

Figure S1. Survival analysis of the validation set.

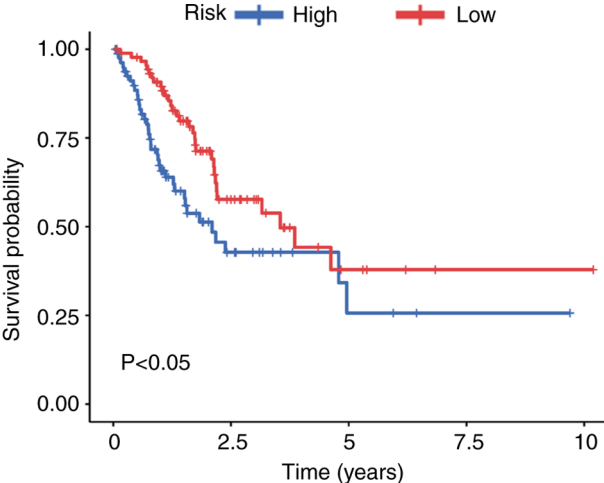


Figure S2. Differentially expressed gene enrichment analysis in high- and low-risk groups. (A) Volcano plot highlighting the DEGs between the high- and low-risk score groups [$\text{Log}(\text{FoldChange}) < 1$; $P < 0.05$]. (B) Gene set enrichment analysis of the data. (C) GO and (D) Kyoto Encyclopedia of Genes and Genomes functional enrichment analysis for DEGs. DEGs, differentially expressed genes; GO, Gene Ontology; BP, Biological Process; CC, Cellular Compartment; MF, Molecular Function.

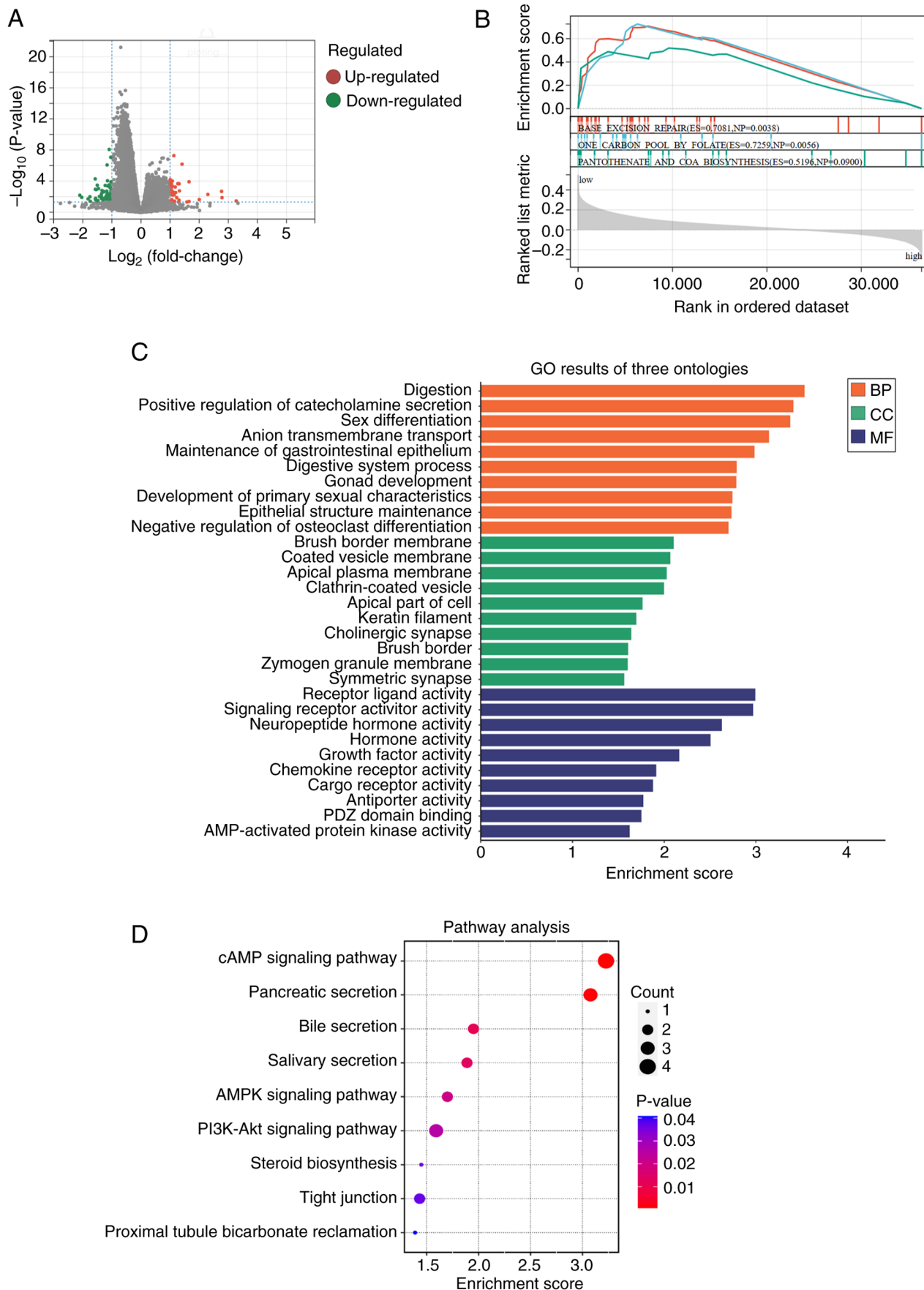


Figure S3. Proportions of *cytosolic iron-sulfur assembly component 1* genes in several cell types of primary and metastatic gastric cancers. NK, natural killer; RBC, red blood cell; pDC, plasmacytoid dendritic cell.

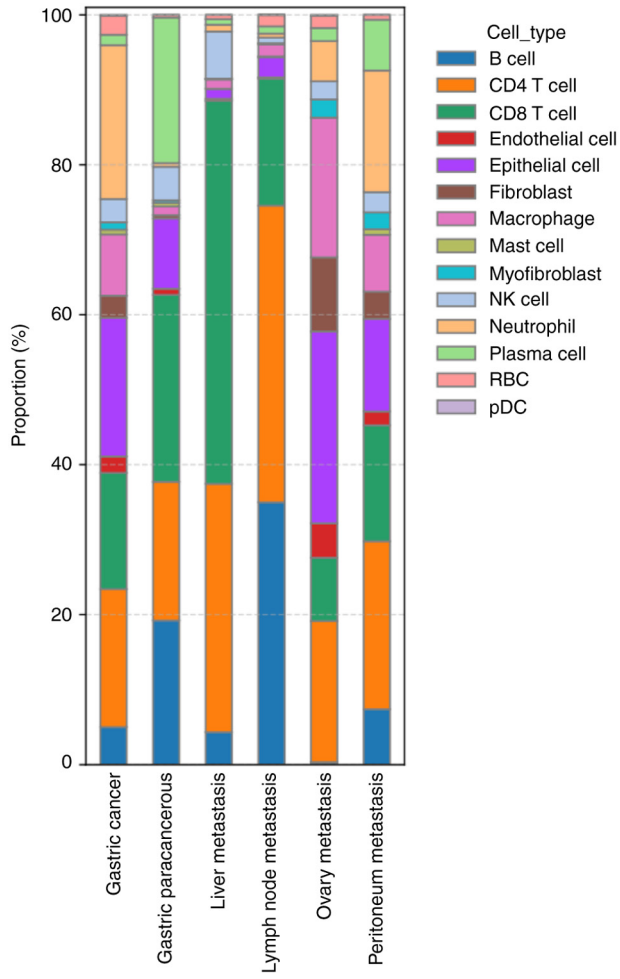


Figure S4. Immune, stromal and tumor purity scores of *CIAO1* in gastric cancer tissues. (A) Immune score, (B) stromal score and (C) tumor purity, comparing the high- and low-*CIAO1* expression groups. * $P < 0.05$ and ** $P < 0.01$. *CIAO1*, cytosolic iron sulfur assembly component 1.

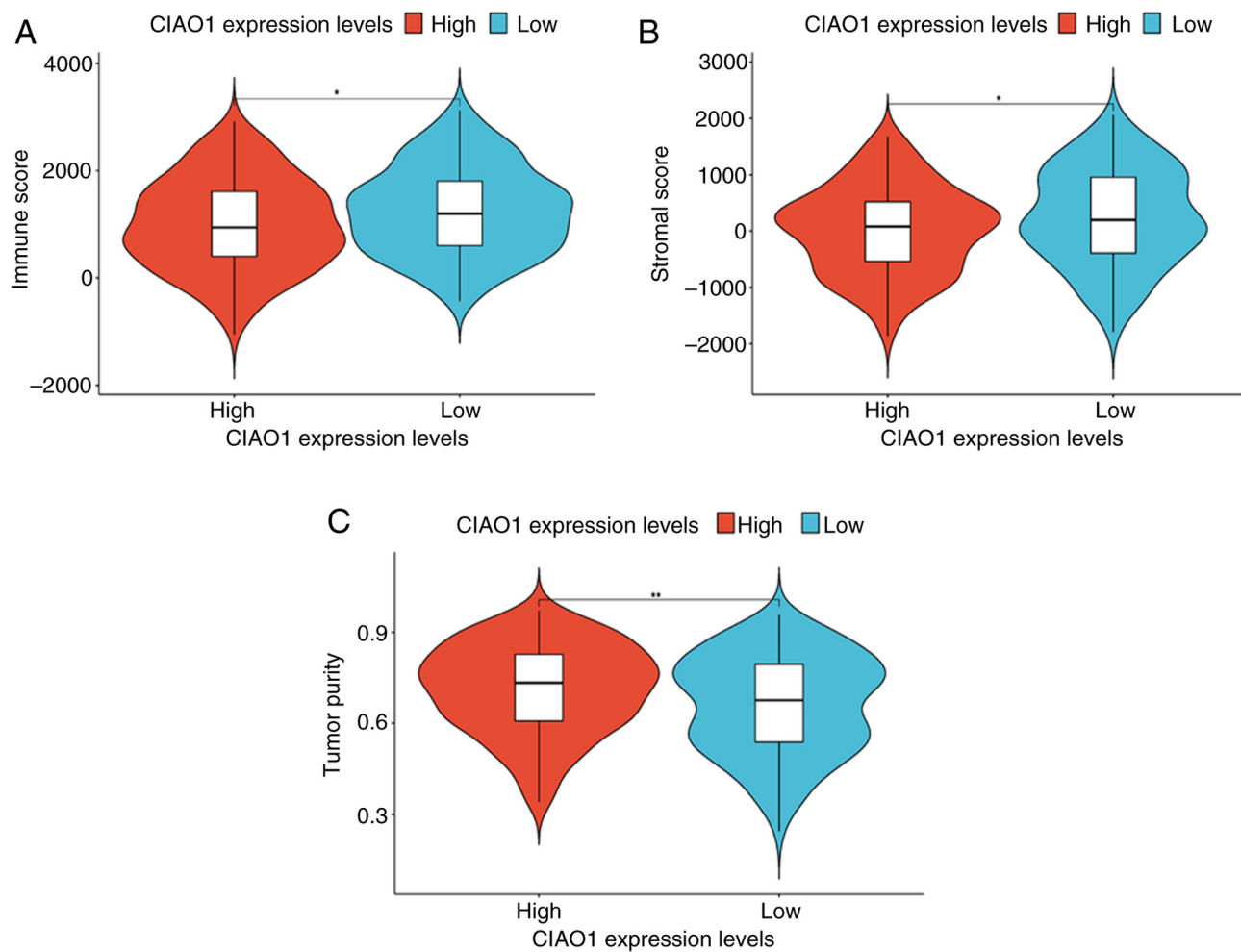


Figure S5. Molecular docking diagram of CIAO1 and its interacting proteins. (A) NUBP2, (B) POLD1, (C) SLC25A5, (D) SLC25A6, (E) ERCC2, (F) MMS19 and (G) NUBP1. CIAO1, cytosolic iron-sulfur assembly component 1; NUBP, NUBP Fe/S cluster assembly factor; POLD1, DNA polymerase $\Delta 1$ catalytic subunit; ERCC2, ERCC excision repair 2; SLC25, solute carrier family 25; MMS19, methyl methanesulfonate sensitivity gene 19.

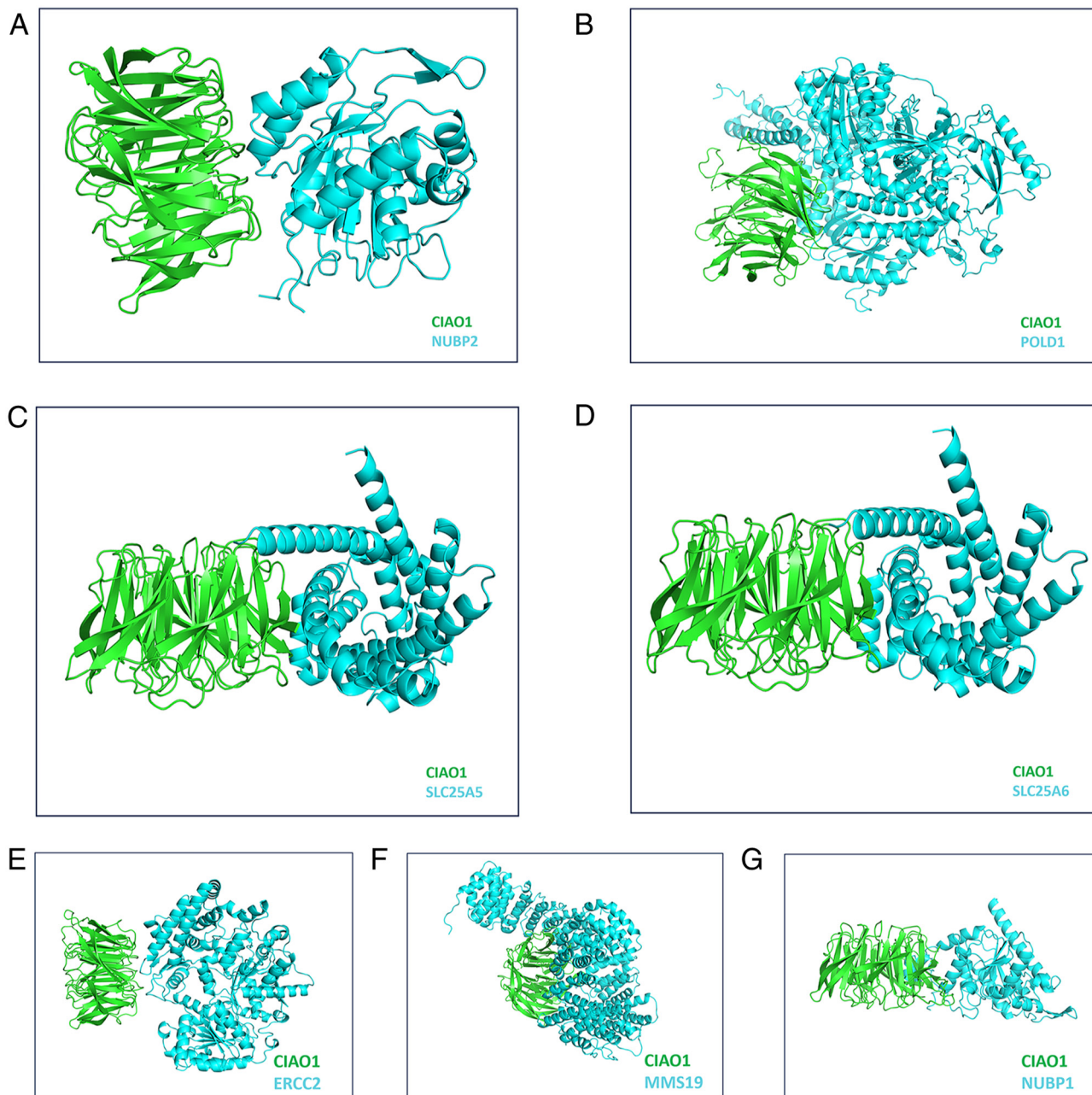


Figure S6. Pharmacological inhibition of CIAO1 induces cuproptosis and inhibits the aggressiveness of HeLa cells. (A) RT-qPCR assays (normalized to β -actin) revealed *CIAO1* expression levels, (B) western blot analysis demonstrated CIAO1, FDX1, LIAS and HSP70 expression levels and (C) immunofluorescence assays revealed localization and fluorescence intensity of CIAO1 expression (scale bar, 10 μ m) in HeLa cells transfected with sh-Scb, sh-*CIAO1* #1 or sh-*CIAO1* #2. (D) Western blot analysis demonstrated CIAO1, FDX1, LIAS and HSP70 expression levels in HeLa cells treated with dasatinib (0.5 nmol/l) or AT-9283 (4 nmol/l). MTT colorimetric assay revealed changes in the viability of HeLa cells (E) transfected with sh-Scb, sh-*CIAO1* #1 or sh-*CIAO1* #2, and (F) treated with dasatinib (0.5 nmol/l) or AT-9283 (4 nmol/l). Representative images (top panels) and quantification of the data (bottom panels) of (G) soft agar and (H) Matrigel invasion assays, showing the anchorage-independent proliferation and invasion of HeLa cells stably transfected with sh-Scb, sh-*CIAO1* #1 or sh-*CIAO1* #2. Magnification, x100x magnification. *** $P < 0.001$; **** $P < 0.0001$ vs. sh-Scb. CIAO1, cytosolic iron-sulfur assembly component 1; RT-qPCR, reverse transcription-quantitative PCR; sh, short hairpin; Scb, scramble; FDX1, ferredoxin 1; LIAS, lipionic acid synthetase; HSP70, heat shock protein 70.

