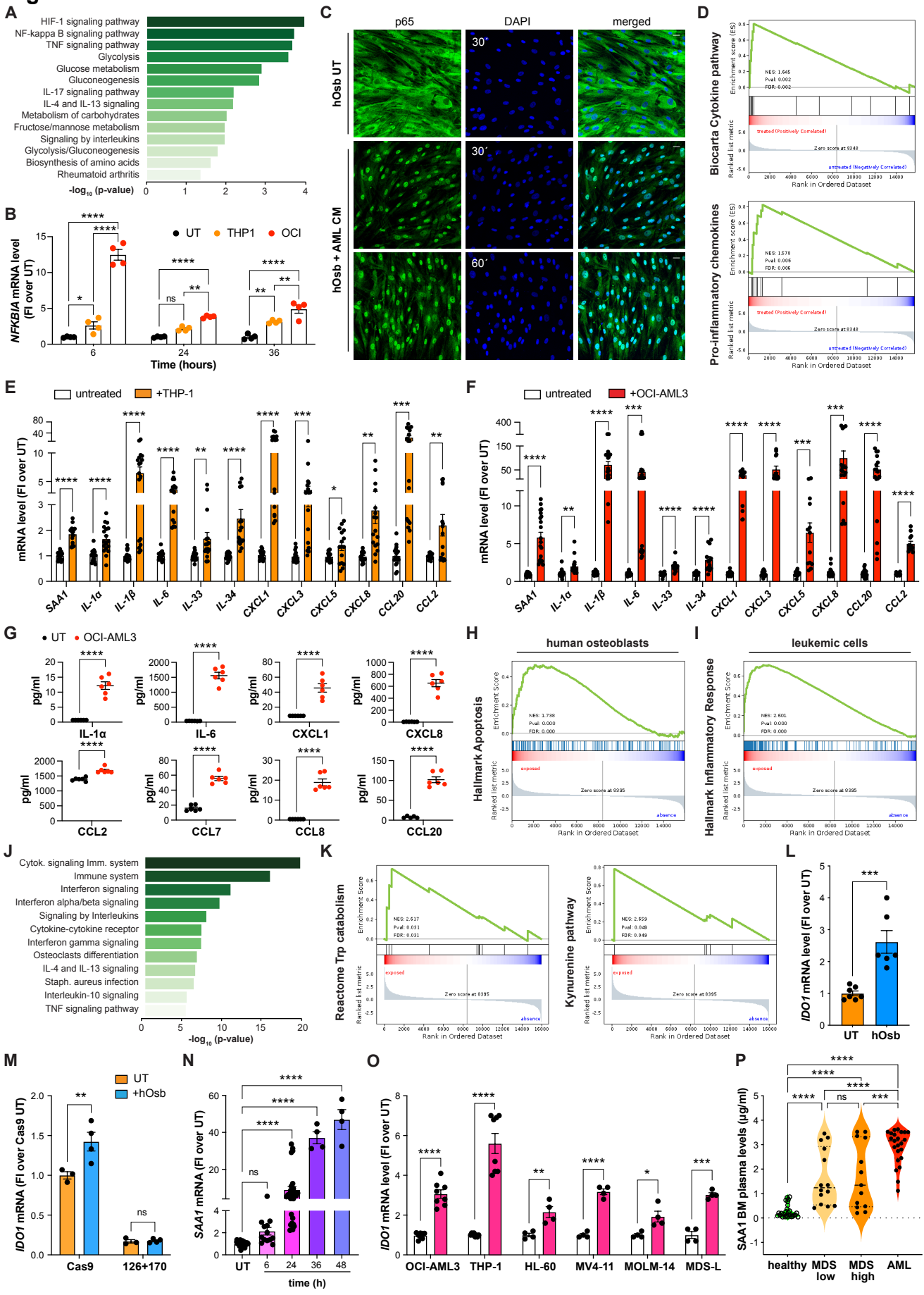


Figure S4



Supplementary Figure S4. AML cells induce a self-reinforcing niche that upregulates *IDO1* expression by eliciting SAA in osteoblasts. Related to Figure 4. (A) RNAseq data KEGG (Kyoto Encyclopedia of Genes and Genomes) pathway enrichment analysis of upregulated genes in primary human osteoblasts exposed to the AML cell-line THP-1. (B) *NFKB1A* mRNA relative level (fold induction -FI- over untreated -UT-) of primary human osteoblasts exposed to the THP-1 or OCI-AML3 cell lines for the indicated time points (n=4); two-way ANOVA. (C) Representative immunofluorescence images of primary human osteoblasts differentiated for 10 days and exposed to conditioned media (CM) for 30-60 min; p65 (green) and DAPI (nuclear stain, blue). Scale 66µm. (D) Gene Set Enrichment analysis (GSEA) plot for cytokines and pro-inflammatory chemokines. (E-F) mRNA relative level (FI over UT) of the indicated molecules in primary osteoblasts co-cultured with the human THP-1 (E) or the OCI-AML3 (F) AML cell-lines for 24h (n=12 replicates from 4 healthy human samples). (G) Protein level of the indicated molecules in supernatants from co-cultures (n=6). (H) GSEA analysis for apoptosis hallmark genes in human osteoblasts exposed to THP-1 cells for 24h. (I) GSEA analysis for inflammatory response hallmark genes in THP-1 cells exposed to human osteoblasts for 24h. (J) KEGG-database enrichment analysis of the upregulated genes in THP-1 AML cell-line exposed to primary human osteoblasts. (K) GSEA for tryptophan (Trp) catabolism and kynurenine pathway in the THP-1 cells exposed to primary human osteoblasts. (L) *IDO1* mRNA relative level in THP-1 cells exposed to primary human osteoblasts (n=6-7). (M) *IDO1* mRNA level in OCI-AML3 cells co-cultured 24h with primary human osteoblasts; (n=3-4), two-way ANOVA. (N) *SAA1* mRNA level in human osteoblasts exposed to OCI-AML3 cells for the indicated timepoints (UT n=48; 6h n=13; 24h n=30 and 36 and 48h n=4); one-way ANOVA. (O) *IDO1* mRNA relative level in the indicated human AML and MDS cells lines exposed o/n to SAA1; (n=4-8). (P) Kyn/Trp levels in BM plasma of healthy (n=27), MDS -stratified in low- (n=14) and high-risk (n=13) groups based on the Revised International Prognostic Scoring System (IPSS-R)- and AML (n=23) patients, one-way ANOVA. Statistical analysis done with unpaired t-test unless otherwise indicated. All data are presented as mean ± SEM.