Expanded View Figures

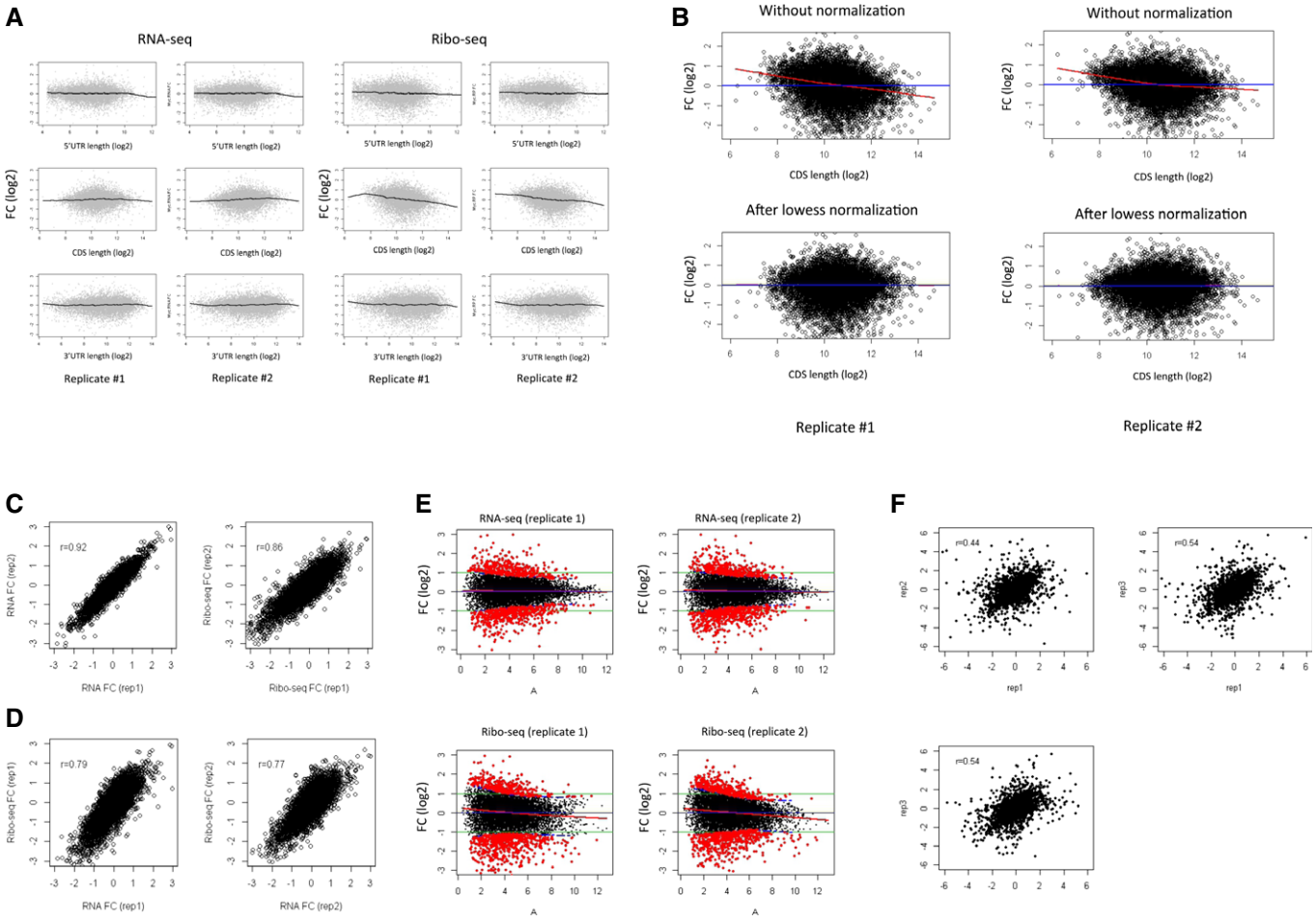

Figure EV1. Analysis of Myc-induced responses at the transcriptome, translatome and proteome levels.

A Relationship between Myc-induced gene responses measured by either RNA-seq (left) or Ribo-seq (right) and transcript 5′UTR (top), CDS (middle), and 3′UTR (bottom) lengths. A global relationship was detected between Ribo-seq fold change (FC, in log2 scale) and CDS length.

B The technical relationship between Ribo-seq fold change and transcripts CDS length was normalized using lowess normalization.

C Highly correlated Myc-induced responses were observed for biologically independent duplicate RNA-seq (left) and Ribo-seq (right) experiments.

D The Myc-induced response measured by Ribo-seq was, as expected, highly correlated with the one measured by RNA-seq.

E An adjusted fold-change threshold was set to define Myc-responsive genes: The standard deviation of fold change (at a log2 scale) was obtained as a function of expression level, and responsive genes (red dots) were defined as those whose fold change was at least 1.5-fold the SD above mean (dashed blue lines). A total number of 724 and 616 genes consistently responded to Myc activation in the RNA-seq (top) and Ribo-seq (bottom) duplicate experiments, respectively (with an overlap of 368 genes).

F Correlations between Myc-induced protein-level fold-change estimates obtained by three independent proteomic assays.

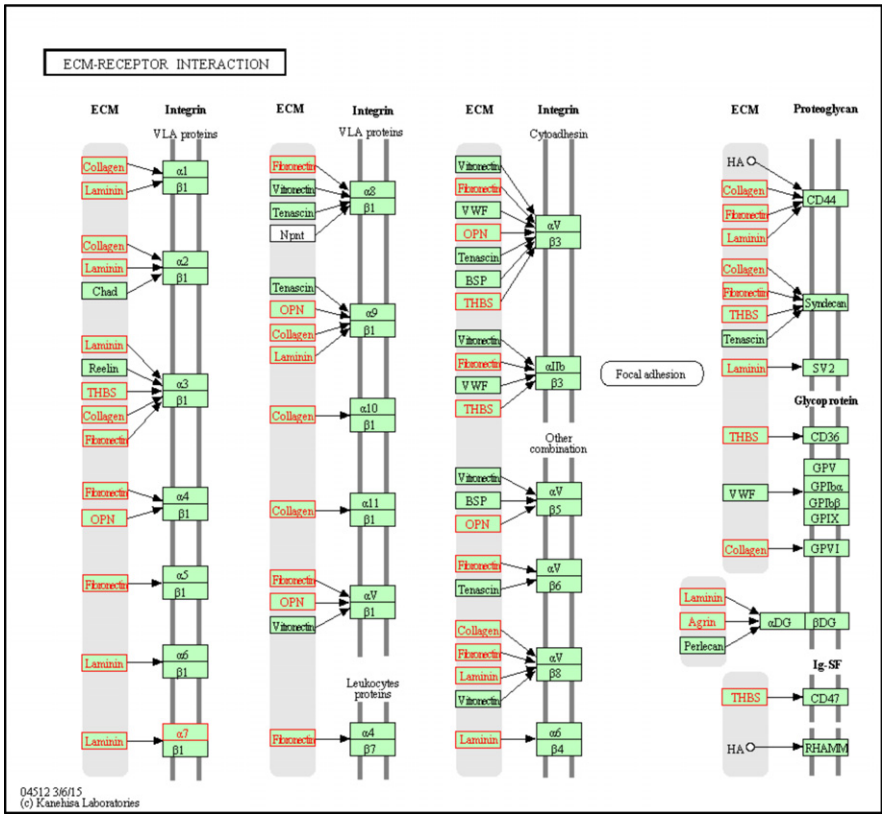

Figure EV2. Repression of an extensive network of ECM-receptor interactions in response to Myc activation.

Shown is the KEGG [62] pathway map for ECM-receptor interactions. Proteins whose encoding genes were assigned to the repressed cluster #2 are colored in red.

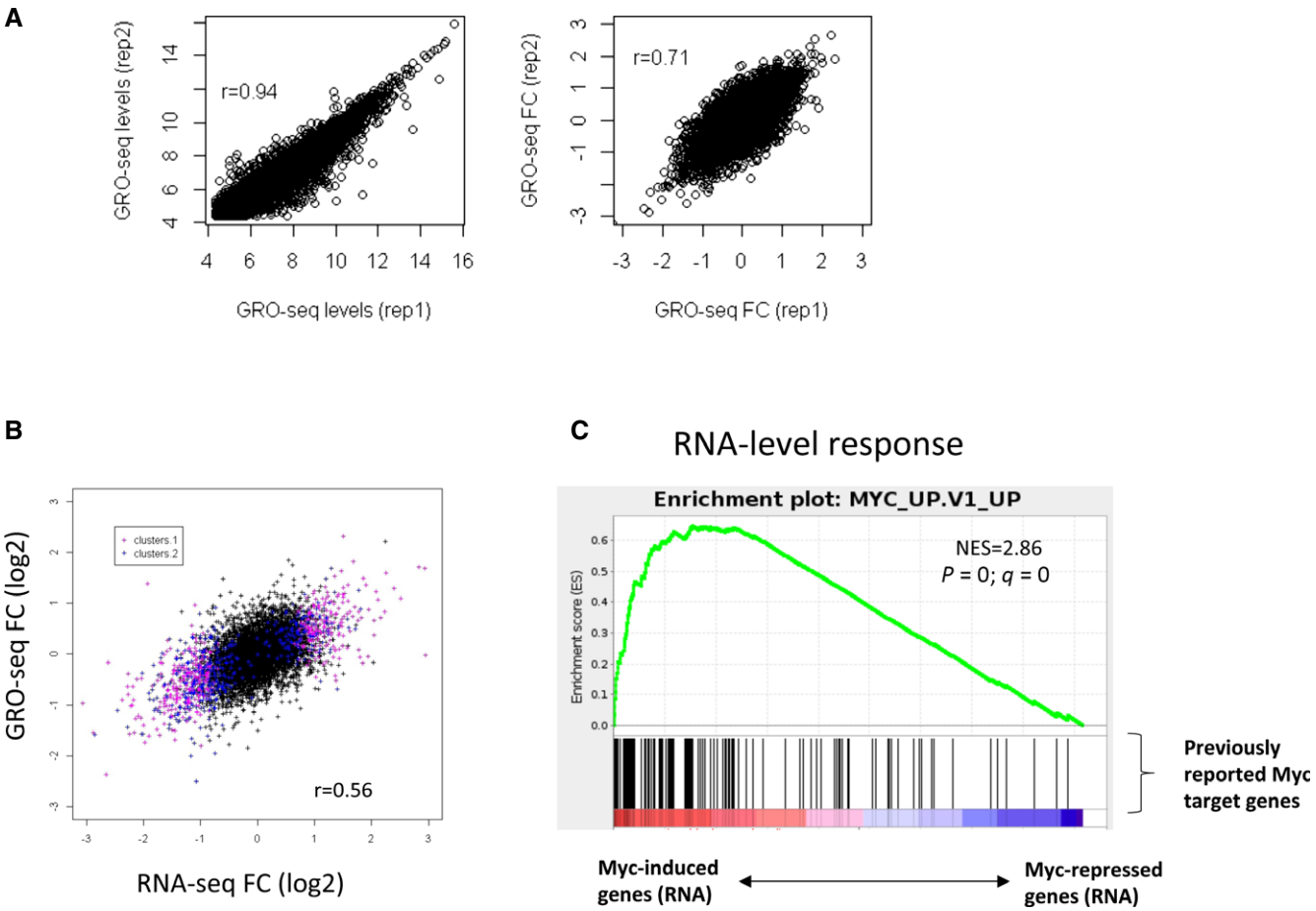

Figure EV3. Myc-induced transcriptional responses.

A Correlations between gene expression level (left) and gene response upon Myc activation (right) between two independent GRO-seq measurements.

B Correlation between changes in mRNA levels (RNA-seq) and transcription rates (GRO-seq) induced by Myc. Genes assigned to clusters #1 and #2 are colored in pink and blue, respectively.

C GSEA analysis applied to genes sorted by their change in mRNA level upon Myc activation showed that the induced genes in our dataset were significantly enriched for Myc target genes reported by previous studies.

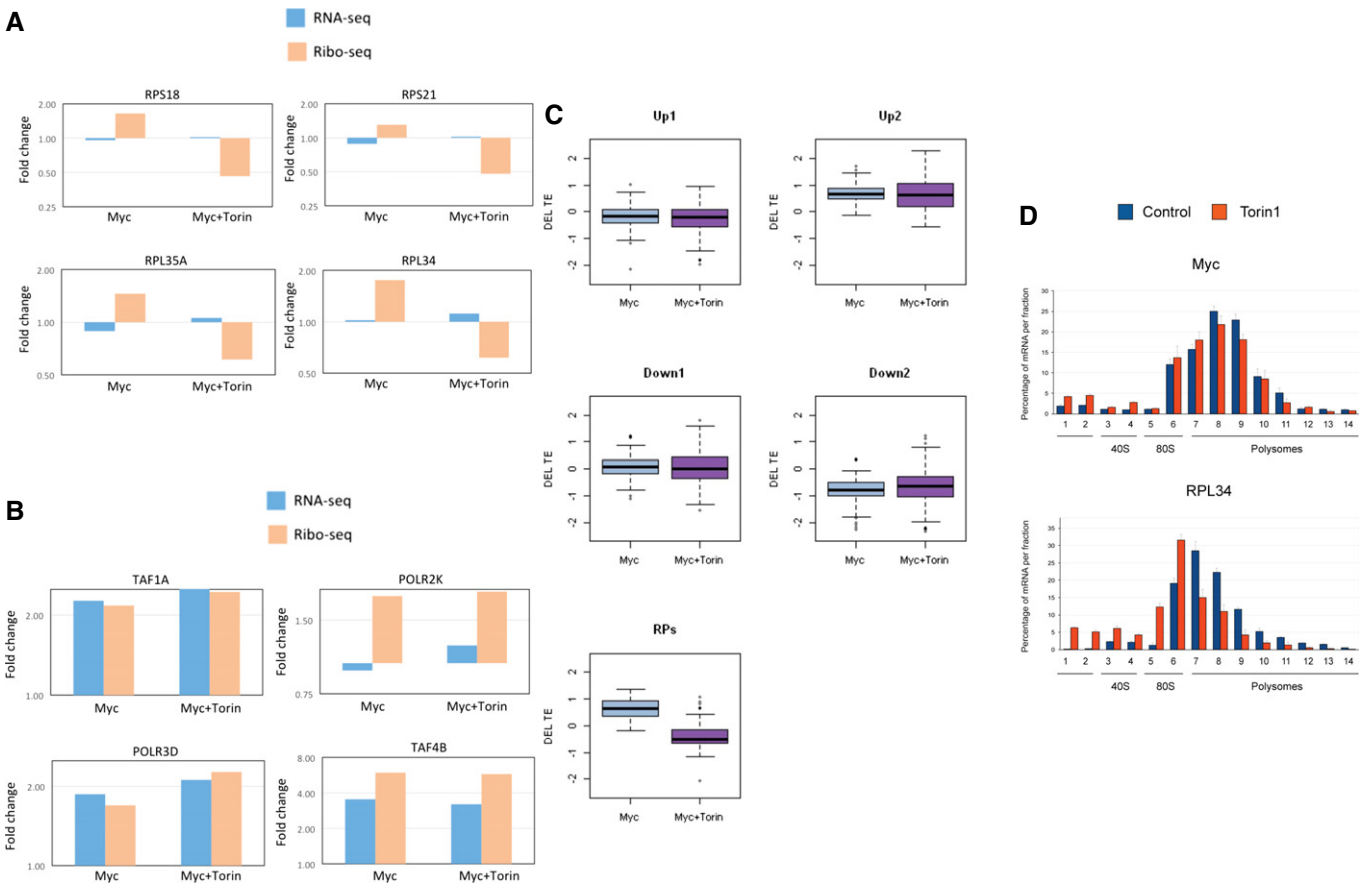

Figure EV4. Myc-induced regulation of protein translation.

A Example of ribosomal protein genes whose Myc-mediated translational induction was repressed and reversed by Torin-1 treatment.

B Example of genes whose Myc-mediated induction was not affected by Torin-1 treatment.

C A global statistical analysis showing that in contrast to the significant effect that Torin-1 had on the modulation of translation efficiency (TE) of ribosomal protein genes (RPs) upon Myc induction (bottom panel), Torin-1 had no significant effect on the TE of other components of the Myc-induced response network (divided by the clusters shown in Fig 1A). For the boxplots, the box indicates the 1st and 3rd quartiles; the horizontal band inside the box indicates the median. The whiskers extend to the most extreme data point which is no more than 1.5-times the interquartile range from the box.

D Using polysome fractionation assay followed by RT–PCR to quantify relative portion of Myc mRNA in each fraction, we confirmed that Torin-1 did not affect Myc TE. As a control, we used RPL34 whose TE is strongly repressed by Torin-1 (see Fig EV4A). The repression of RPL34 is reflected by the strong shift of the signal toward the subpolysomal fractions. Experiments were done in independent triplicates; error bars represent SD.

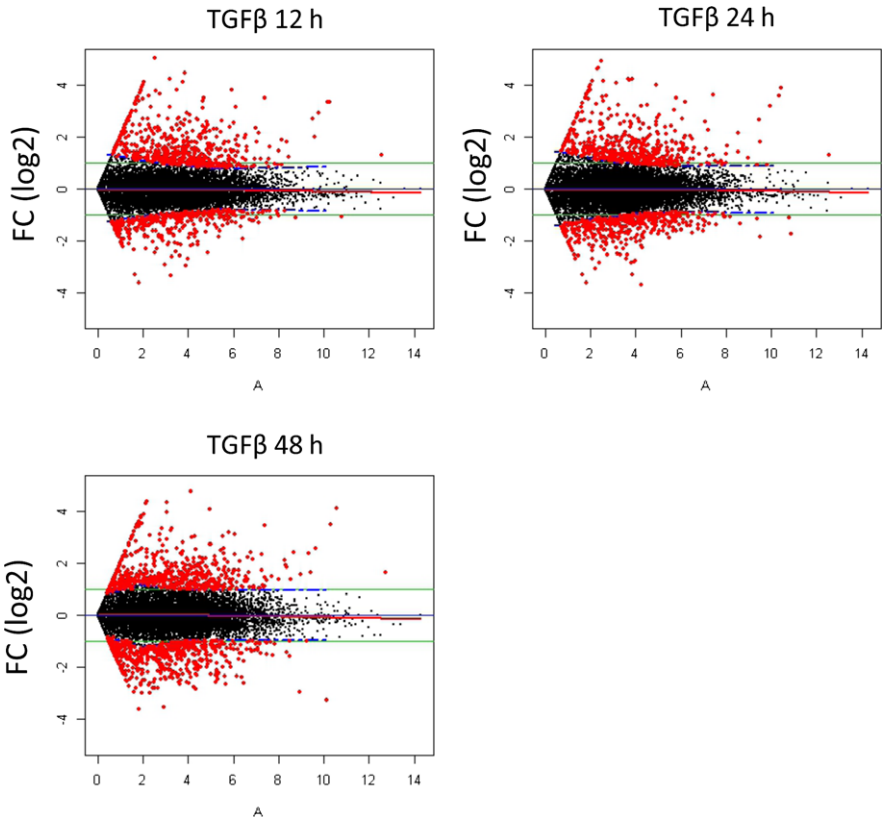

Figure EV5. TGF-beta-induced transcriptional responses.
Similar to Fig EV1E, an adjusted fold-change threshold was set to define TGFβ-responsive genes in MCF10A cells. The union of the responsive genes identified by this analysis was subjected to cluster analysis shown in Fig 5C.