

Figure S1. Spatial annotation of cell types and markers for ESCC in tumor sections, as well as the corresponding H&E-staining sections.

Figure S2. Single-cell analyses characterize the landscape of ESCC participants with resistance to neoadjuvant therapy. (A) tSNE plot of individual cells colored by cell type. Overview of intercellular communication networks in post-nICT ESCC tissues from resistant patients, showing (B) the count of interactions and (C) interaction intensity among key cell populations. (D) Representative ligand–receptor networks in epithelial and T cells.

Figure S3. (A) Schematic illustration of the Transwell-based co-culture system, in which T cells were placed in the upper chamber and ESCC cells in the lower chamber. Flow-cytometry assays of tumor-cell apoptosis at ESCC:CD8⁺ T-cell ratios of (B) 1:5 and (C) 1:10.

Figure S4. CXCL10 enhances the antitumor efficacy of anti-PD1 therapy in an ESCC CDX model. (A) Schematic illustration of the ESCC CDX model and treatment regimen. (B) Representative images of mice from each treatment group. (C) Representative images of excised tumors collected at the study endpoint. (D) Body-weight changes during treatment. (E) Tumor-growth curves showing tumor volume over time. (F) Final tumor weights of each treatment group. Data are presented as mean ± SD. Statistical significance was determined using one-way ANOVA followed by multiple-comparison tests. *P < 0.05, ns, not significant.

Figure S5. Hallmark GSEA comparing high- and low-risk groups defined by the ornithine metabolism signature in TCGA-ESCC. The dot plot shows normalized enrichment scores (NES) for representative Hallmark pathways. High-risk patients exhibited significant enrichment of cell cycle, hypoxia, and interferon response pathways.

1 Figure S6. Stable isotope-labeled arginine tracing and targeted metabolomic analyses
2 following ODC inhibition. (A) Experimental design of the stable isotope-labeled
3 arginine tracing assay in the Eca-109–T-cell Transwell coculture system treated with
4 anti-PD1 in the presence or absence of DFMO. (B) Heatmap showing differentially
5 abundant metabolites identified by targeted LC–MS/MS between the anti-PD1 and
6 anti-PD1 + DFMO groups. (C) Metabolic pathway enrichment analysis of differential
7 metabolites between the anti-PD1 and anti-PD1 + DFMO groups.

8

9 Supplementary table 1 Characteristics of ESCC patients in this study.

10