

Cell Reports, Volume 26

## Supplemental Information

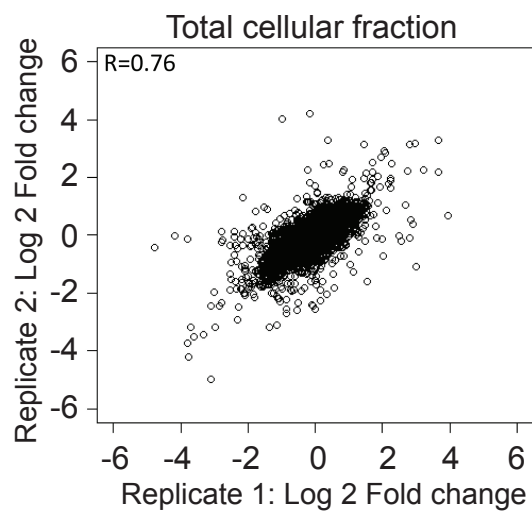
**Integrated Phosphoproteome and Transcriptome**

**Analysis Reveals *Chlamydia*-Induced**

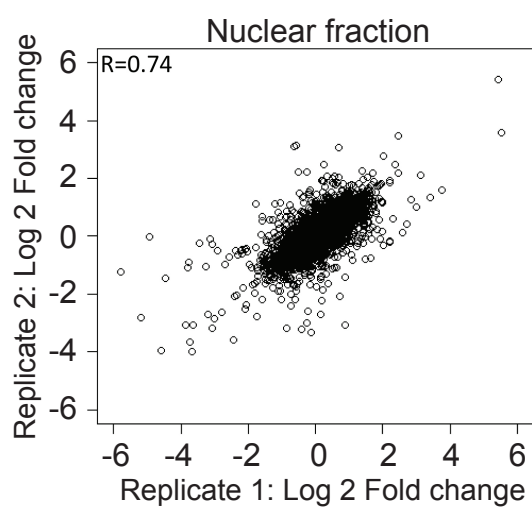
**Epithelial-to-Mesenchymal Transition in Host Cells**

**Piotr K. Zadora, Cindrilla Chumduri, Koshi Imami, Hilmar Berger, Yang Mi, Matthias Selbach, Thomas F. Meyer, and Rajendra Kumar Gurumurthy**

A

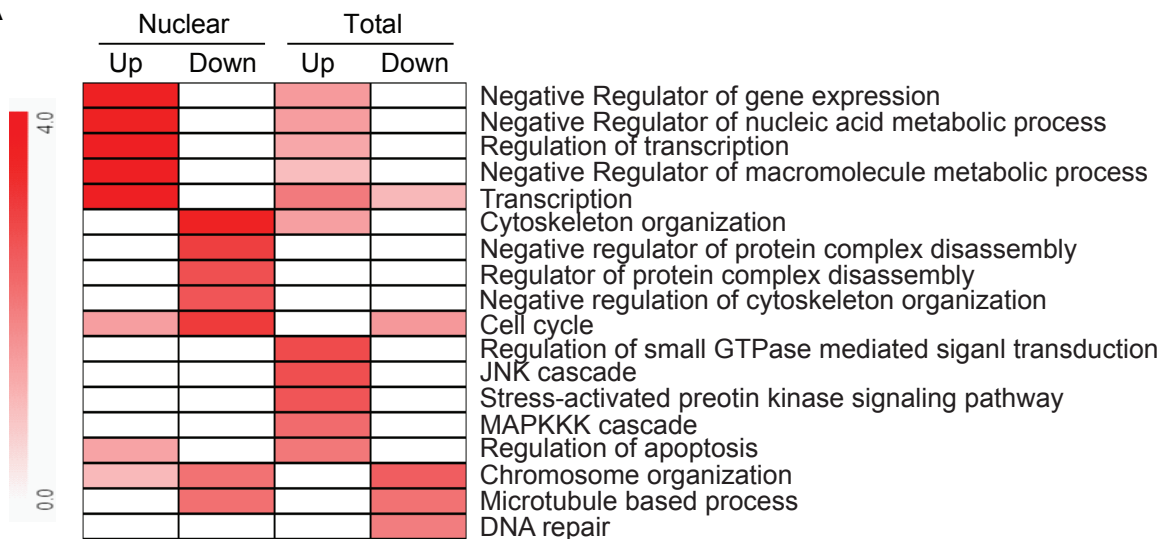


B





**Figure S1. SILAC labeling efficiency and global phosphoproteome correlation analysis. Related to Figure 1. (A-B)** Pearson's correlation coefficient analysis of log2-transformed phosphopeptide fold changes of two biological replicates from (A) total cellular fraction and (B) nuclear fraction shows high reproducibility.

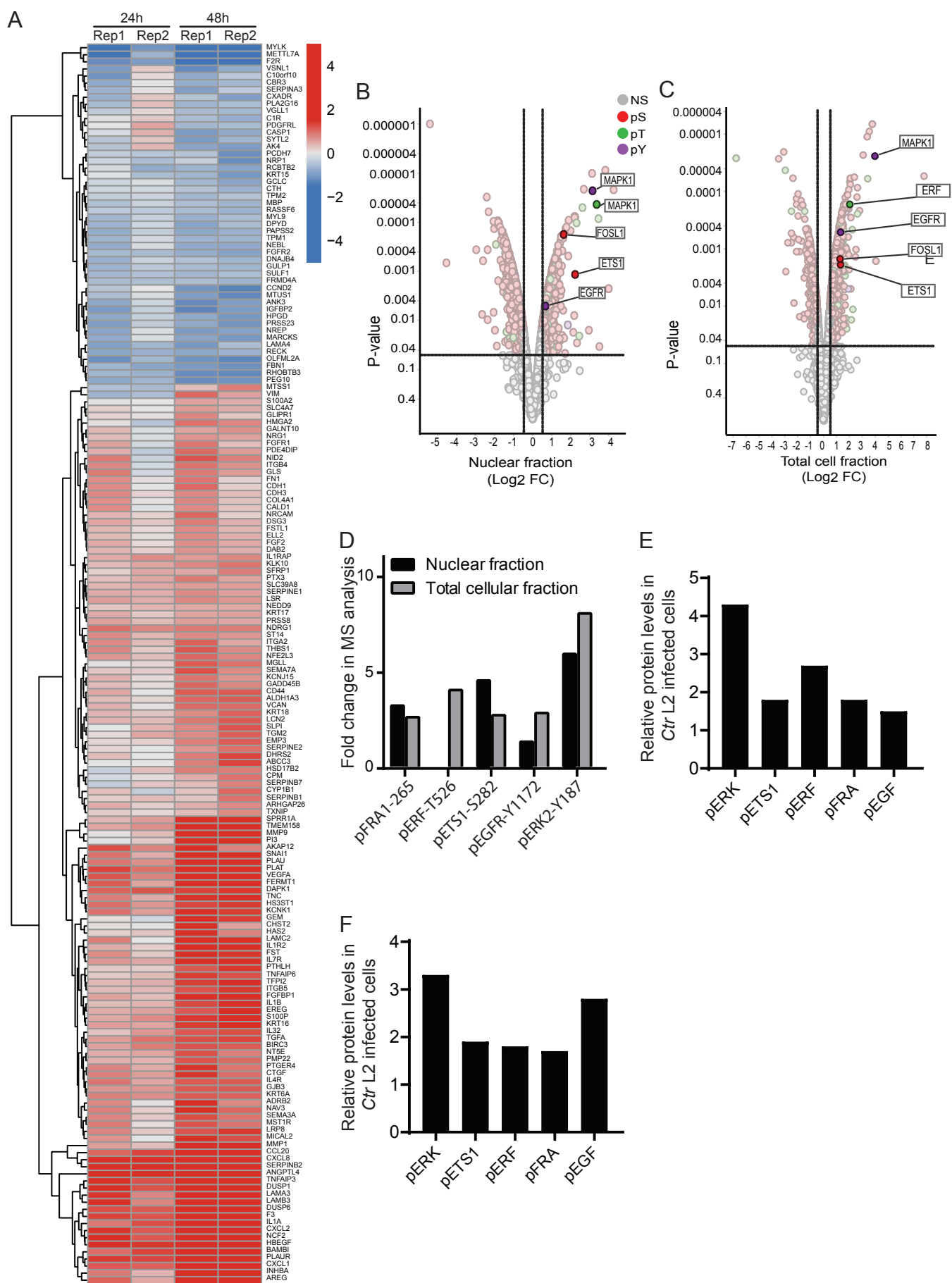
A



B

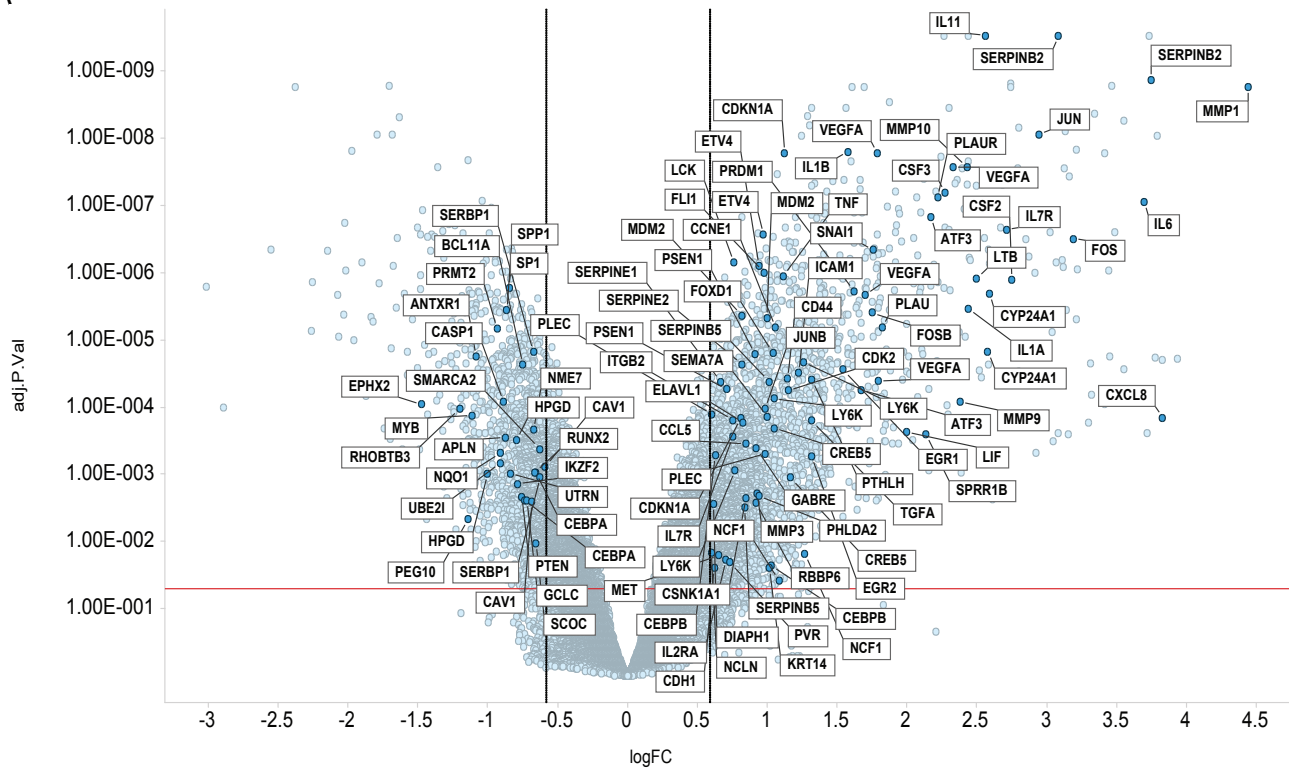
Top Diseases and Bio Functions			
Diseases and Disorders			
Name		p-value range	# Molecules
Cancer		5.59E-04 – 1.55E-45	929
Gastrointestinal Disease		3.45E-04 – 6.91E-37	703
Hepatic System Disease		2.66E-04 – 1.81E-23	460
Organismal Injury and Abnormalities		5.59E-04 – 4.65E-17	563
Reproductive System Disease		3.41E-04 – 2.47E-15	473
<div>1 2 3 4 5 6 7 8 9 &gt;</div>			
Molecular and Cellular Functions			
Name		p-value range	# Molecules
Gene Expression		2.37E-04 – 2.70E-26	293
Cellular Growth and Proliferation		5.08E-04 – 1.07E-23	413
Cell Cycle		5.70E-04 – 6.78E-21	249
Cellular Assembly and Organization		4.55E-04 – 2.55E-17	293
Cellular Function and Maintenance		3.91E-04 – 2.55E-17	225

**Figure S2. Functional analysis of *Ctr*-regulated global host phosphoproteome hits. Related to Figure 2.** (A) Shown are the top five biological processes from Gene Ontology (GO) term analysis for up- and downregulated phosphoproteins upon *Ctr* infection with  $p < 0.01$ . (B) Ingenuity pathway analyses for diseases and disorders as well as molecular and cellular functions enriched among phosphoproteins regulated upon *Ctr* infection. The five top-scoring hits are presented in each category, with significance scores (P-values) and the number of molecules in each class.

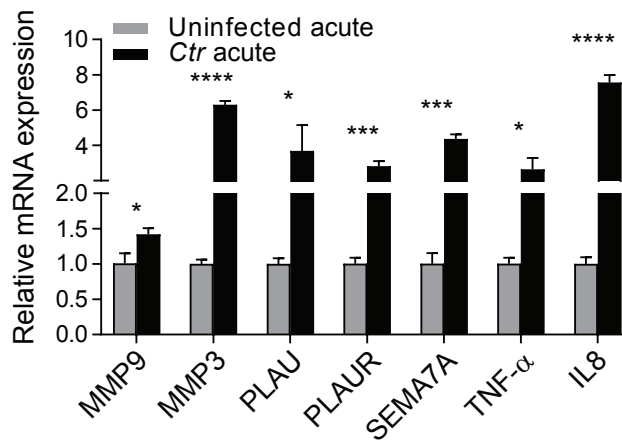


**Figure S3. Related to Figure 3.** (A) Heat map of 365 EMT-associated genes (Gröger et al., 2012) that are differentially regulated in *Ctr*-infected compared to uninfected cells at 24 and 48 h p.i. (B-C) Volcano plot representing global phosphoproteome data set from (B) nuclear fraction and (C) total cell extract according to log2 fold change (FC) (x-axis) and p-value (y-axis). The cut-off of  $\pm 0.5$  log2 FC (dashed vertical lines) and 0.05 p-value (dashed horizontal line) was applied. Phospho hits are highlighted depending on phosphorylated residues colored as labelled. The hits selected for validation are highlighted and labelled. (D) Shown is the fold-change in selected phosphosites after *Ctr* infection from the global phosphoproteome data. (E-F) Densitometric values of immunoblots from End1/E6E7 (E) and hCEcto (F) cells.

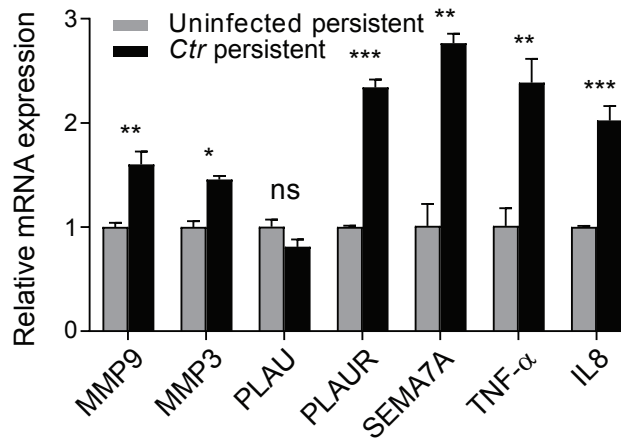
A



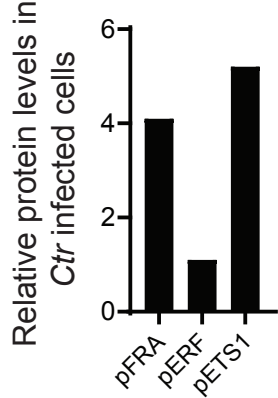
B



C



D



**Figure S4. Transcriptional regulation of epithelial to mesenchymal transition-associated genes upon *Ctr* infection. Related to Figure 4.** (A) Volcano plot highlighting *Ctr*-regulated genes that are known to be downstream of ERF, ETS1 and FRA1. (B-C) End1/E6E7 cells were (B) acutely or (C) persistently infected with *Ctr* for 32 h and 8 d, respectively. Shown is the relative mRNA expression of selected ERF, ETS1 and FRA1 target genes analyzed by qRT-PCR. Data shown as mean  $\pm$  SD from three technical replicates. \*\*\*\* $p < 0.0001$ , \*\*\* $p < 0.001$ , \*\* $p < 0.01$ , \* $p < 0.05$ ,  $p > 0.05$ , ns - not significant Student's t-test. (D) Densitometric values of immunoblots shown in Figure 4E.