Supplementary Information

The reciprocal interplay between TNFα and the circadian clock impacts on cell proliferation and migration in Hodgkin lymphoma cells

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Supplementary Materials

**Table S1** – Pairwise interactions Clock-Immune Genes
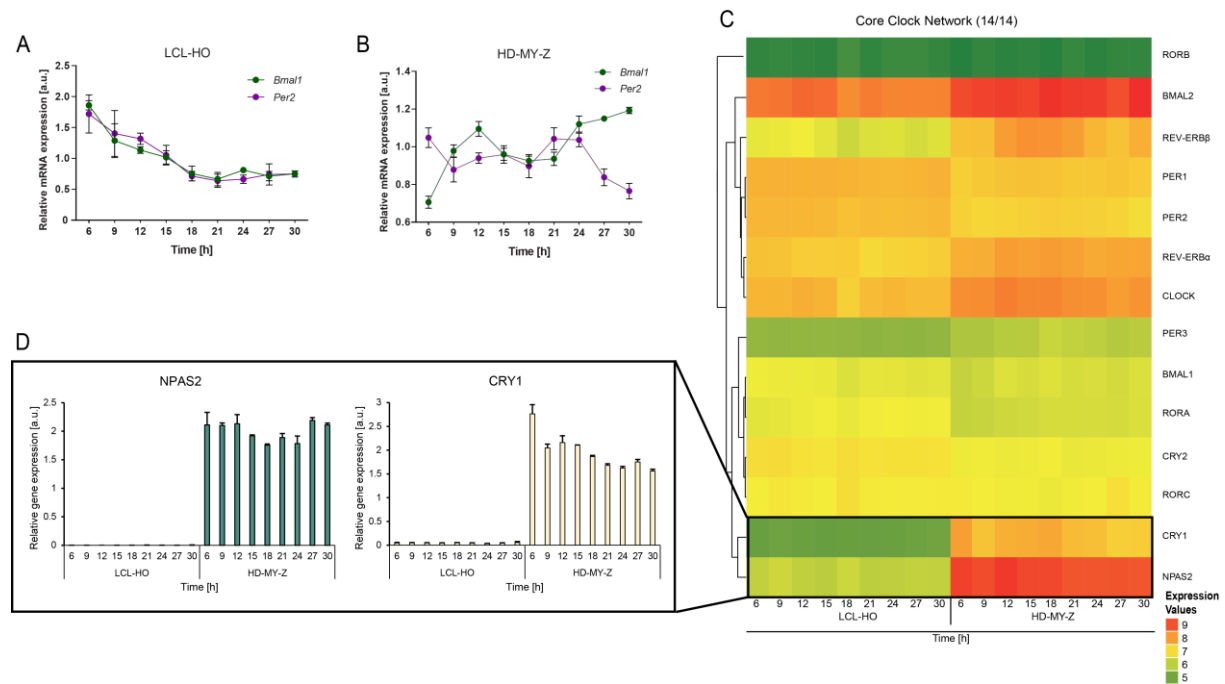
Figure S1 – LCL-HO and HD-MY-Z have differential circadian phenotypes Depicted are gene expression values of *Bmal1* and *Per2* for LCL-HO (**A**) and HD-MY-Z (**B**) over 24 h (n=3, mean ± SEM), expression of core-clock genes in the HTA 2.0 for both cell lines (**C**) and (**D**) 24h time-course qPCR for *NPAS2* and *CRY1* genes (n=3, mean ± SEM).
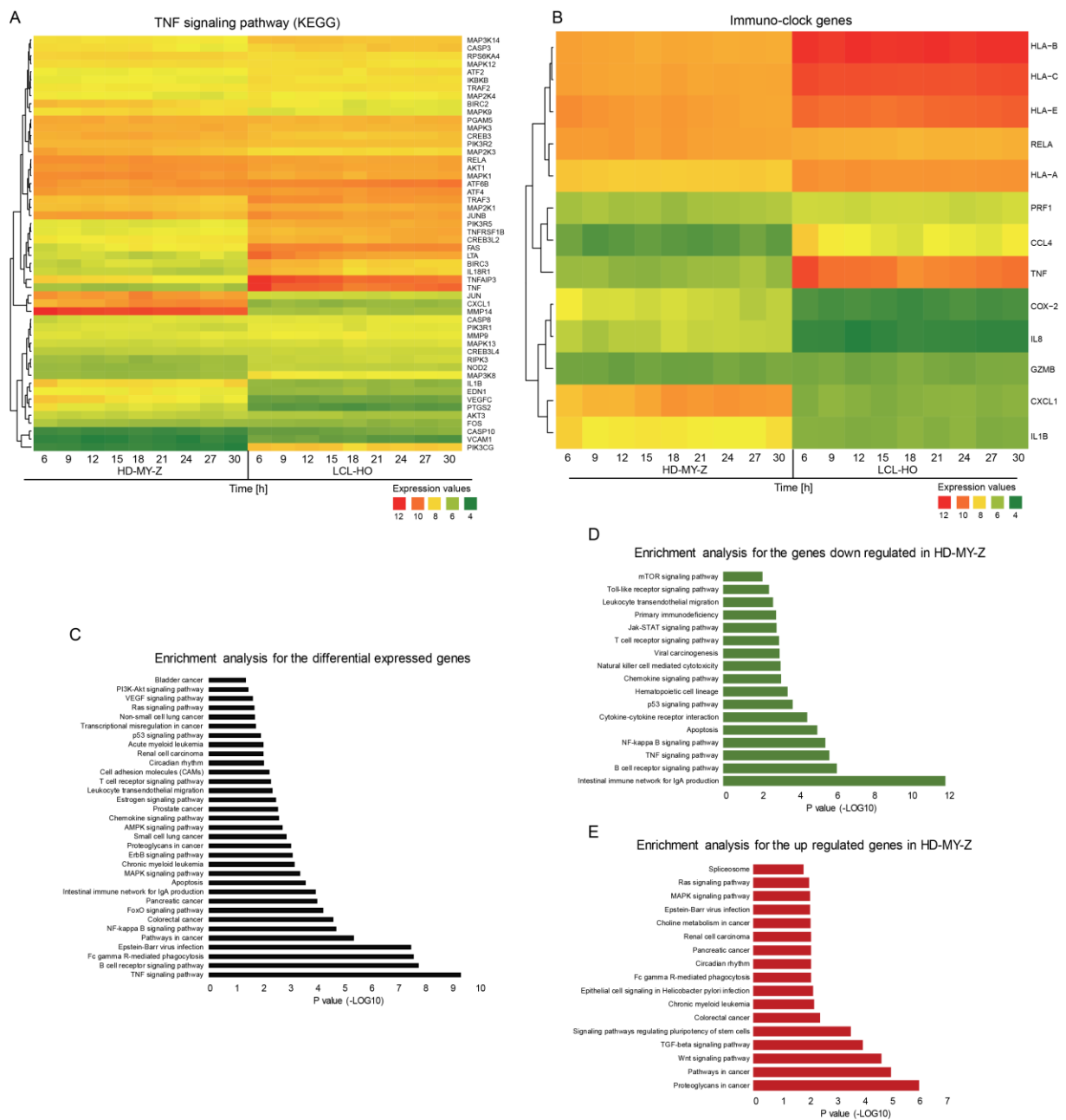
Figure S2 – The TNF signalling pathway is deferentially expressed in HL. (**A** and **B**) heatmaps showing the relative expression of the differentially expressed genes in the TNF signalling pathway and immune-clock genes (from Figure 1). (**C**, **D** and **E**) enrichment analysis for KEEG pathways of the differentially expressed genes in LCL-HO and the HL cell line HD-MY-Z.
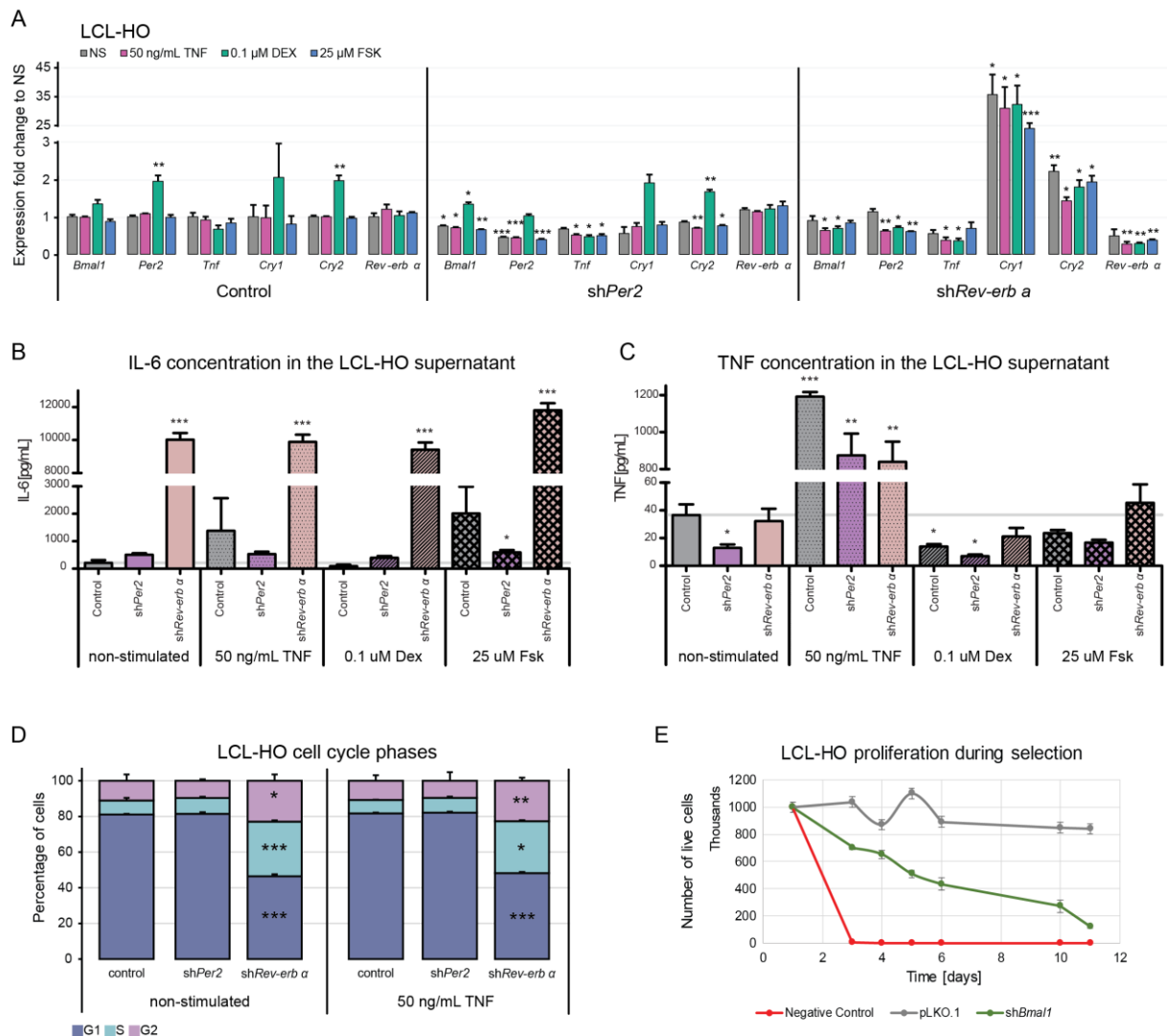
Figure S3 – TNF affects cell cycle and expression of core-clock genes in LCL-HO cell line. (**A**) Gene expression qPCR data (n=3, mean ± SEM), (**B**) IL-6 and (**C**) TNF concentration in the supernatant (n=3, mean ± SEM) (**D**) measurements of cell cycle phases (n=3, mean ± SEM) after shRNA KD of *Per2* and *Rev-erb α* and stimulation for 3 days with 50 ng/mL of recombinant human TNF, 0.1 µM of dexamethasone and 25 µM of forskolin. (**E**) proliferation measurements after sh*Bmal*1 (n=3, mean ± SEM). Statistics for the compassion to non-stimulated using unpaired t test: * p < 0.05, ** p < 0.01, *** p < 0.001. LCL-HO WT, LCL-HO transduced with pLKO.1 empty vector and transduced with sh*Bmal1* KD vector proliferation during the selection with puromycin.
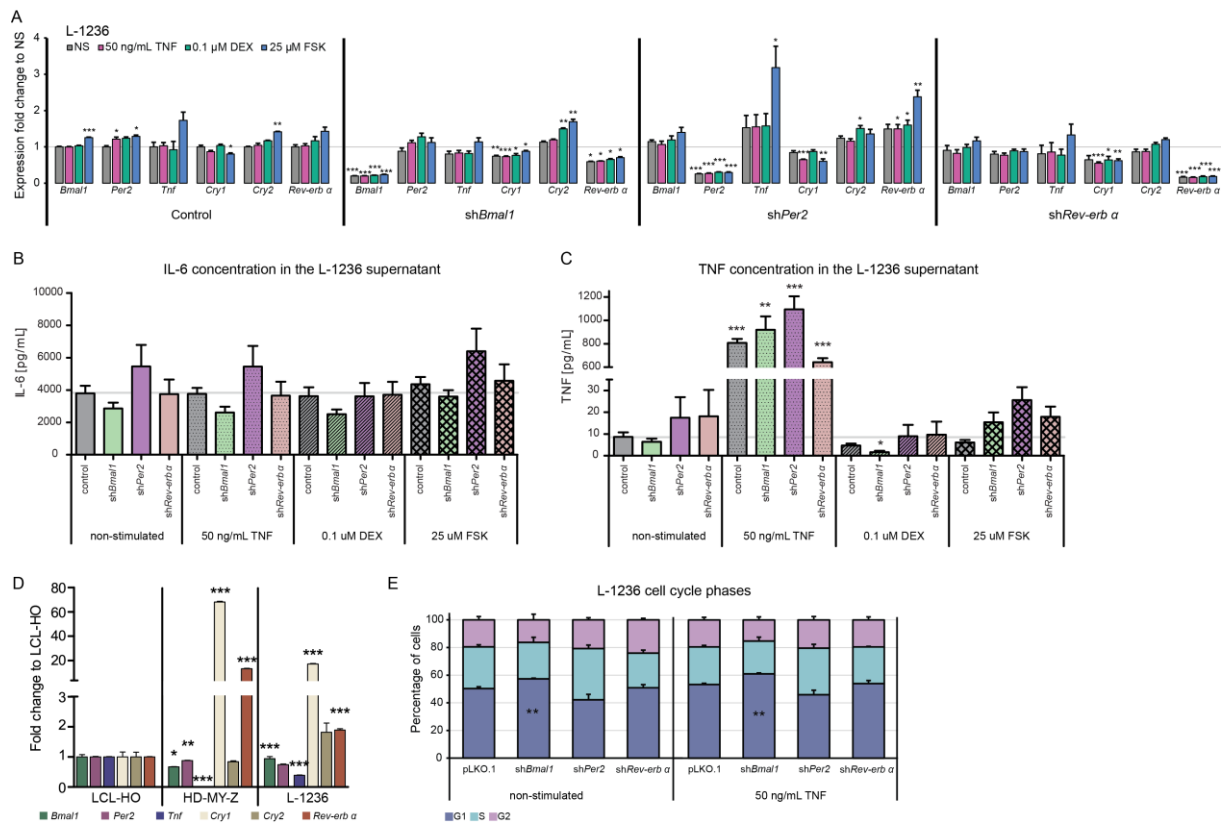
Figure S4 – TNF affects cell cycle and expression of core-clock genes in the HL cell line L-1236. (**A**) Gene expression qPCR data (n=3, mean ± SEM), (**B**) expression levels of core-clock genes and Tnf in HL cell lines, qPCR data (n=3, mean ± SEM) (**C**) IL-6 and (**D**) TNF concentration in the supernatant (n=3, mean ± SEM) (**E**) measurements of cell cycle phases (n=3, mean ± SEM) after shRNA KD of *Bmal1*, *Per2* and *Rev-erb α* and posterior stimulation for 3 days with 50 ng/mL of recombinant human TNF, 0.1 μM of dexamethasone and 25 μM of forskolin. (Statistics for the comparison to non-stimulated using unpaired t-test: * p < 0.05, ** p < 0.01, *** p < 0.001.
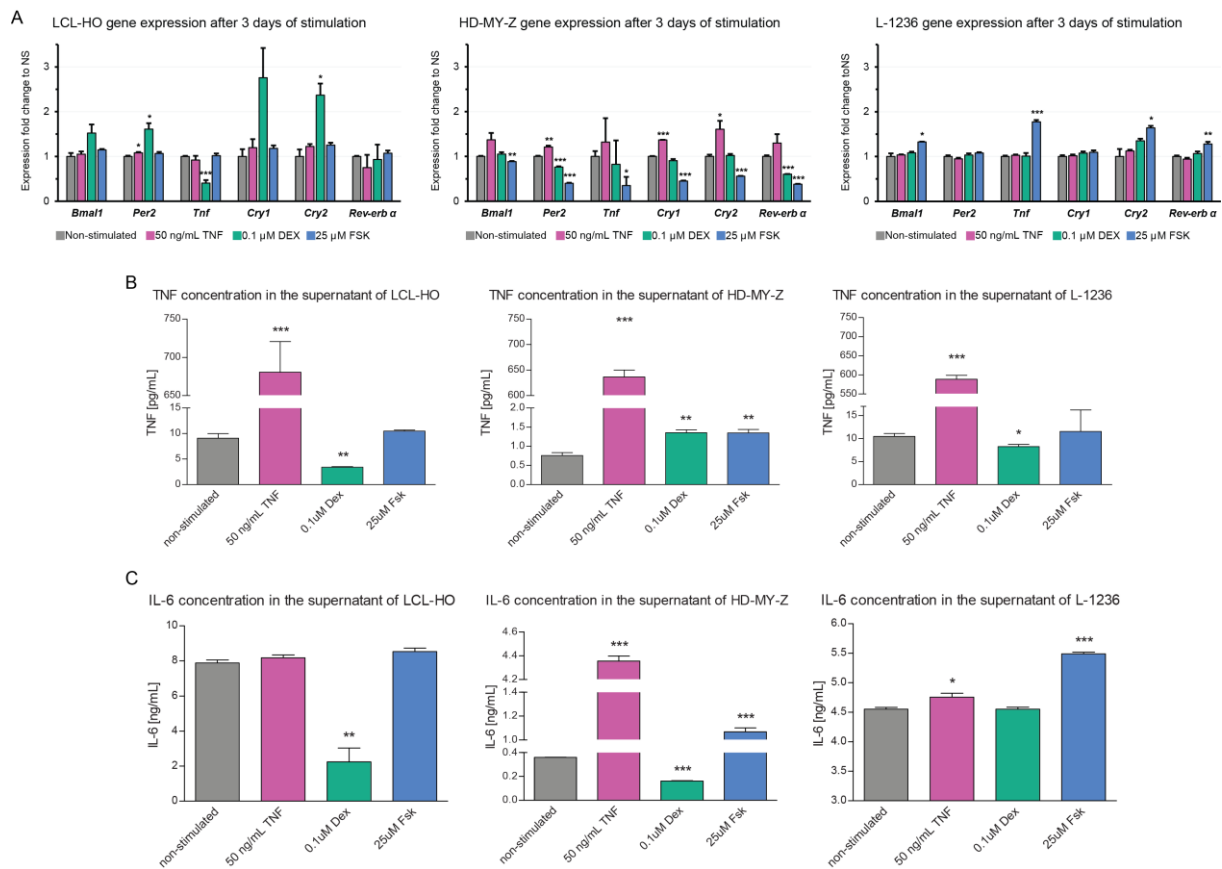
Figure S5 – Overall comparison for the effects of TNF on the core-clock in HL cell lines. (**A**) Gene expression qPCR data (n=3, mean ± SEM), (**B**) TNF and (**C**) IL-6 concentration in the supernatant (n=3, mean ± SEM) for the 3 cell lines (WT) after 3 days stimulation with 50 ng/mL of recombinant human TNF, 0.1 µM of dexamethasone and 25 µM of forskolin. Statistics for the compassion to non-stimulated using unpaired t test: * p < 0.05, ** p < 0.01, *** p < 0.001.
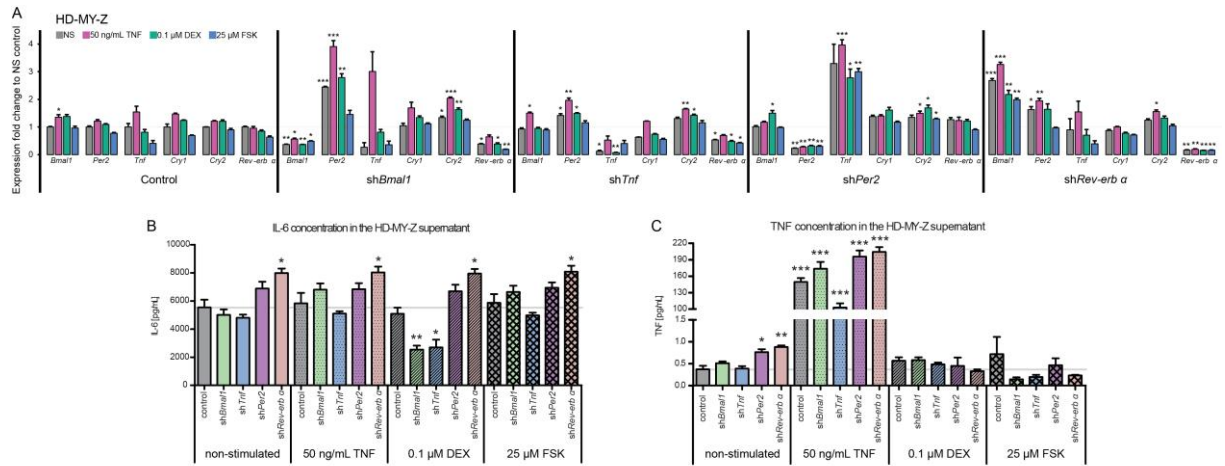
Figure S6 – The core-clock affects expression of TNF and IL-6 in the HL cell line HD-MY-Z. (**A**) Gene expression qPCR data (n=3, mean ± SEM), (**B**) IL-6 and (**C**) TNF concentration in the HD-MY-Z supernatant (n=3, mean ± SEM) for the 3 cell lines (WT) after 3 days stimulation with 50 ng/mL of recombinant human TNF, 0.1 µM of dexamethasone and 25 µM of forskolin. Statistics for the compassion to non-stimulated using unpaired t test: * p < 0.05, ** p < 0.01, *** p < 0.001.