

Fig. S1

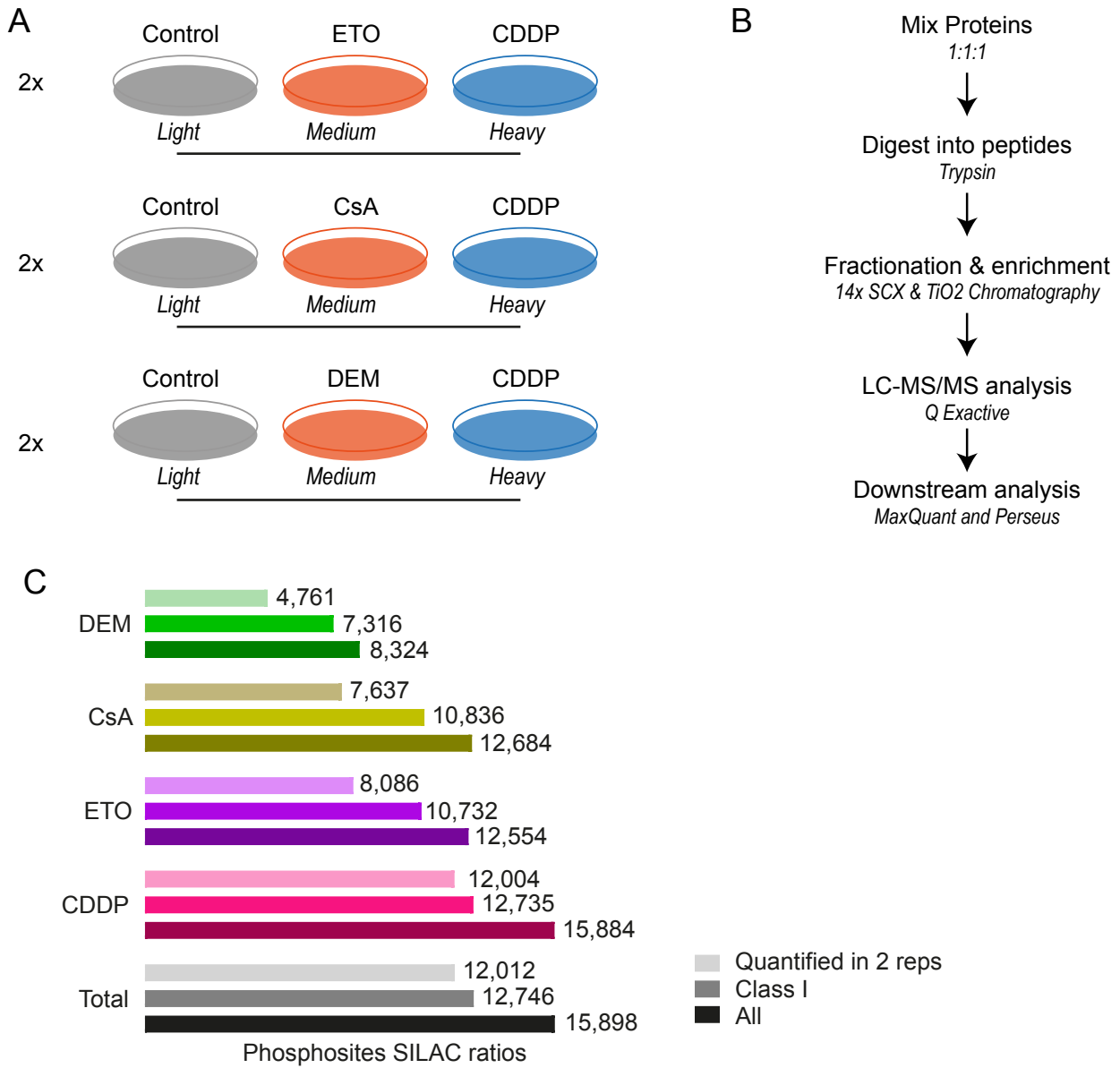


Fig. S2

**Total phosphosites
(n= 15,898)**

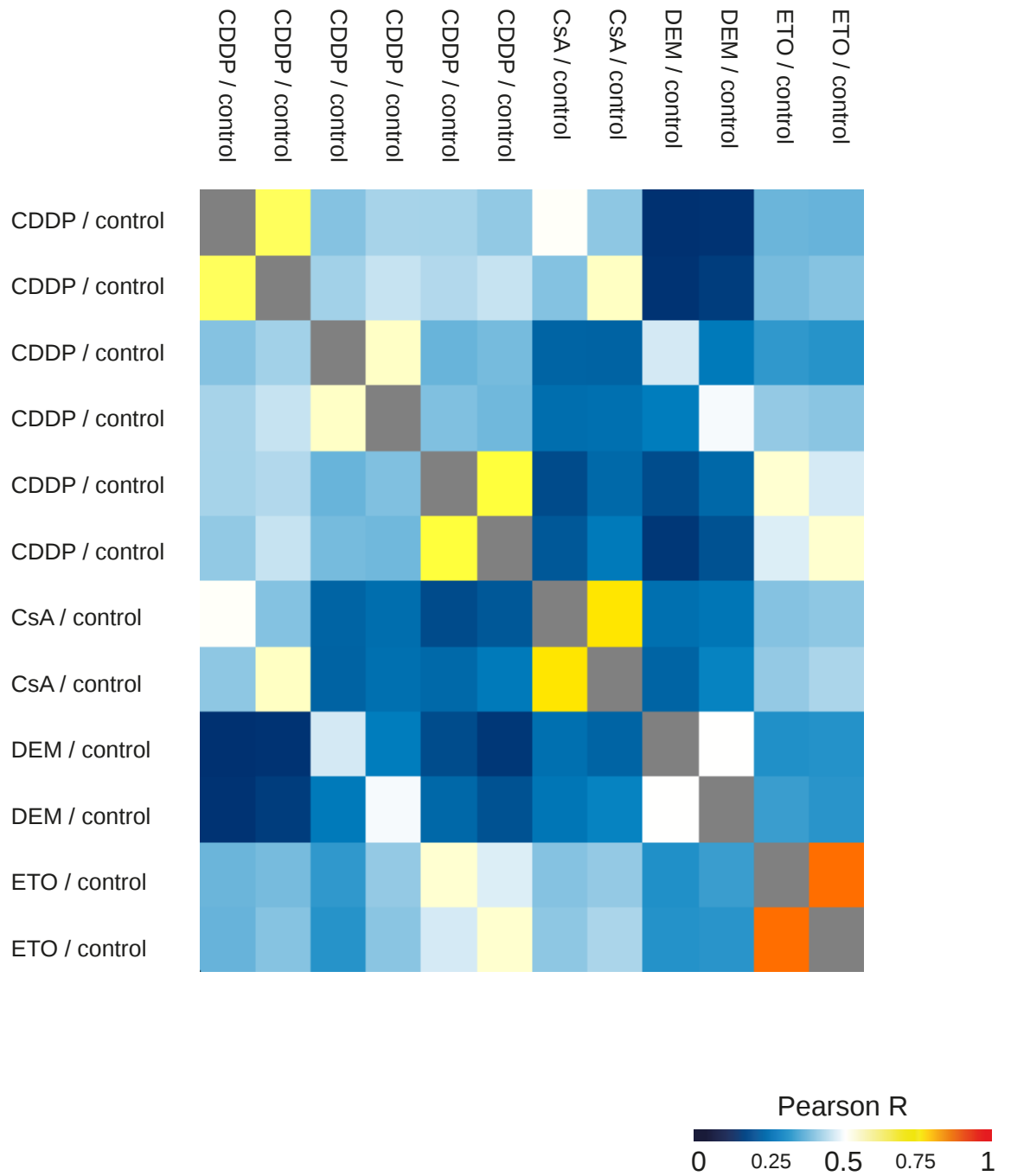


Fig. S3

Regulated phosphosites (quantified in all stressors)
1.5 fold change n=1,022

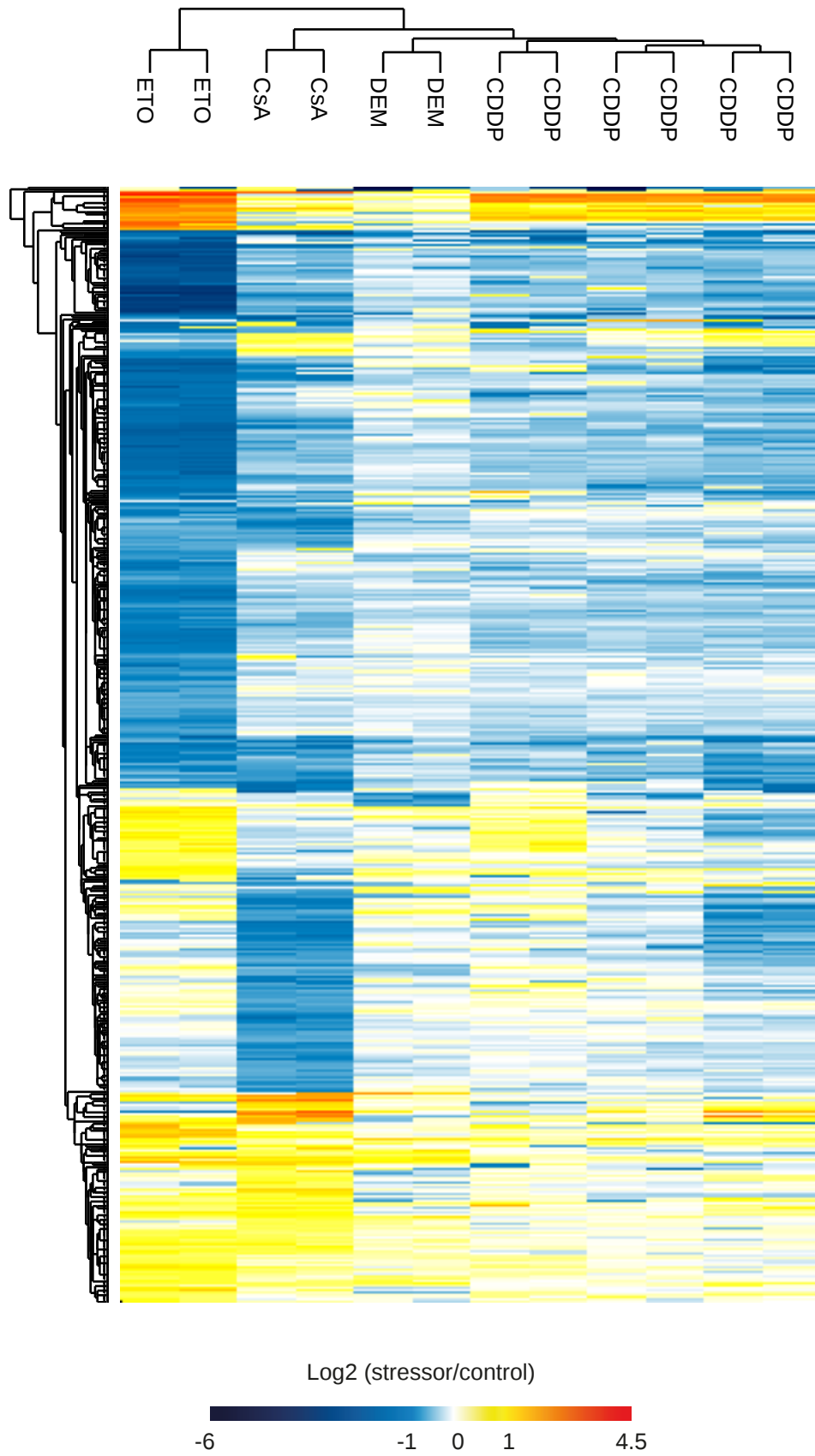


Fig. S4

Top 50 up-regulated phosphosites

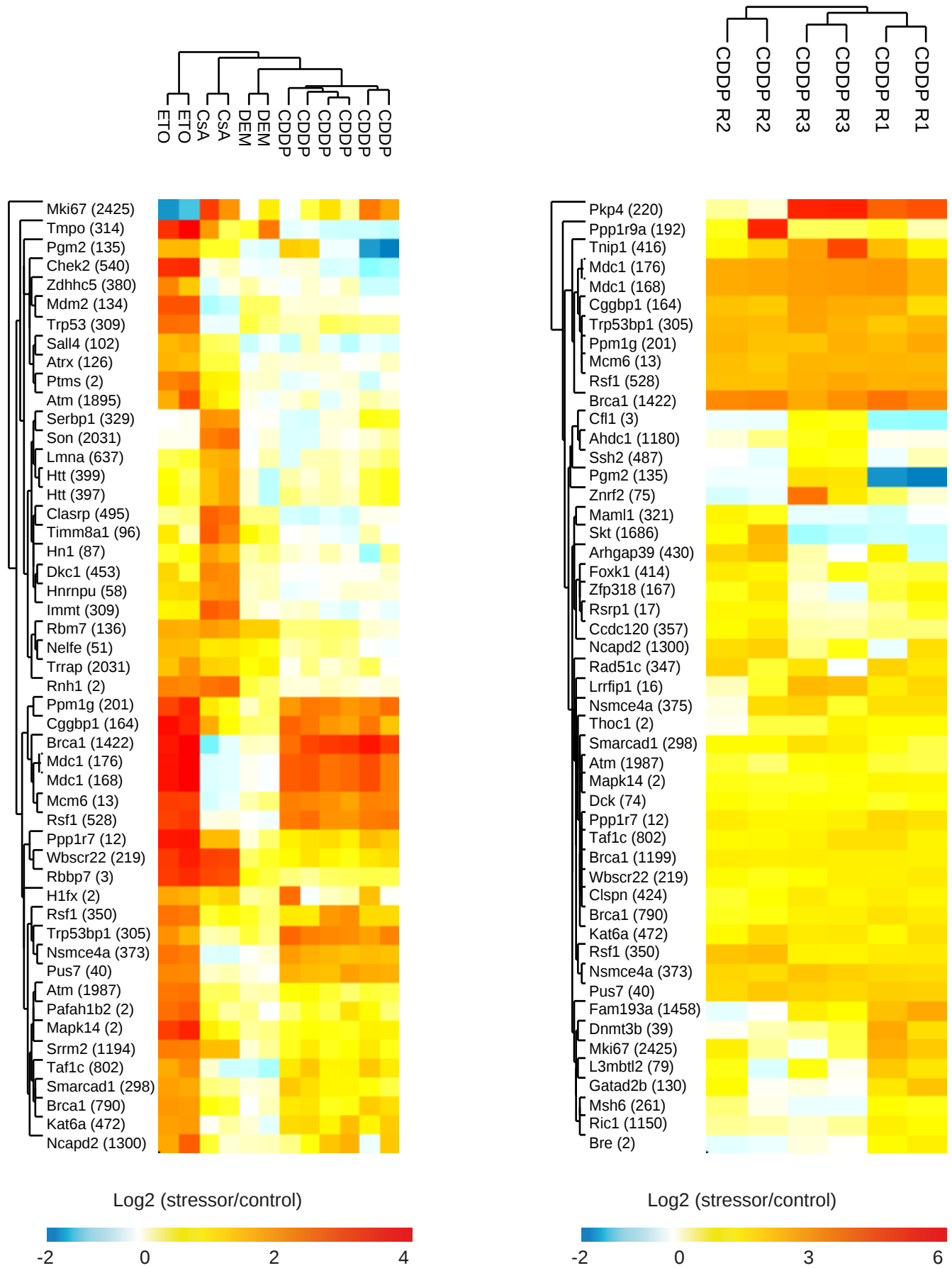


Fig. S5

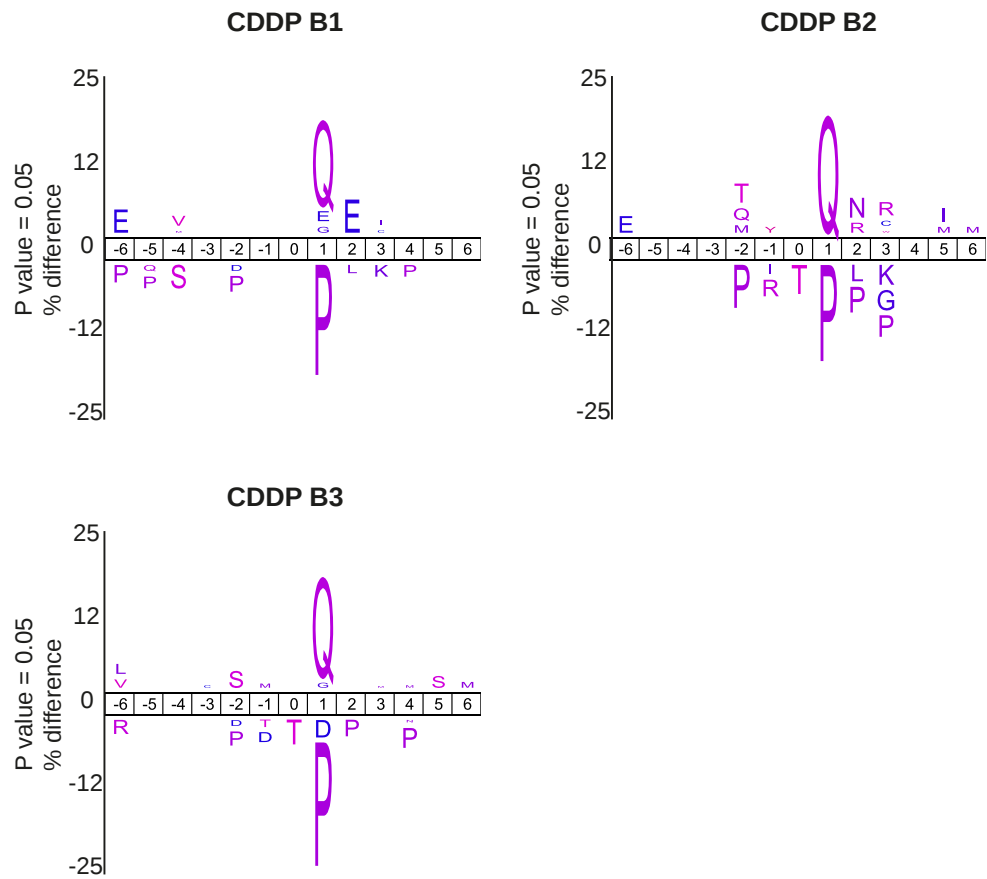
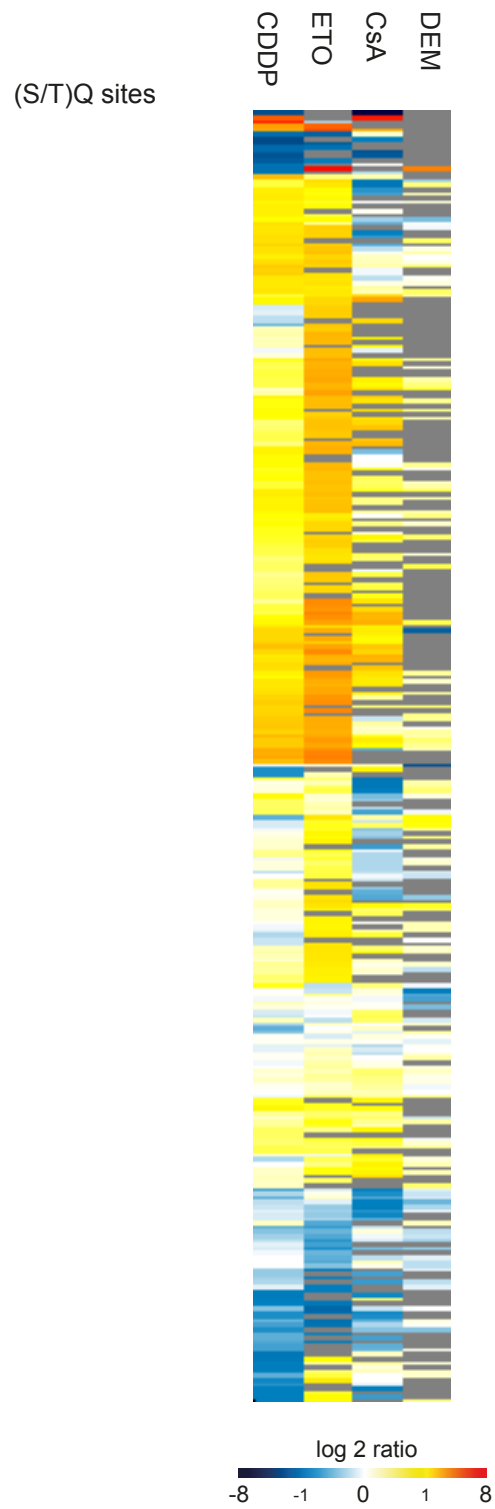


Fig. S6



Stressor	up-reg sites	up-reg p(S/T)Q sites	up-reg p(S/T)Q sites (%)
CDDP	708	136	19%
ETO	1473	180	12%
CsA	1039	76	7%
DEM	327	15	4.5%

Fig. S7

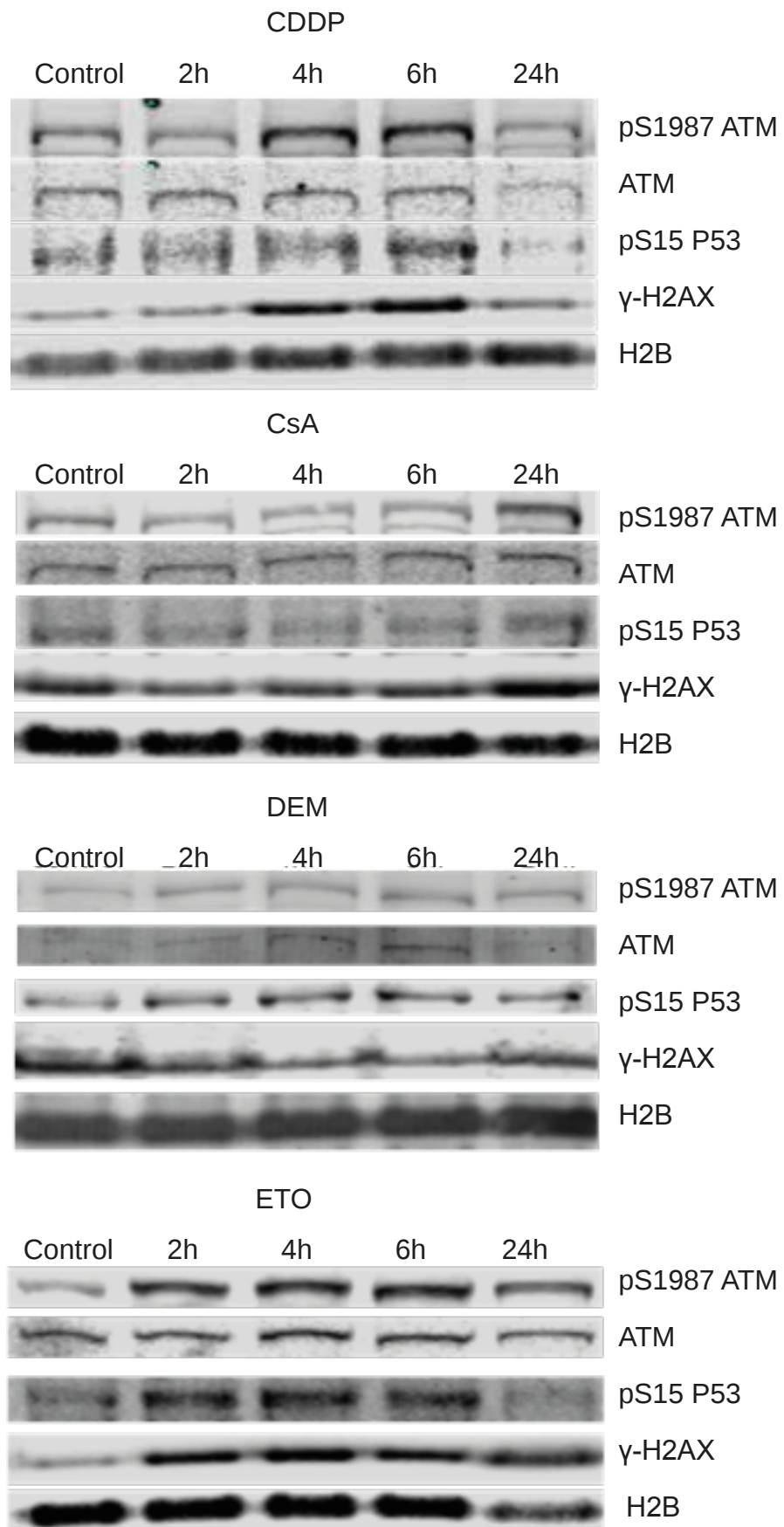
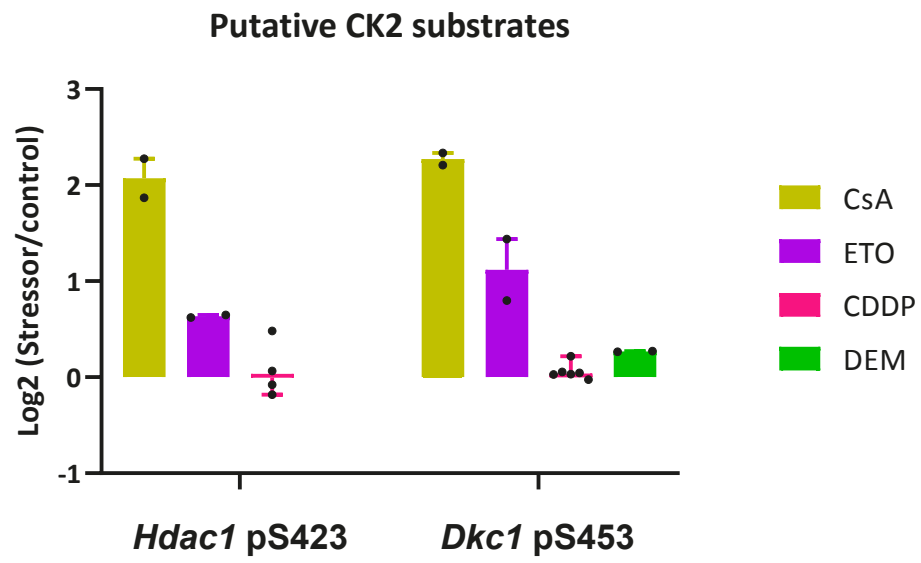


Fig. S8



Supplementary figure legends

Figure S1. Stressor-induced phosphoproteomes. (A) Experimental setup. In all three experiments (performed in biological duplicates) Light SILAC and Heavy SILAC cells were mock-treated and CDDP-treated, respectively. Medium SILAC cells were treated with either ETO, CsA or DEM. (B) Step-by-step phosphoproteomics workflow with specific methodology indicated in *italics*. (C) Coverage of the global phosphoproteome indicating the number of phosphosites quantified for each experiment.

Figure S2. Heatmap shows Pearson R values of log₂ phosphosite SILAC ratios (stressors over control) of all quantified 15,858 phosphosites.

Figure S3. Heatmap showing the log₂ phosphosite SILAC ratios over untreated controls that are quantified in all four stressors (including all replicates) that passed a 1.5-fold change cut-off.

Figure S4. Heatmap showing log₂ phosphosite SILAC ratios of top 50 up-regulated phosphosites that are quantified in all four stressors including all replicates (left panel) and that are quantified in all six CDDP replicates (right panel).

Figure S5. Motif analyses of differentially regulated phosphosites for each biological replicate of CDDP. Sequence motifs analyses and visualization was performed using the IceLogo tool. For each replicate, the sequences of all quantified phosphosites were used as a statistical background. A p-value cut-off of 0.05 was applied. The phosphorylated amino acid is located at position 0. Motifs enriched for the indicated replicate of CDDP among the up-regulated phosphosites.

Figure S6. Heatmap showing the log₂ phosphosite SILAC ratios over untreated controls of phosphosites with (S/T)Q motif that responded to any of the stressors. **Table inlet** displays an overview of the fraction of up-regulated (S/T)Q phosphosites.

Figure S7. Western blot probing dynamic changes of pS1987 ATM, ATM, pS15 p53, γ-H2AX and H2B after treatment with CDDP (top panel), CsA (second panel from top), DEM (second panel from bottom) and ETO (bottom panel)

Figure S8. Bar chart of site-specific quantifications of two example putative CK2 substrates.