

Supplementary Figures

Loss of PGRMC1 delays the progression of hepatocellular carcinoma via suppression of pro-inflammatory immune responses

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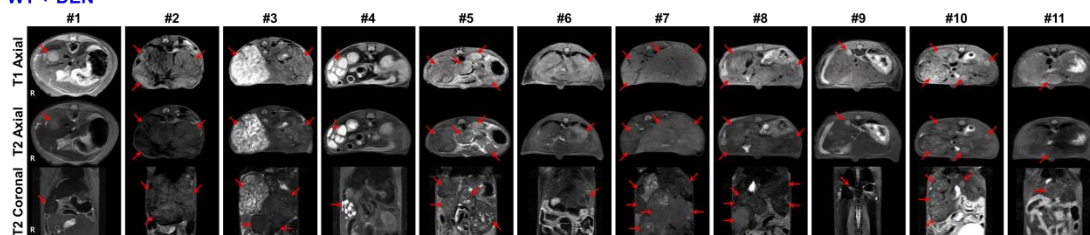
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WT + DEN



Pgrmc1-null + DEN

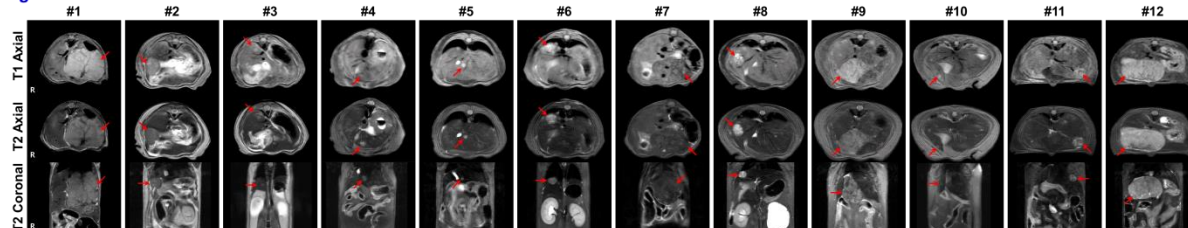


Figure S1. Magnetic resonance images (T1w [axial], T2w [axial and coronal]) of HCCs in HCC-bearing WT (n=11) and *Pgrmc1*-null (n=12) mice. Arrows denote the tumors.

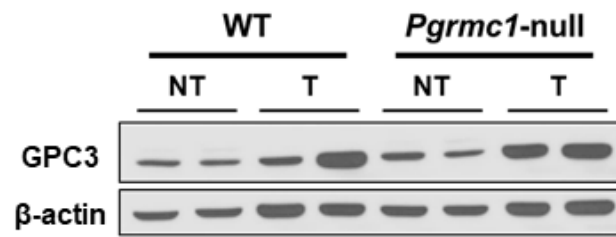


Figure S2. Western blot analysis of Glypican-3 (GPC3) in non-tumor lesions and tumors of WT and *Pgrmc1*-null mice. β-actin was used as an internal control. NT, non-tumor; T, tumor region

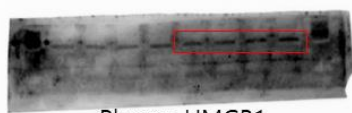


Fig. 3C

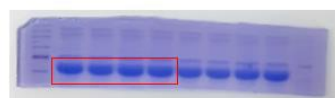


Fig. 3C

Plasma albumin



Fig. 4A

PGRMC1



Fig. 4A

EGFR



Fig. 4A

pEGFR



Fig. 4A

β-actin



Fig. 6A

PGRMC1



Fig. 6A

EGFR



Fig. 6A

pEGFR



Fig. 6A

IκBα

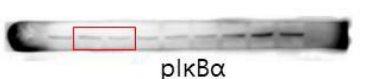


Fig. 6A

pIκBα



Fig. 6A

NF-κB (p65)



Fig. 6A

pNF-κB (p65)



Fig. 6A

β-actin



Fig. 4C

PGRMC1



Fig. 4C

EGFR



Fig. 4C

pEGFR



Fig. 4C

β-actin



Fig. 4D

PGRMC1



Fig. 4D

EGFR



Fig. 4D

pEGFR



Fig. 4D

β-actin



Fig. 6C

PGRMC1



Fig. 6C

EGFR

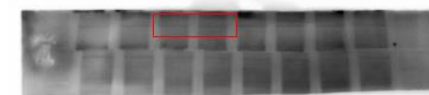


Fig. 6C

pEGFR



Fig. 6C

IκBα



Fig. 6C

pIκBα

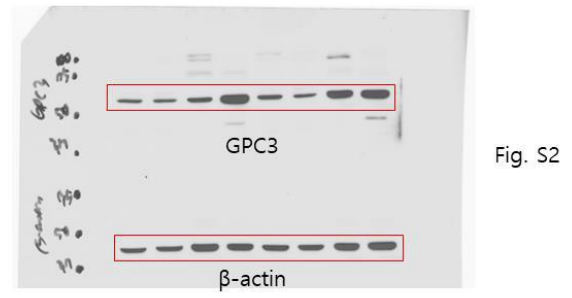
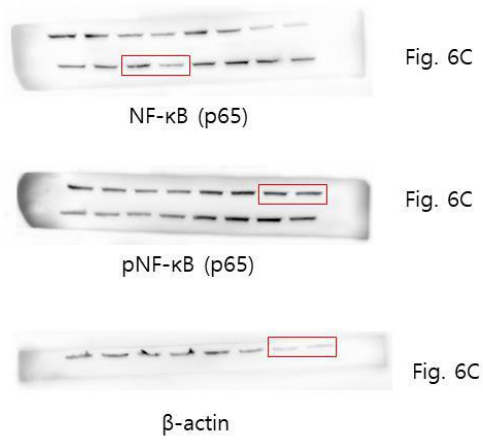


Figure S3 Full blot images.

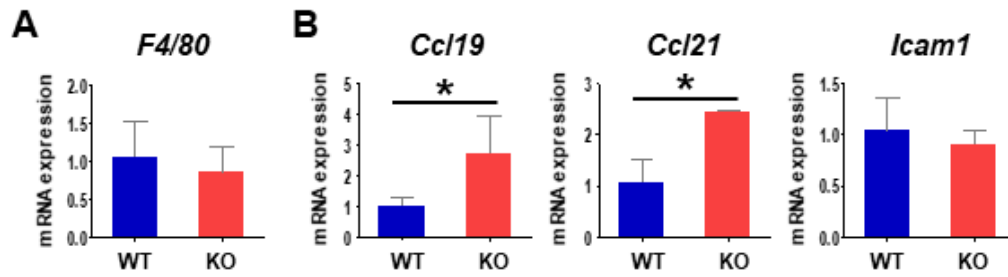


Figure S4. mRNA expression levels of *F4/80*, *Ccl19*, *Ccl21*, and *Icam1* in the livers of DEN-injected WT and *Pgrmc1*-null mice. The expression levels were normalized to that of *Rplp0* and expressed relative to the WT group. * $p < 0.05$ in Student's *t*-test.

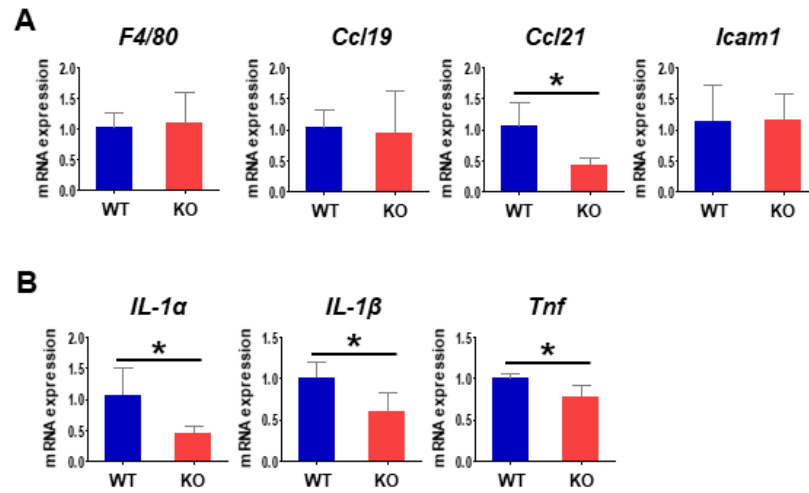


Figure S5. mRNA expression levels of *F4/80*, *Ccl19*, *Ccl21*, *Icam1*, *IL-1α*, *IL-1β*, and *Tnf* in the HCC of DEN-injected WT and *Pgrmc1*-null mice. The expression levels were normalized to that of *Rplp0* and expressed relative to the WT group. * $p < 0.05$ in Student's *t*-test.

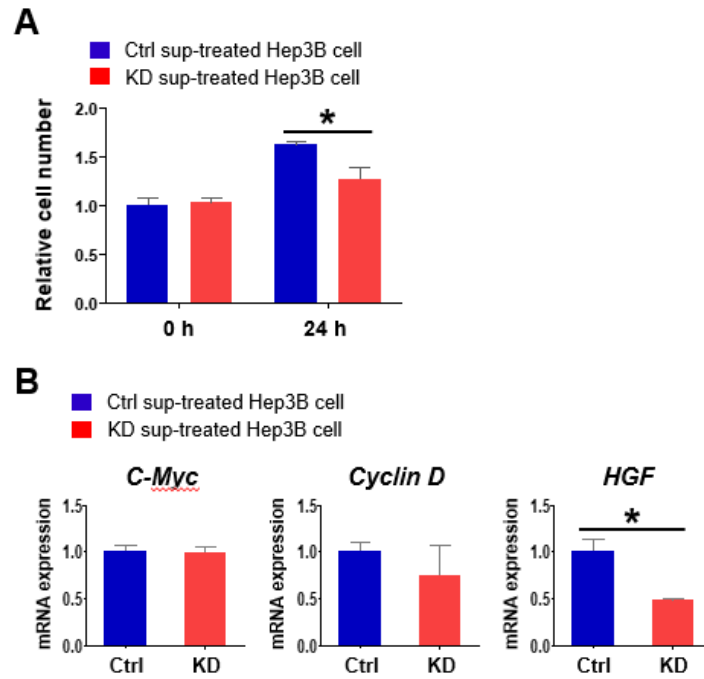


Figure S6. Cancer cell proliferation measured after treatment of Raw264.7 cell culture supernatant. (A) Cell number counted by trypan blue staining in 0 and 24 h treatment of Raw264.7 cell culture supernatant to Hep3B cells. FBS was excluded in Hep3B cell culture medium. Cell culture supernatant was collected from Control (Ctrl) or Pgrmc1-siRNA (KD)-transfected Raw 264.7 cells treated with necrotic debris for 3 h. (B) mRNA expression levels of *C-Myc*, *Cyclin D*, and *HGF* in Hep3B cells cultured with Raw 264.7 cell culture supernatant. Cell culture supernatant was collected from Control (Ctrl) or Pgrmc1-siRNA (KD)-transfected Raw 264.7 cells treated with necrotic debris for 3 h. Cell culture supernatant was incubated for 3 h in Hep3B cells. * $p < 0.05$ in Student's *t*-test.

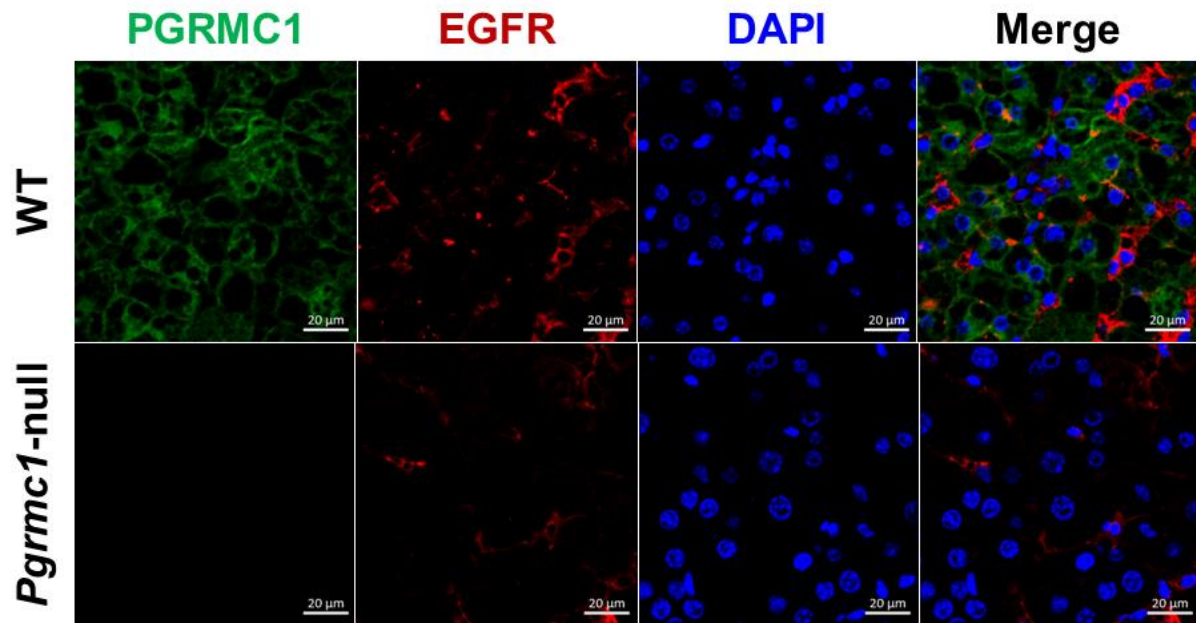
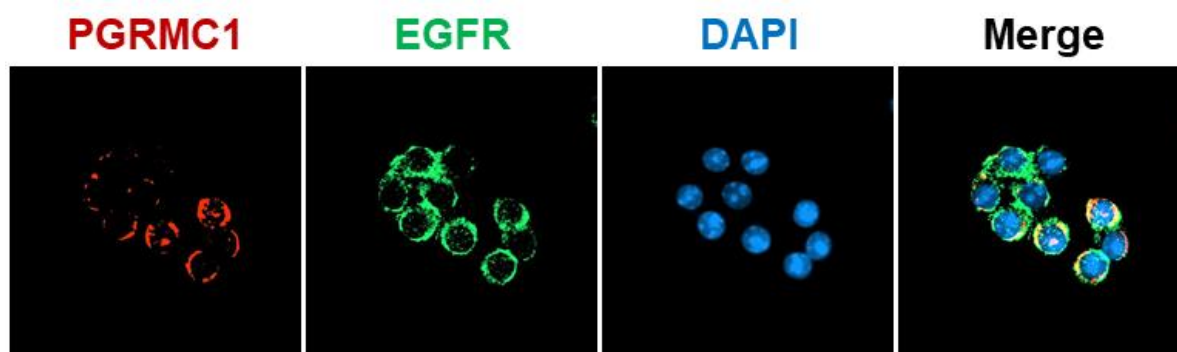


Figure S7. Co-immunostaining of PGRMC1 and EGFR in the HCC of WT mice and *Pgrmc1*-null mice. PGRMC1 (green) and EGFR (red) were stained with the corresponding antibodies. DAPI (blue) was used for nuclear staining.



Raw264.7 cell

Figure S8. Co-immunostaining of PGRMC1 and EGFR in Raw 264.7 cell. PGRMC1 (red) and EGFR (green) were stained with the corresponding antibodies. DAPI (blue) was used for nuclear staining.