

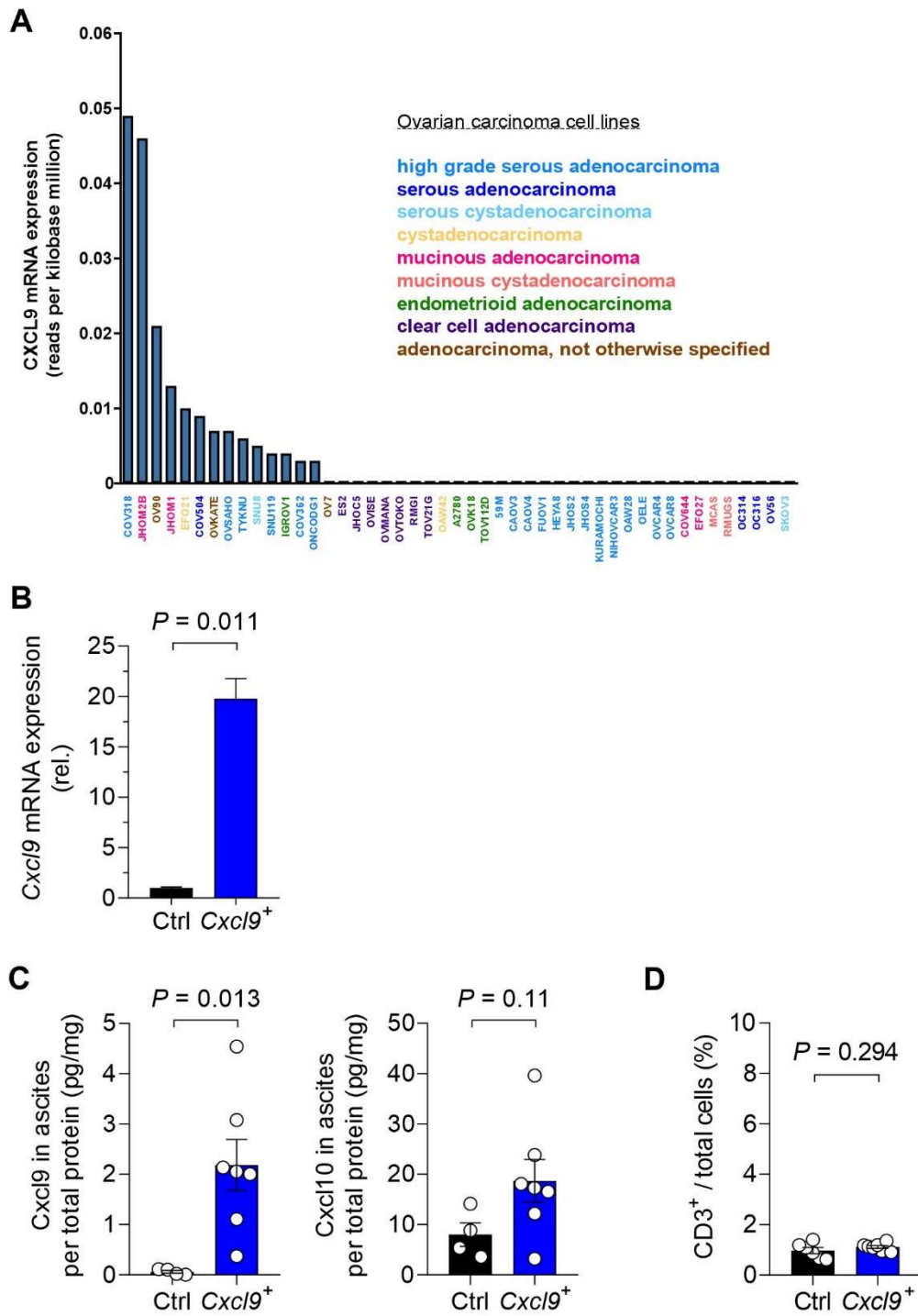
## Supplementary Material and Methods

**Quantitative reverse transcriptase PCR.** For relative gene expression analysis, RNA was extracted from cell pellets or granulated tumor tissue using the RNeasy® Plus Mini Kit (Cat. #74134) from Qiagen following the manufacturer's protocol. 1 µg of total RNA was used to synthesize cDNA via SuperScript™ IV First-Strand Synthesis System (Cat. #18091050) from Thermo Fisher Scientific. cDNA (150 ng) was amplified using Brilliant III Ultra-Fast QPCR Master Mix (Cat. #600890-51, Agilent Technologies), universal probes for murine Cxcl9 (Probe #1 Cat. #318501) and murine Hprt (Probe #95 Cat. #263935, Roche Molecular Systems) as well as corresponding primer pairs targeting the full-length proteins (mCxcl9 forward: 5'-GCA TCG ATT CCT TAT CA-3'; mCxcl9 reverse: 5'-CTT TTC CTC TTG GGC ATC AT-3'; mHprt forward: 5'-CCT CCT CAG ACC GCT TTT T-3'; mHprt reverse: 5'-AAC CTG GTT CAT CAT CGC TAA-3'; Metabion). For quantification of PD-L1 and Hprt, Thermo Fisher probes supplemented with associated primers were used (#Mm00452054\_m1 Cd274, #Mm03024075\_m1 Hprt, Thermo Fisher Scientific). Cycle parameters were as follows: 3 minutes: 95°C; 40X (15 s: 95 °C; 1 minute: 60°C). Gene expression levels were normalized to Hprt housekeeping gene and quantification was performed using the  $\Delta$ Ct calculation method.

**MTT proliferation assay.** Metabolic activity was compared by MTT assay. Briefly, cells were seeded at  $2 \times 10^3$  per well in 96-well plate triplicates within 70 µl cell culture media (DMEM with 5% FCS). After 4, 24 and 48 hours, 14 µl MTT reagent was added for 3 h at 37°C. Subsequently, cells were lysed by adding 150 µl DMSO and optical density was measured at 570 nm in a microplate reader.

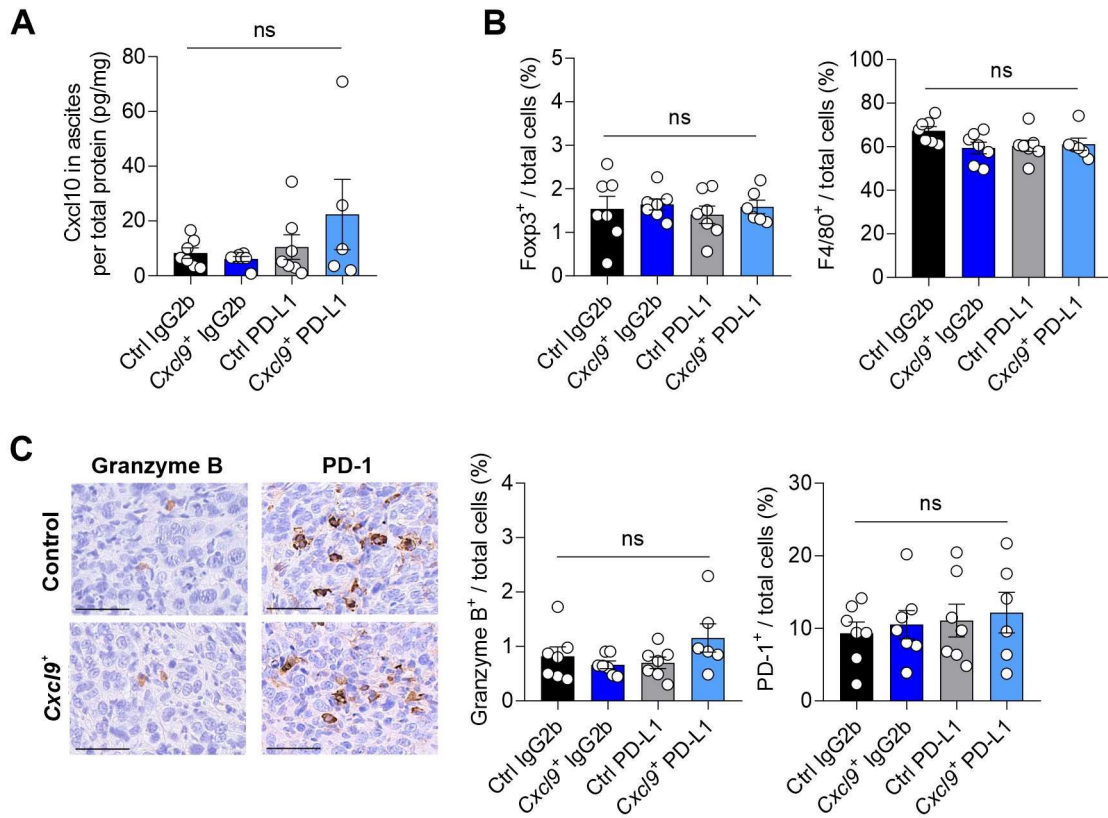
**Western Blot analysis.** Murine cells were collected as cell pellets without treatment. Human cells were seeded at 50% confluency overnight in 6-well plates and treated with 100 ng/mL recombinant human CXCL9 (Cat. #300-26, Peprotech) or 10 ng/mL human IFN $\gamma$  (Cat. #300-02, Peprotech) for 72 h. Protein concentrations of cell lysates were assessed using Pierce™

BCA Protein Assay Kit (Cat. #23225, Thermo Fisher Scientific) according to the manufacturer's instructions. 30 µg of cell lysates were separated on 12% SDS-polyacrylamide gels and electrotransferred to PVDF membranes using standard procedures. Membranes were blocked with 5% skim milk in TBS-Tween20 (0.05%) for 1 hour at room temperature. Primary antibody incubation was conducted overnight at 4°C with 1:600 polyclonal rabbit anti-PD-L1/CD274 (Cat. #17952-1-AP, Proteintech). Corresponding secondary goat-anti-rabbit IgG-HRP antibody (1:10000, Cat. #G21234, Thermo Fisher Scientific) was incubated for 1 hour at room temperature. Between incubation steps, membranes were washed 3 x 10 min with TBS-T. Blots were developed using Immobilon® Western Chemiluminescent HRP Substrate (Cat. #WBKLS0500, Millipore) and signals were detected using Bio-Rad ChemiDoc™ XRS+. GAPDH was used as a loading control (1:10000 monoclonal mouse anti GAPDH, Cat. #MAB374, Sigma-Aldrich and 1:10000 goat-anti-mouse IgG-HRP, Cat. #G21040, Invitrogen).

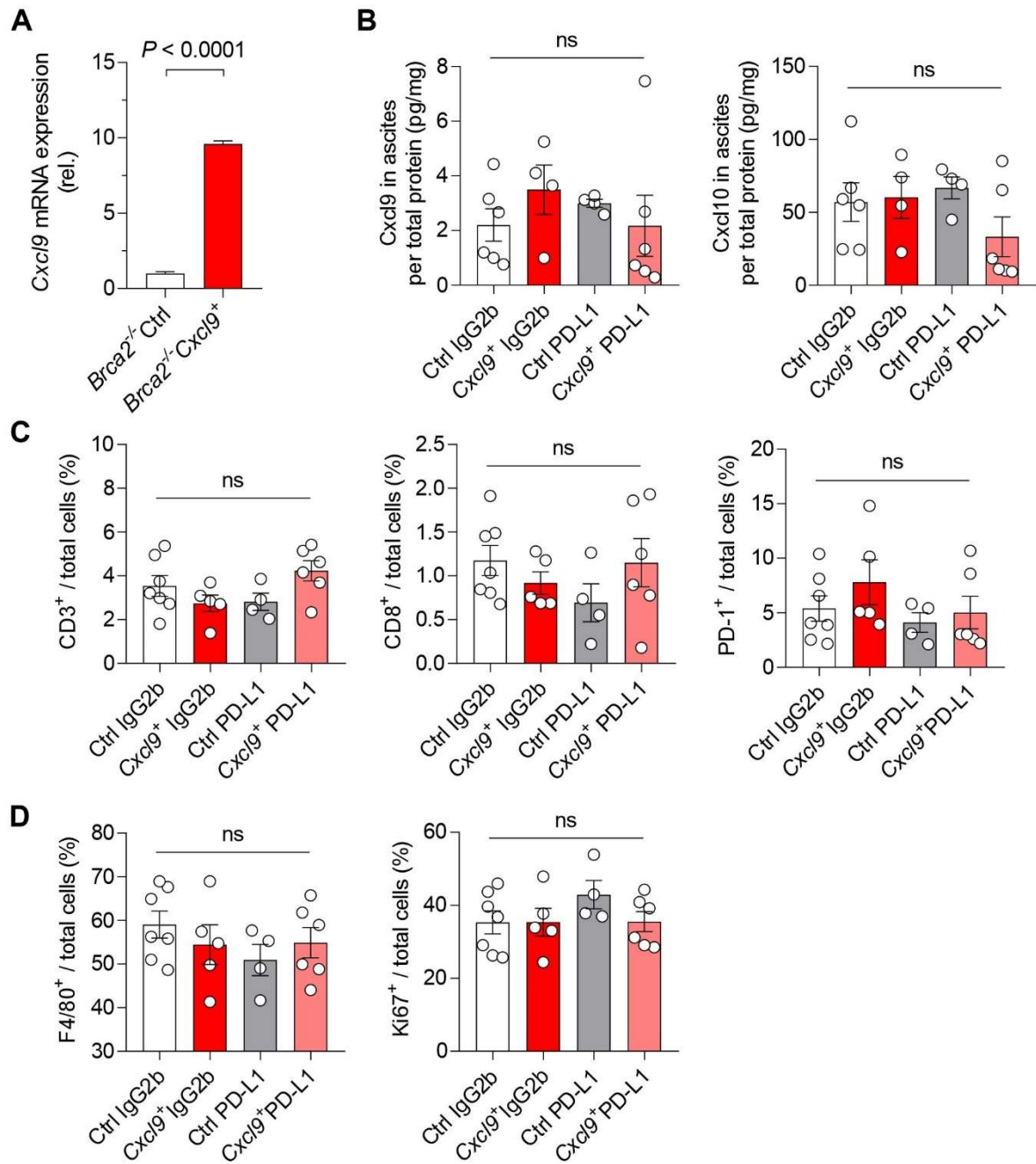


**Figure S1. A,** CXCL9 mRNA expression values given as “RPKM” (reads per kilobase million) in 47 different ovarian carcinoma cell lines including 8 distinct histological subtypes (publicly available data set was obtained from cBioPortal website). **B,** Verification of *Cxcl9* overexpression at RNA-level via quantitative PCR analysis of ID8-*Trp53*<sup>-/-</sup>Control (Ctrl) and ID8-*Trp53*<sup>-/-</sup>*Cxcl9*<sup>+</sup> (*Cxcl9*<sup>+</sup>) cells. **C,** Soluble *Cxcl9* or *Cxcl10* measured in ascites of athymic nude mice inoculated with ID8-*Trp53*<sup>-/-</sup>Control or *Cxcl9*<sup>+</sup> cells via ELISA and normalized to total

protein amount. **D**, Digital Analysis of intratumoral CD3<sup>+</sup> T-cells within ID8-*Trp53*<sup>-/-</sup>Control or *Cxcl9*<sup>+</sup> tumors in the athymic nude model. Bars represent mean  $\pm$  SEM. Each dot indicates data of one individual mouse.



**Figure S2. A**, Soluble Cxcl10 measured via ELISA in ascites of anti-PD-L1 or IgG2b treated mice stratified by Cxcl9 overexpression in the ID8-*Trp53*<sup>-/-</sup> model. Values were normalized to total protein amount. **B-C**, Digital analysis of immunohistochemically detected immune cell population markers (Foxp3, F4/80, Granzyme B, PD-1). Representative IHC pictures of Granzyme B and PD-1 staining are presented with 50  $\mu$ m scale bars. Each dot in A-C indicates data of one individual mouse. Bars represent mean  $\pm$  SEM.



**Figure S3. A**, Lentiviral overexpression of Cxcl9 in ID8-*Trp53*<sup>-/-</sup>*Brca2*<sup>-/-</sup> cells (*Brca2*<sup>-/-</sup>Cxcl9<sup>+</sup>) was tested via quantitative PCR and compared to ID8-*Trp53*<sup>-/-</sup>*Brca2*<sup>-/-</sup> empty vector (*Brca2*<sup>-/-</sup> Ctrl) cells. **B**, Soluble Cxcl9 or Cxcl10 in ascites samples of anti-PD-L1 or IgG2b treated mice stratified by Cxcl9 overexpression in the *Brca2*-deficient model was quantified by ELISA and normalized to total protein amount. **C-D**, Digital analysis of immunohistochemical markers (CD3, CD8, PD-1, F4/80, Ki-67) within the tumor microenvironment. Bars represent mean  $\pm$  SEM. Each dot indicates data of one individual mouse.