

MATLAB Software to enable use of the statistical model is available at

<https://sourceforge.net/projects/ncspline/>

ADDITIONAL RESOURCES

None

SUPPLEMENTAL DATASETS

Dataset S1. Related to Figures 1 and 2 and STAR Methods section “Selected Reaction Monitoring Proteomics” (Attached as Excel spreadsheet). SRM parameters and intensities. List of detected SRM transitions and intensities as measured by peak area integration in Skyline software. Transitions and raw intensities are compiled from 3 data sets, across 7 injections, before normalization with housekeeping proteins.

Dataset S2. Related to Figures 1 and 2 and STAR Methods section “Ribosome profiling and mRNA-seq” (Attached as Excel spreadsheet). RPKM of genes monitored by ribosome profiling and mRNA seq and biological processes in Upreg and Downreg transcripts in response to bortezomib treatment. “MM1.S – DAVID analysis” sheet includes most enriched results from DAVID analysis (32) with default settings and human as species in MM1.S experiments. Top five most enriched terms shown measured from “Upreg” and “Downreg” clusters, as in Figure S2A.

Dataset S3. Related to Figures 2 and 4 and STAR Methods section “Estimation of absolute protein copy number at baseline” (Attached as Excel spreadsheet).

iBAQ values. MM1.S and EBV-immortalized B-cell tryptic peptide samples were analyzed using the iBAQ algorithm implemented in MaxQuant. For comparison to SRM and sequencing data, proteins were matched based both on UniProt identifier in MaxQuant protein group and gene name.

Dataset S4. Related to Figures 2 and 4 and STAR Methods section “Prediction of translational rate parameters and protein synthesis rates using ribosome profiling and RNA-seq measurements” (Attached as Excel spreadsheet).

Master dataset used for model calculations in both MM1.S and EBV-immortalized B-cell experiments and estimation and prediction of translational rate parameter $k_g^s(t)$ and protein synthesis rates in MM1.S experiments.

Absolute protein abundance (SRM intensity scaled by baseline iBAQ values; all values baseline corrected for “heavy” channel intensity at 0h) and mRNA and ribosome footprint density mapping to gene across the time course. All the values in this dataset are the fitted values by functional data analysis. “MM1.S - estimation & prediction” sheet includes the Pearson correlation coefficient and relative mean absolute error (RMAE) of the estimation and prediction of translational rate parameter and protein synthesis rate averaged over all the genes were compared to those between the two mass spectrometry replicates We define the relative mean absolute error between two time series $f_1(t)$ and $f_2(t)$ as $\frac{1}{T} \int_0^T |f_1(t) - f_2(t)| / |f_2(0)| dt$.

KEY RESOURCES TABLE

REAGENT or RESOURCE	SOURCE	IDENTIFIER
Antibodies		
Rabbit monoclonal anti-GAPDH	Cell Signaling Technology	Cat# 2118L
Rabbit monoclonal anti-vimentin	Cell Signaling Technology	Cat# 5741P
Rabbit polyclonal anti-Bid	Cell Signaling Technology	Cat# 2002P
Mouse monoclonal anti-puromycin	KeraFast	Cat# 3RH11
Rabbit monoclonal anti-4E-BP1	Cell Signaling Technology	Cat# 53H11
Rabbit monoclonal anti-phospho-4E-BP1 (T37/46)	Cell Signaling Technology	Cat# 236B4
Chemicals, Peptides, and Recombinant Proteins		
Bortezomib	LC Laboratories	B-1408
Cycloheximide	Sigma-Aldrich	Cat# C4859; CAS: 66-81-9
L-Lysine 4,4,5,5-D4 (Lys4)	Cambridge Isotope Laboratories	Cat# DLM-2640
L-Arginine ¹³ C ₆ (Arg6)	Cambridge Isotope Laboratories	Cat# CLM-2265-H
L-Lysine ¹³ C ₆ ¹⁵ N ₂ (Lys8)	Cambridge Isotope Laboratories	Cat# CNLM-291-H
L-Lysine ¹³ C ₆ ¹⁵ N ₄ (Arg10)	Cambridge Isotope Laboratories	Cat# CNLM-593-H
L-Lysine	Sigma-Aldrich	Cat# 8662; CAS: 657-27-2
L-Arginine	Sigma-Aldrich	Cat# A6969; CAS: 1119-34-2
TRIzol® reagent	Life Technologies	Cat# 15596026
HALT protease and phosphatase inhibitor single use cocktail	Thermo Fisher	Cat# 78443
Sequencing Grade Modified Trypsin	Promega	Cat# V5111
GAPDH recombinant protein	Abcam	Cat# ab82633
Vimentin recombinant protein	Peptrotech	Cat# 110-10
Bid recombinant protein	Sino Biological	Cat# 10468-HNCE-59
Critical Commercial Assays		
Rneasy mini kit	QIAGEN	Cat# 74104
Quantifluor RNA assay	Promega	Cat# E3310
Oligo(dT)25 Magnetic Beads kit	New England BioLabs	Cat# S1419S
Pierce™ BCA Protein Assay Kit	Thermo Fisher Scientific	Cat# 23225
SepPak C18 columns	Waters	Cat# WAT020515
CellTiter-Glo® Luminescent Cell Viability Assay	Promega	Cat# G7570
Caspase-Glo® 3/7 Assay Systems	Promega	Cat# G8090
Invitrogen MyOne streptavidin C1 dynabeads	Thermo Fisher	Cat# 65001
Deposited Data		

Raw genetic sequencing data	This paper	http://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?token=chohoqacvbyffwf&acc=GSE69047
Raw SRM data	This paper	https://panoramaweb.org/labkey/translation_model_2015.url
Human reference genome NCBI build 37, GRCh37	Genome Reference Consortium	http://www.ncbi.nlm.nih.gov/projects/genome/assembly/grc/human/
HMCL66_Transcript_Expression_FPKM	Keats Lab (Translational Genomics Research Institute, USA)	http://www.keatslab.org/data-repository
Experimental Models: Cell Lines		
MM.1S	ATCC	Cat# CRL-2974
EBV-transformed B-cell	Markus Muschen (University of California San Francisco, USA)	N/A
Sequence-Based Reagents		
miRNA Cloning Linker 1 DNA oligo: 5' AppCTGTAGGCACCATCAAT/3ddC 3'	IDT	N/A
rRNA subtraction DNA oligo oNTI309: (biotin)-TCCTCCCGGGGCTACGCCTGTCTGAGCGTCGCT	IDT	N/A
rRNA subtraction DNA oligo oNTI301r : (biotin)-GGCCTCGATCAGAAGGACTTGGGCCCCCCACGA	IDT	N/A
rRNA subtraction DNA oligo oNTI305r : (biotin)-GGCGAGACGGCCGGTGGTGCGCCCTCGGCGGA	IDT	N/A
rRNA subtraction DNA oligo oNTI307hr : (biotin)-GCGGGGACCGGCTATCCGAGGCCAACCGAGGCTC	IDT	N/A
rRNA subtraction DNA oligo oNTI298r : (biotin)-TGATCTGATAAATGCACGCATCCCCC	IDT	N/A
rRNA subtraction DNA oligo oNTI303hr : (biotin)-CGCGCCGTGGGAGGGTGGCCCGGCC	IDT	N/A
Rev Transcription DNA oligo oNTI225-Link1 (DNA): 5'/5Phos/GATCGTCGGACTGTAGAACTCTGAACCTGT CCGTGGTCGCCGTATCATT/iSp18/CACTCA/iSp18/CAAGCAGAAGACGGCATAACGAATTGATGGTGCCCTACAG 3'	IDT	N/A
Amplification DNA primer oNTI230 (DNA): 5'-AATGATACGGCGACCACCGA	IDT	N/A
Amplification DNA primer oNTI231 (DNA): 5'-CAAGCAGAAGACGGCATAACGA	IDT	N/A
Software and Algorithms		
MaxQuant v1.5.1.2	Cox and Mann, 2008	http://www.biochem.mpg.de/5111795/maxquant

Skyline v2.5	MacLean <i>et al.</i> , 2010	https://skyline.ms/project/home/software/Skyline/begin.view
Ribomap	Wang <i>et al.</i> , 2016	https://github.com/Kingsford-Group/ribomap
Star v.2.4.0j	Dobin <i>et al.</i> , 2012	https://github.com/alxdobin/STAR/releases/tag/STAR_2.4.0j
Cluster 3.0	de Hoon <i>et al.</i> , 2004	http://bonsai.hgc.jp/~mdehoon/software/cluster/software.htm
Java TreeView	Saldanha, A.J. 2004	http://jtreeview.sourceforge.net
DAVID Bioinformatics Resource	Huang <i>et al.</i> , 2009	http://david.ncifcrf.gov
MATLAB R2013a	Mathworks	http://www.mathworks.com/products/matlab/
MATLAB spline fit script	This paper	https://sourceforge.net/projects/ncspline/
Other		