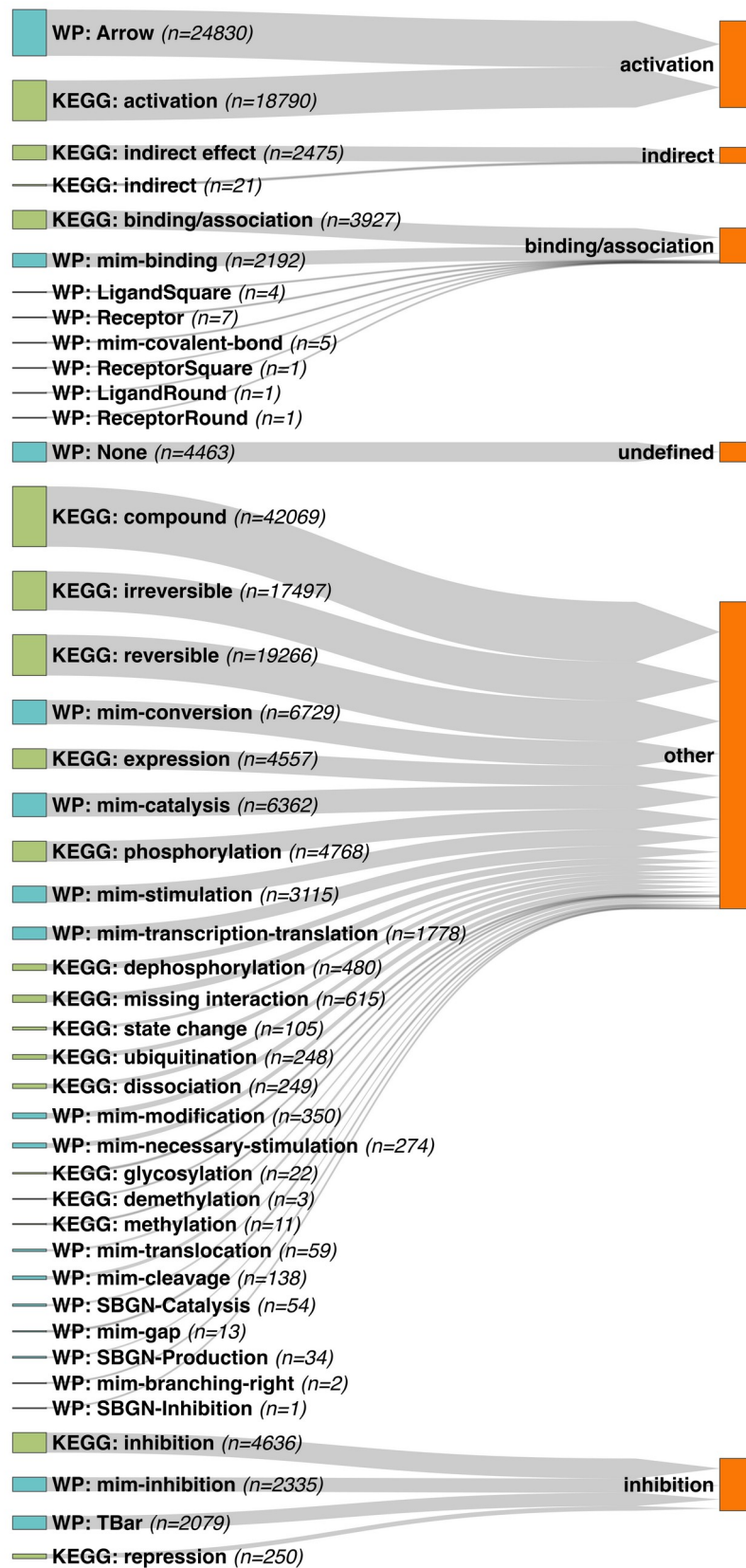


Supplementary Figure 2. Mapping of Node Types from KEGG and WikiPathways to PTMNavigator.

On the left side all node types are listed that appear in the KEGG KGML or WikiPathways GPML files of the ten imported organisms. On the right side are the 4 node types that are defined in PTMNavigator. Source data are provided as a Source Data file.

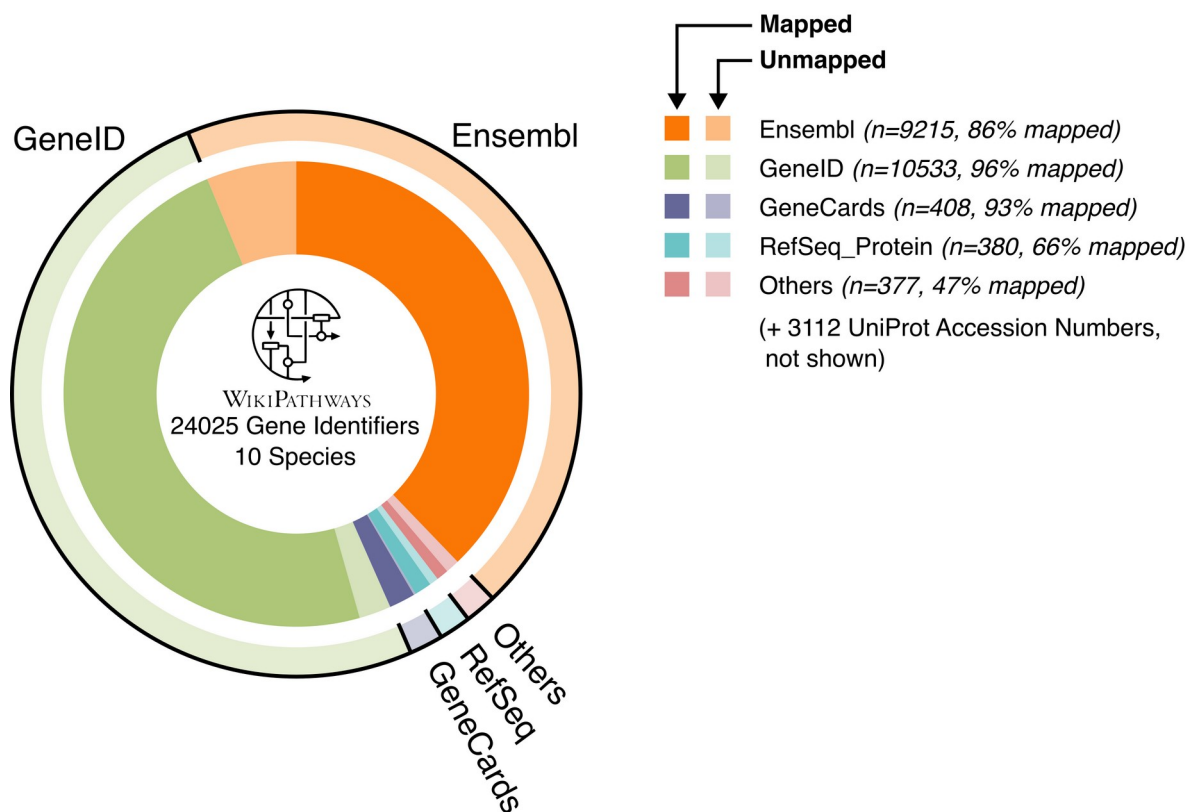
Original Edge Types

Mapped Edge Types



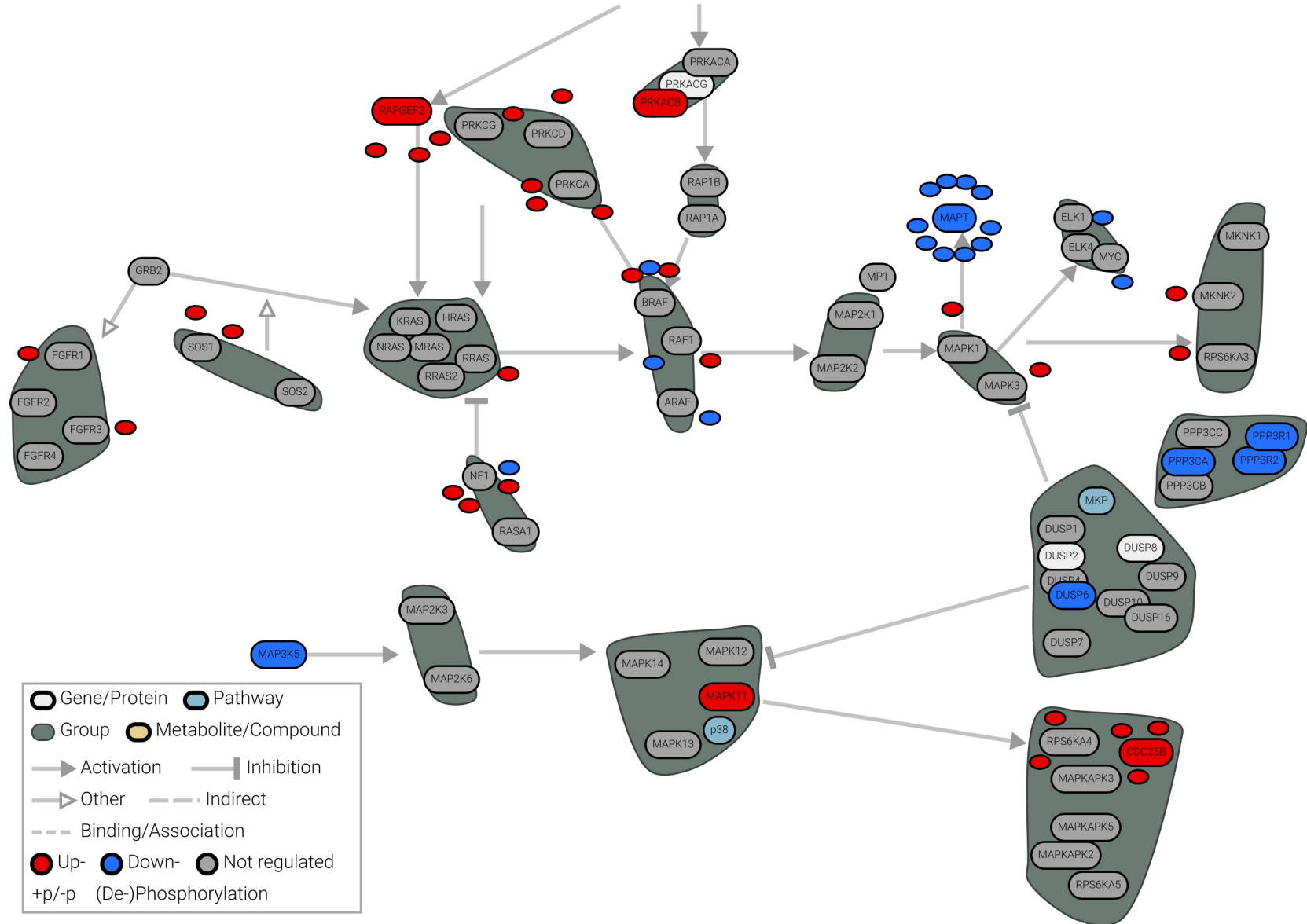
Supplementary Figure 3. Mapping of Edge Types from KEGG and WikiPathways to PTMNavigator.

On the left side all edge types are listed that appear in the KEGG KGML or WikiPathways GPML files of the ten imported organisms. The right side shows how they are mapped to the internal JSON representation in ProteomicsDB. Raw edge types are converted either to 'activation', 'inhibition', 'binding/association', 'indirect' or 'undefined'. The other edges are not converted and retain their original type (in the figure, these edge types are summarized as 'other'). Source data are provided as a Source Data file.



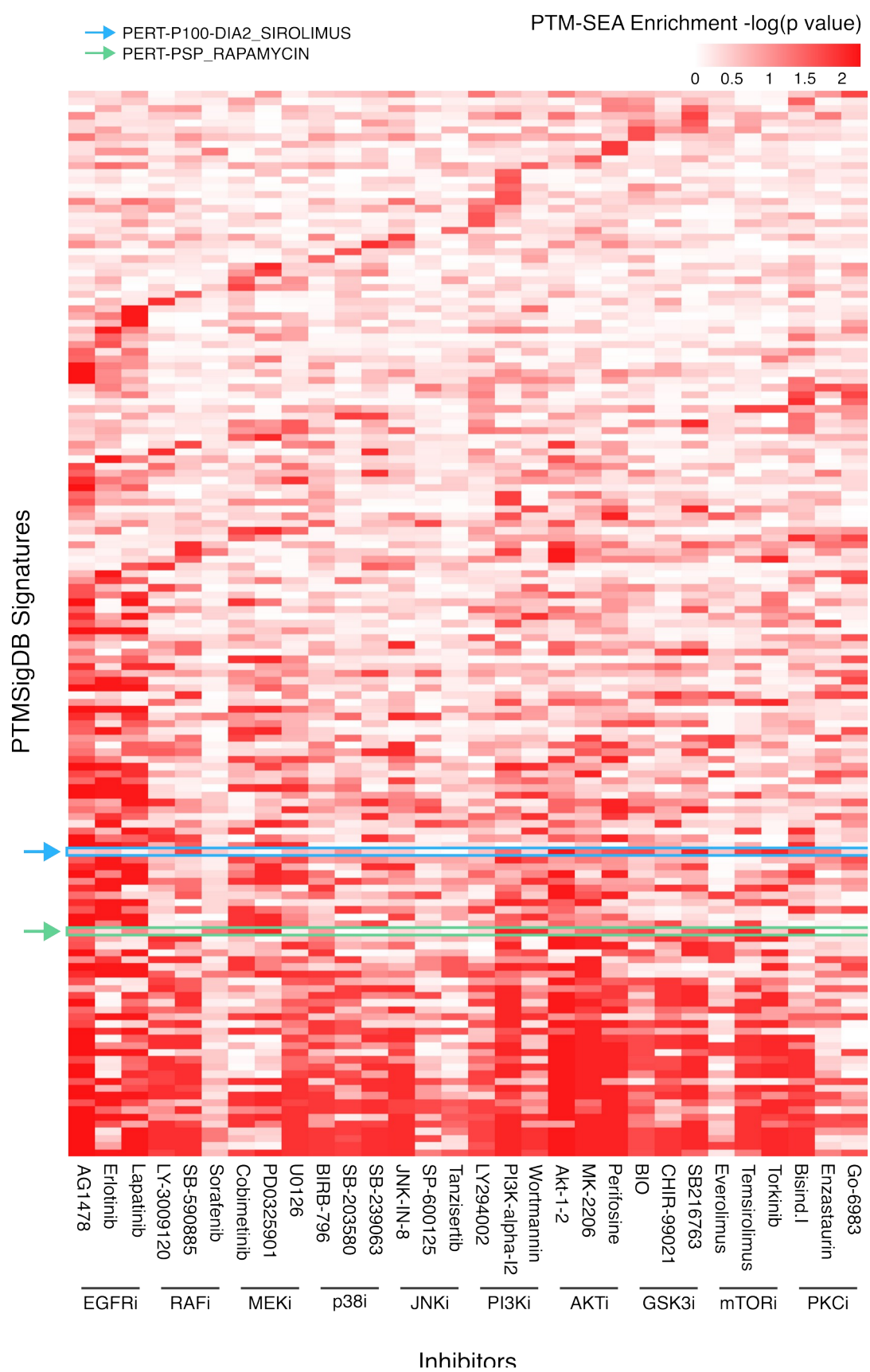
Supplementary Figure 4. Gene Identifier Conversion for WikiPathways Diagrams.

WikiPathways uses 9 different gene identifier types in the diagrams processed in this study. The UniProt API was used to map them to UniProt Accession Numbers. Overall, 91% of gene identifiers (21956 of 24025) could be mapped (or were already in UniProt format). 'Ensembl' summarizes identifiers of type 'Ensembl' and 'Ensembl_Transcript'. 'Others' summarizes 'KEGG', 'EC', and 'HGNC' identifiers. Source data are provided as a Source Data file. The WikiPathways Logo is licensed under the Creative Commons BY-SA license (<https://creativecommons.org/licenses/by-sa/4.0/deed.en>) by Alexander Pico (<https://www.wikipathways.org/authors/AlexanderPico.html>).



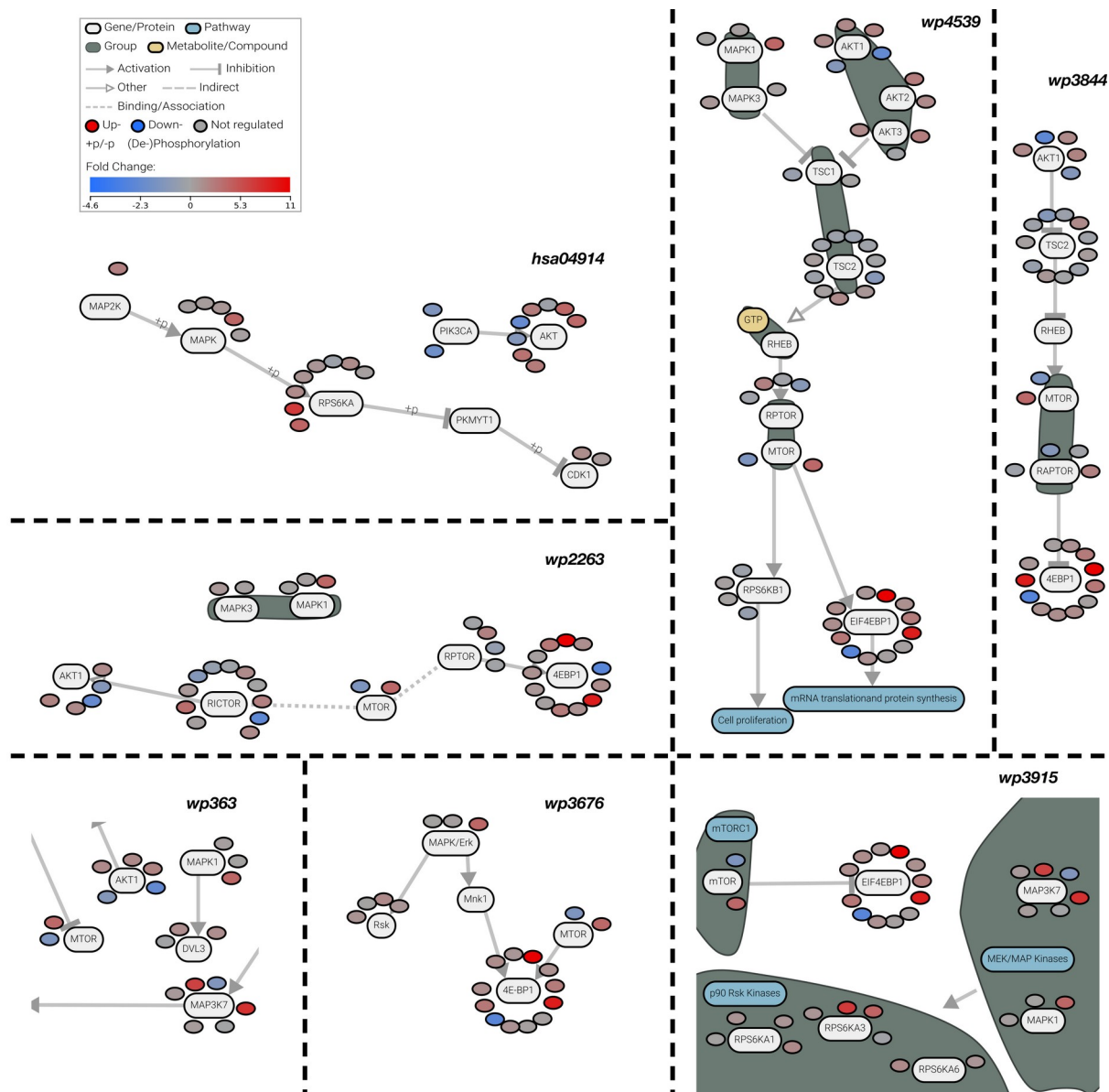
Supplementary Figure 5. Visualization of protein-level and peptide-level data in PTMNavigator.

Comparison of baseline proteomes and phosphoproteomes of the cell lines A204 and RDES projected onto the diagram 'MAPK Signaling - WP382'. Colors indicate whether expression is higher in A204 (red) or RDES (blue). Grey color indicates peptides/proteins that were not sufficiently different between the two cell lines (as thresholds, a difference of 3 was used for proteomic and 4 for phosphoproteomic data). White protein nodes indicate proteins that were not measured in the experiment. Dataset generated by Lee et al. Source data are provided as a Source Data file.



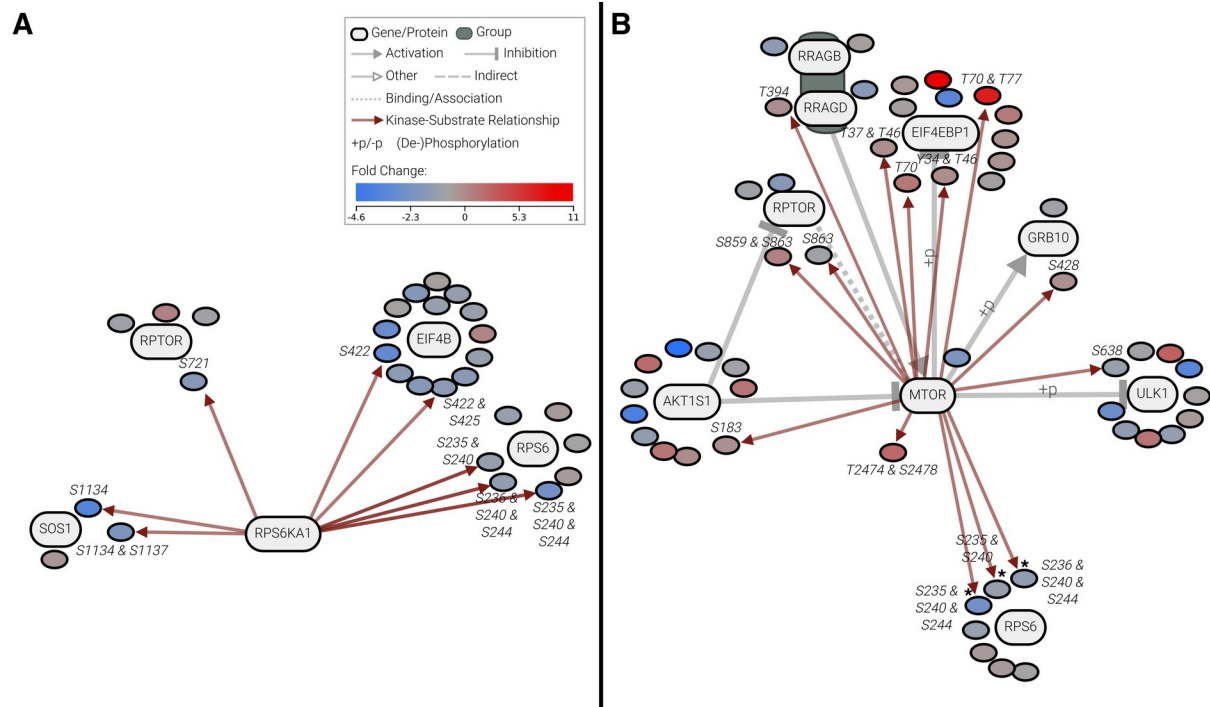
Supplementary Figure 6. Results of PTM-SEA analysis of the Bekker-Jensen dataset.

Shown are signatures with $p\text{-value} < 0.05$, sorted by the number of datasets in which they were reported as enriched (larger numbers on the bottom). Inhibitors are grouped by their designated targets. Source data are provided as a Source Data file.



Supplementary Figure 7. Common subnetworks of the high-scoring pathways in the Go-6983 dataset.

Edited Screenshots from PTMNavigator showing excerpts from 7 different canonical pathway diagrams. All of these pathways showed significant enrichment in the gene-centric-redundant ssGSEA. PTM nodes are colored according to their fold change. Edges labeled with '+p' indicate a phosphorylation of the target protein. Source data are provided as a Source Data file.

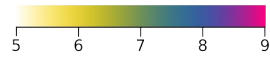


Supplementary Figure 8. PTMNavigator makes PTM-SEA and kinase activity scores transparent.

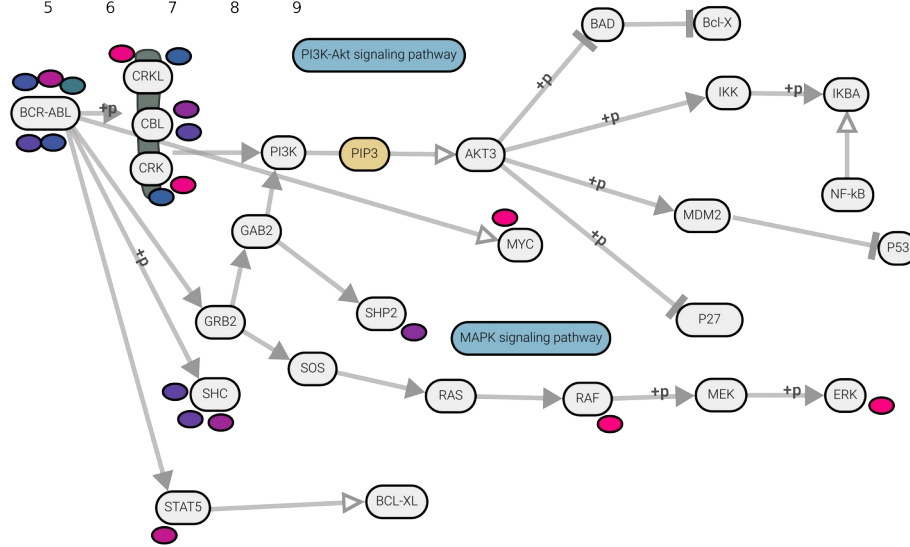
Edited Screenshots from PTMNavigator showing excerpts from the projection of the Go-6983 dataset onto the pathway ‘mTOR Signaling Pathway - hsa04150’. Shown are the proteins phosphorylated by RPS6KA1 (A) and mTOR (B). Red arrows indicate kinase-substrate annotations from PhosphoSitePlus. Both PTM-SEA and KSEA reported a downregulation of RPS6KA1 activity as well as an upregulation of mTOR activity. The visualizations of PTMNavigator confirm this, with all annotated RPS6KA1 substrates in the diagram showing reduced expression, whereas most mTOR substrates showed increased expression. Notably, the sites on RPS6 (indicated by asterisks “”) are not exclusively associated with mTOR, but are also substrates of RPS6KA1, RPS6KA3, and PRKCD. Edges labeled with ‘+p’ indicate a phosphorylation of the target protein. Source data are provided as a Source Data file.*

hsa05220 - Chronic Myeloid Leukemia

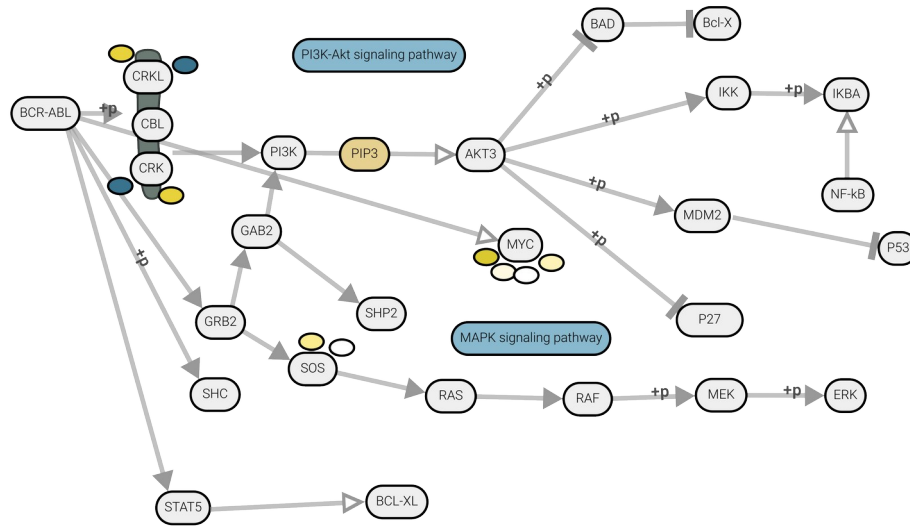
pEC50 [M]:



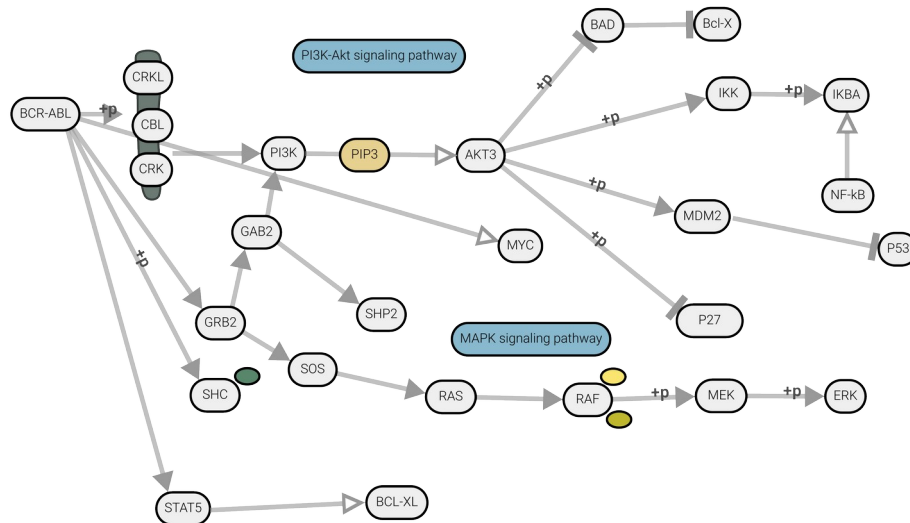
K562 Cells



A431 Cells

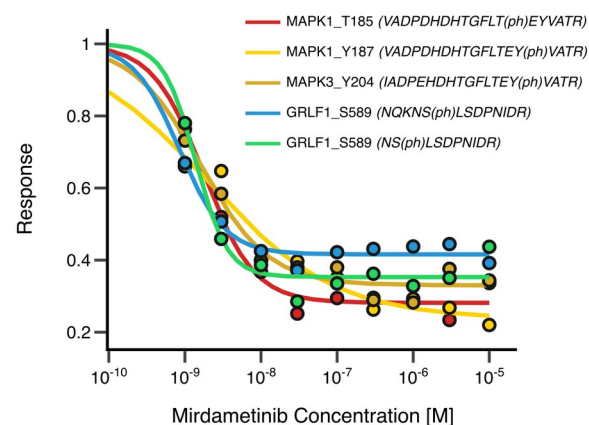
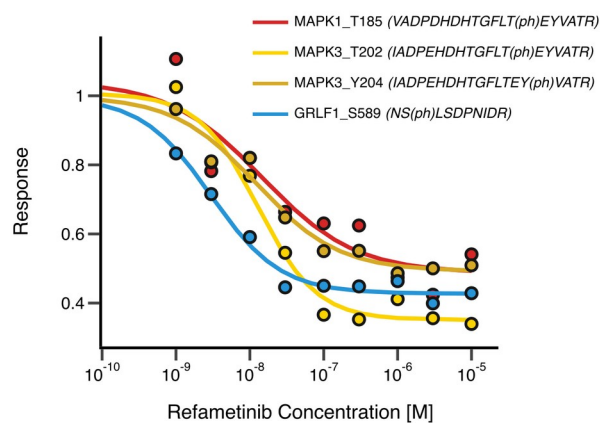
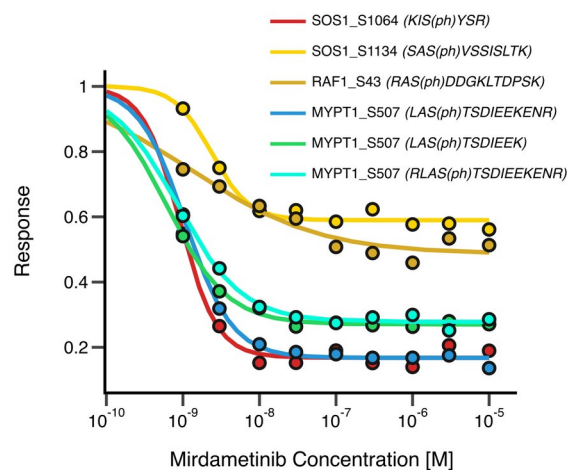
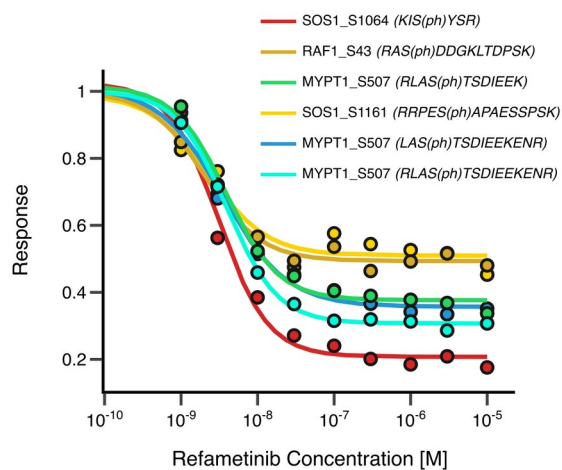


A549 Cells



Supplementary Figure 9. Comparison of dose-dependent Dasatinib treatment in three different cell lines.

Edited screenshots from PTMNavigator. Projection of data from Zecha et al. onto the KEGG diagram 'hsa05220 – Chronic Myeloid Leukemia'. Edges labeled with '+p' indicate a phosphorylation of the target protein. Top: K562 lymphoblast cells. Middle: A431 epidermoid carcinoma epithelial cells. Bottom: A549 lung adenocarcinoma cells. Source data are provided as a Source Data file.



Supplementary Figure 10. decryptM curves from the Refametinib and Mirdametinib datasets.

Source data are provided as a Source Data file.