**Supplementary Figure legends:**

**Supplementary Figure 1:** Representative malignant (CA) and benign (BE) tissue cores of two patients with low and high Robo 4 and Slit 2 expression, scale bar = 100 μm.

**Supplementary Figure 2:** Representative pictures of CD31 expression in malignant (CA) and benign (BE) tissue cores of patients with low and high grade prostate cancer, scale bar = 100 μm.

**Supplementary Figure 3:**
Robo 4 mRNA expression using qRT-PCR after A) upregulation and B) downregulation of Robo 4; C) Protein expression (Western blot) after Robo 4 overexpression in PC3 cells. Data represent mean + SEM from at least 3 independent experiments, *p<0.05; **p<0.01; ***p<0.001.

**Supplementary Figure 4:** Apoptosis (flow cytometry) measurements of PC3 after (A) Robo 4 knockdown and (B) overexpression. Data represent mean + SEM from at least 3 independent experiments; ns = not significant.
Supplementary Figure 1
Supplementary Figure 2

CD 31

Benign

high grade PCa

Cancer

low grade PCa

100µm
Supplementary Figure 3

Panel A: Image of cells stained with blue and green fluorescence.

Panel B: Bar graph showing the expression levels of Robo4 mRNA in PC3 cells transfected with empty vector or plasmid. The expression levels are compared using a 2^-dCT method with Robo4- (TBP+HPRT1) as a reference. The graph includes statistical significance with asterisks.

Panel C: Bar graph illustrating the expression levels of Robo4 mRNA in PC3 cells treated with control (Crl) or siRNA. The expression levels are compared using a 2^-dCT method with Robo4- (TBP+HPRT1) as a reference. The graph includes statistical significance with asterisks.

Panel D: Western blot analysis showing the expression of Robo4 and GAPDH in LNCaP, PC3, and DU145 cells transfected with empty vector or plasmid.
Supplementary Figure 4

A. PC3

B. PC3

% apoptosis

ns

ns

Ctrl siRNA

Empty vector Plasmid

0.0 0.5 1.0 1.5 2.0

% apoptosis

0.0 0.5 1.0 1.5 2.0 2.5 3.0 3.5 4.0 4.5 5.0 5.5 6.0 6.5 7.0 7.5 8.0

ns