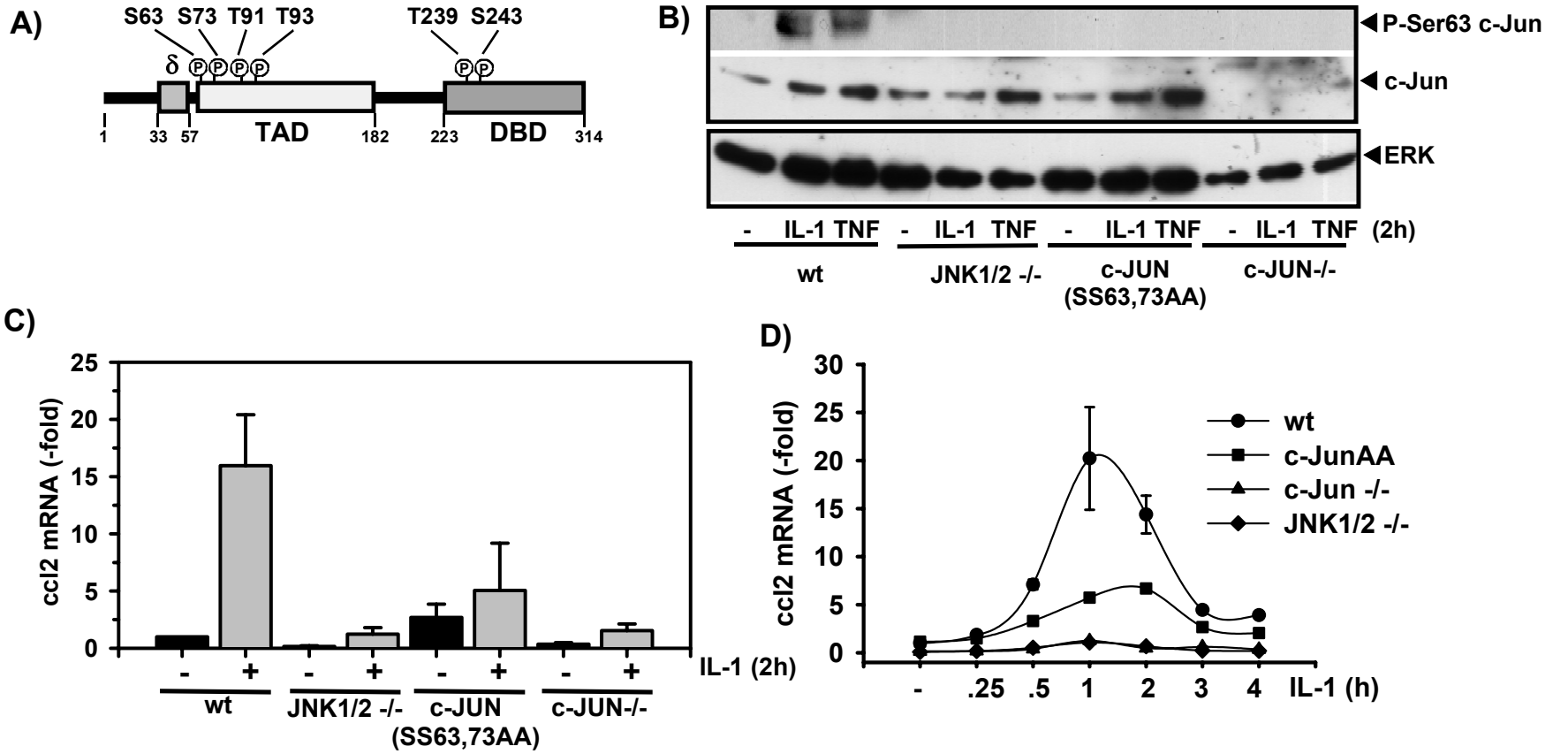

JNK- and c-Jun-dependent ccl2 mRNA expression in genetically altered mouse fibroblasts

A) Schematic representation of c-Jun structure indicating the N-terminal transactivation domain (TAD), the C-terminal DNA-binding domain (DBD) and the JNK-binding (d) domain. P- indicates phosphorylation sites. B) Wild type (wt) embryonic fibroblast cell lines, cells lacking JNK1 and JNK2 genes (JNK1/2 -/-), or c-Jun (c-Jun -/-), or cells that were isolated from mice carrying a c-Jun allel mutated in two of the four JNK phosphoacceptor sites (c-Jun SS63/73AA) were kept in low serum (0.1%) for 48h. Thereafter, cells were treated for 2h with 10ng/ml IL-1a or were left untreated. Phosphorylation and expression of c-Jun were analysed by western blotting of whole cell extracts. ERK antibodies were used to control for protein loading. C) The cells described in B) were kept in low serum (0.1%) for 48h. Thereafter, they were treated for two hours with IL-1a (10ng/ml). ccl2 mRNA expression was determined by Taqman real-time PCR using total RNA. Shown is the mean ccl2 expression +/- S.E.M. from three independent experiments relative to the unstimulated wild type control cells. D) ccl2 mRNA expression was determined in cells kept in low serum as in D) that were treated for the indicated times with IL-1a (10ng/ml). Expression of ccl2 was analysed as in C). Shown is the mean ccl2 expression +/- S.E.M. from two experiments relative to the unstimulated wild type control cells. For details see(Wolter et al., MolCellBiol., 2008)