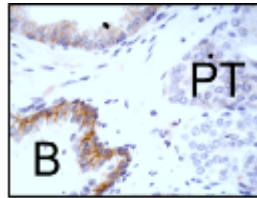


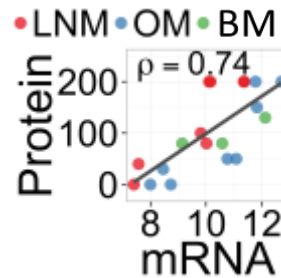
Supplementary Figures

- S1. CDCP1 protein expression and mRNA/protein correlation in a tissue microarray**
- S2. Gene set enrichment analysis of CDCP1 and EMT genes**
- S3. Original data of CDCP1 and E-cadherin surface expression in DU145 and PC3 cell lines**
- S4. CDCP1 Knock-down and model of CTCs**
- S5. Xenografts from CDCP1 positive and negative DU145 cells**
- S6. CDK5 kinase assay and TALIN complex**

A



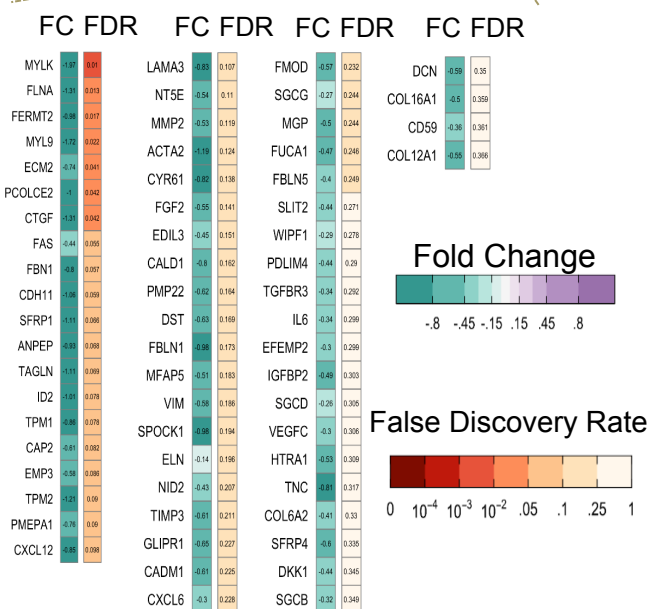
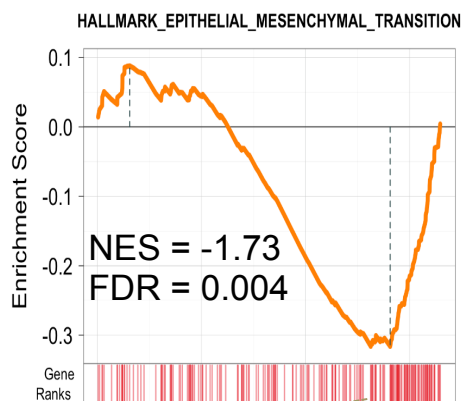
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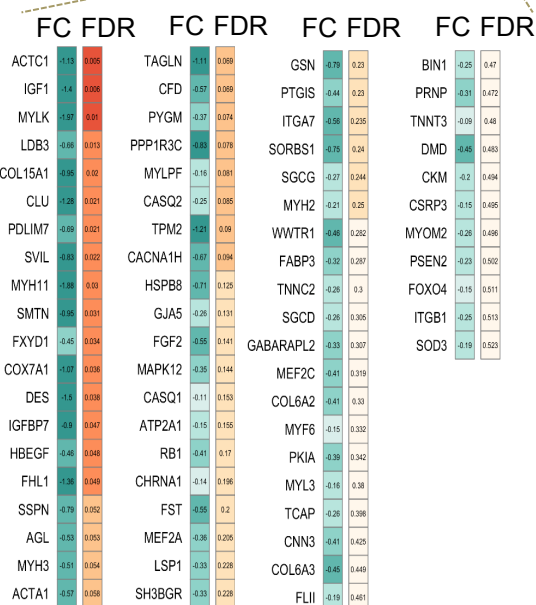
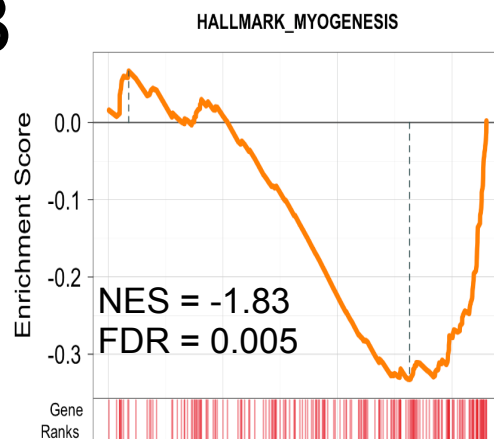
Supplementary Figure 1. CDCP1 protein expression and mRNA/protein correlation in a tissue microarray

A. CDCP1 IHC in a tissue section of benign prostate gland (B) and prostate tumor (PT). **B.** LuCaP xenografts were stained for CDCP1 protein expression and analyzed for CDCP1 mRNA. The correlation between CDCP1 mRNA and protein expression in xenografts derived from lymph node metastasis (LNM), other metastasis (OM), and bone metastasis (BM). Y-axis protein expression calculated by multiplying intensity of antibody staining (0, 1, 2) times the fraction of cells that stained (scale 0 – 100)

A

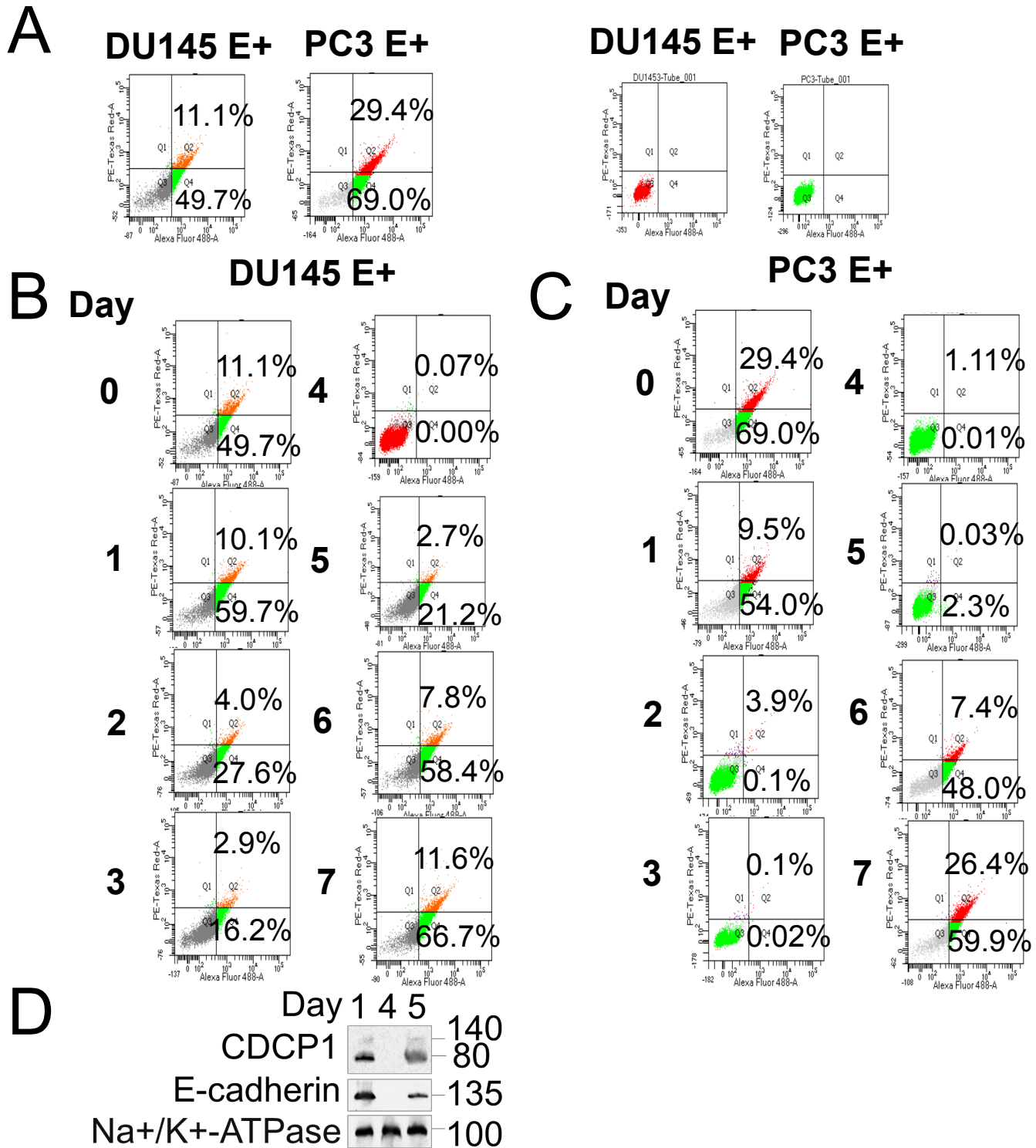


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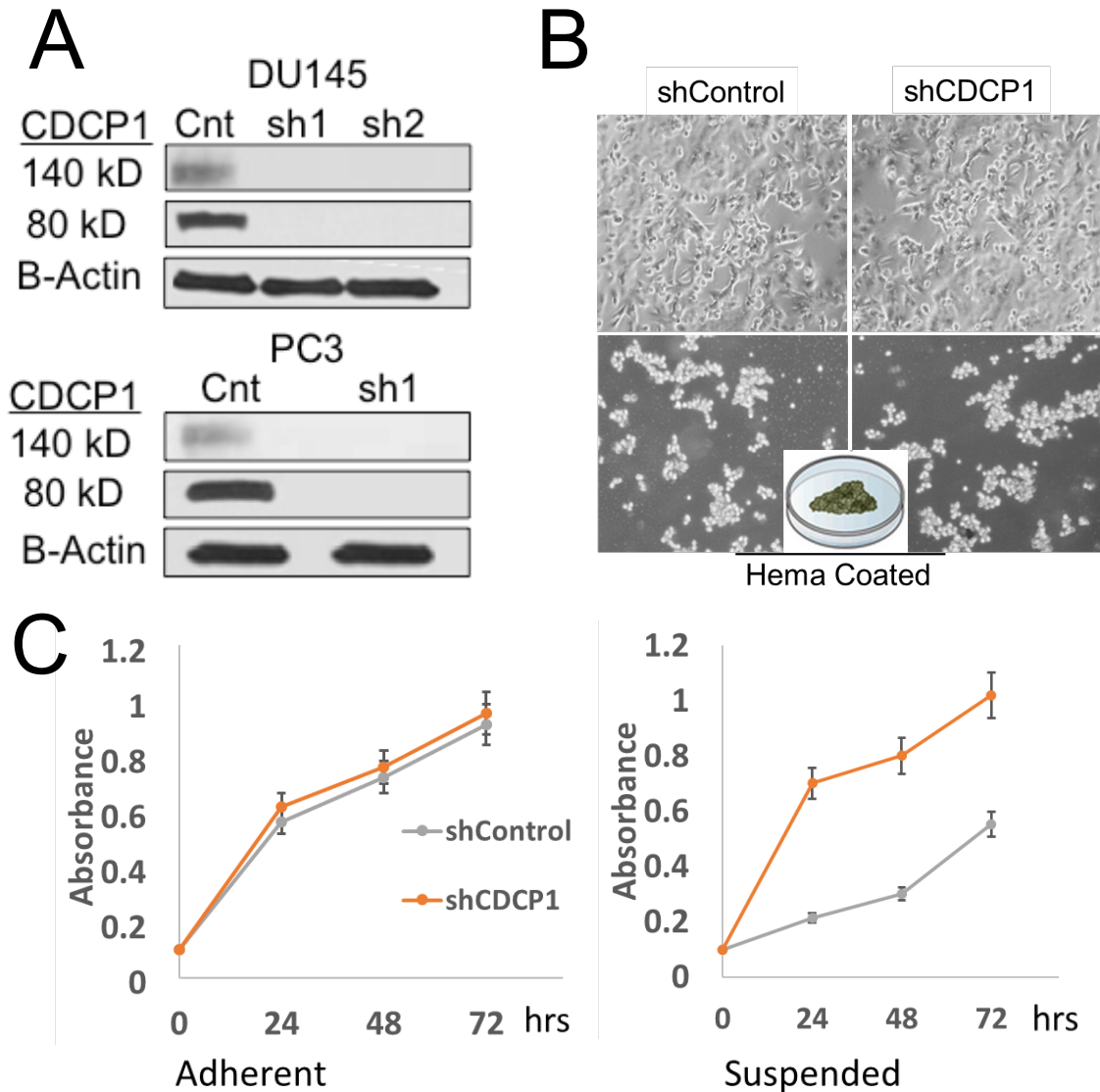


Supplementary Figure 2. Gene set enrichment analysis of CDCP1 and EMT genes
A. GSEA of hallmark EMT transition signature and **B.** hallmark myogenesis signature in prostate cancer metastasis. The identity, fold change (FC) and false discovery rates (FDR) of leading edge genes are indicated underneath the graph; NES, normalized enrichment score.

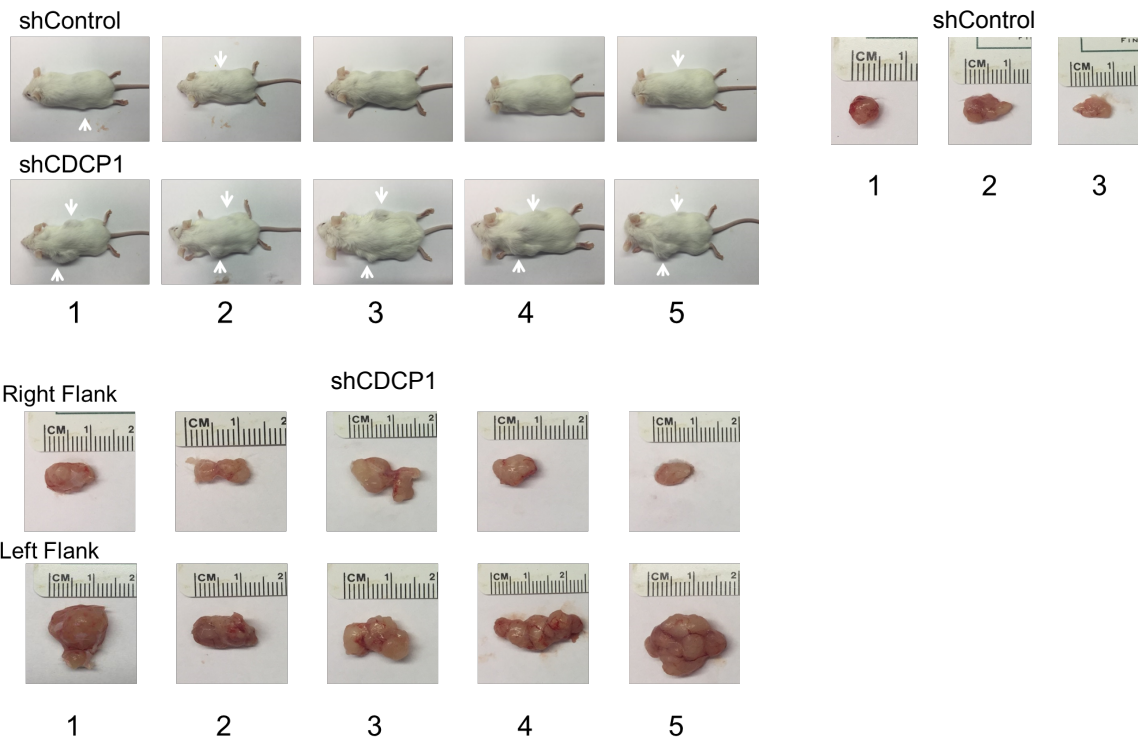
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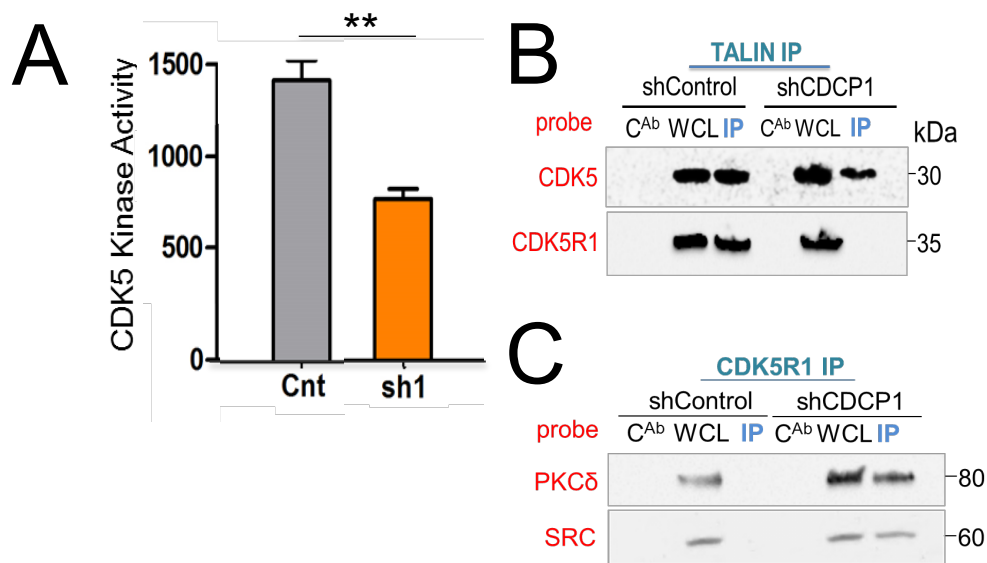
Supplementary Figure 3. Raw data of CDCP1 and E-cadherin surface expression in DU145 and PC3 cell lines
A. Scatterplots from FACS analysis of E-cadherin Alexa Fluor 488 and CDCP1 Alexa Fluor 555 double-stained DU145 E-cadherin + and – cells used in the graph of Figure 2 B. **B.** FACS analysis of surface E-cadherin Alexa Fluor 488 and CDCP1 Alexa Fluor 555 stained DU145 E-cadherin + cells. Cells were treated for 4 days with 10ng/mL TGFβ1, the the treatment was removed for last 3 days of the 7-day time course. Data were used to generate the graph in Figure 2C. **C.** FACS analysis of PC3 E-cadherin + cells for data generation in Figure 2C. **D.** Western blot of DU145 E+ membrane fraction in TGFβ1 timecourse days 1, 4 and 5 as in **B.** probed with antibodies reactive to CDCP1, E-cadherin and Na+/K+-ATPase.



Supplementary Figure 4. CDCP1 Knock-down and model of CTCs, growth curve of CDCP1 positive (shControl) and negative cells (shCDCP1) **A.** Western blot of DU145/shControl (Cnt), shCDCP1 hairpin #1 (sh1) and shCDCP1 hairpin #2 probed for CDCP1 and B-Actin expression to demonstrate the efficiency of the CDCP1 silencing with 2 CDCP1 hairpins. Western blot of PC3 cells transfected with PC3/shControl (Cnt) or shCDCP1 hairpin #1 and probed for CDCP1 and B-Actin expression. **B.** DU145 shControl and shCDCP1 cells in adherent culture and suspended on poly-HEMA coated plates. **C.** Growth curve of DU145 E- cells adherent on cell culture plates or suspended on HEMA coated plates as in **B.** at 24, 48 and 72 hours.



Supplementary Figure 5. Xenografts from CDCP1 positive and negative DU145 cells
A. Pictures of mice with bi-lateral subcutaneous xenografts from injected 1×10^6 DU145/shControl and DU145/shCDCP1 cells per flank after 9 weeks. Arrows indicate visible tumors **B.** Excised tumors from shControl and shCDCP1 injected mice.



Supplementary Figure 6. CDK5 kinase assay and TALIN complex. CDK5 kinase activity and complex formation with TALIN are altered with CDCP1 loss. **A.** Kinase assay of CDK5 activity in DU145/shControl and DU145/shCDCP1 suspended for three hours. Experiment repeated in triplicate. **B.** TALIN immunoprecipitation in DU145/shControl (shControl) and DU145/shCDCP1 (shCDCP1). IgG Control (C^{Ab}), whole cell lysate (WCL) and TALIN IP (IP) probed with antibodies reactive to CDK5 and CDK5R1. **C.** CDK5R1 immunoprecipitation in DU145/shControl (shControl) and DU145/shCDCP1 (shCDCP1). IgG Control (C^{Ab}), whole cell lysate (WCL) and CDK5R1 IP (IP) probed with antibodies reactive to PKC δ and SRC.