

### **Additional file 3**

#### **Figure S3 IL-17 activates AKT and IL-6/STAT3, and up-regulates proinvasive**

**factors production in Huh7 cells. (A)** Western blotting showed that phosphorylation

of JAK2, STAT3 and AKT were obviously increased as early as 3 h after IL-17

treatment and lasted for 24 h after IL-17 stimulation. Huh7 cells were incubated with

IL-17 at the indicated concentrations for 24 h or at 50 ng/ml for the indicated time. **(B**

**and C)** Cells were cultured for 24 h with IL-17 (50 ng/ml). In siRNA-STAT3-Huh7

cells, IL-17-induced AKT and JAK2 phosphorylation were not affected, while in

siRNA-AKT- Huh7, IL-17-induced JAK2/STAT3 phosphorylation was significantly

reduced. **(D)** HCC cells were cultured for 24 h with IL-17 (50 ng/ml) and/or IL-6

mAb (10 ng/ml), and concentrations of the proinvasive factors in culture supernatants

were measured by ELISA. IL-17 selectively up-regulated the production of IL-6, IL-8,

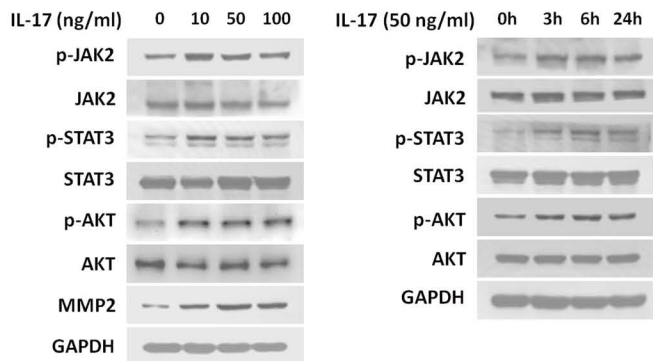
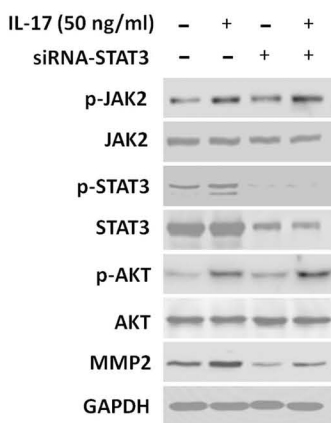
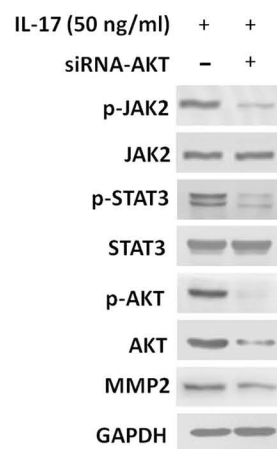
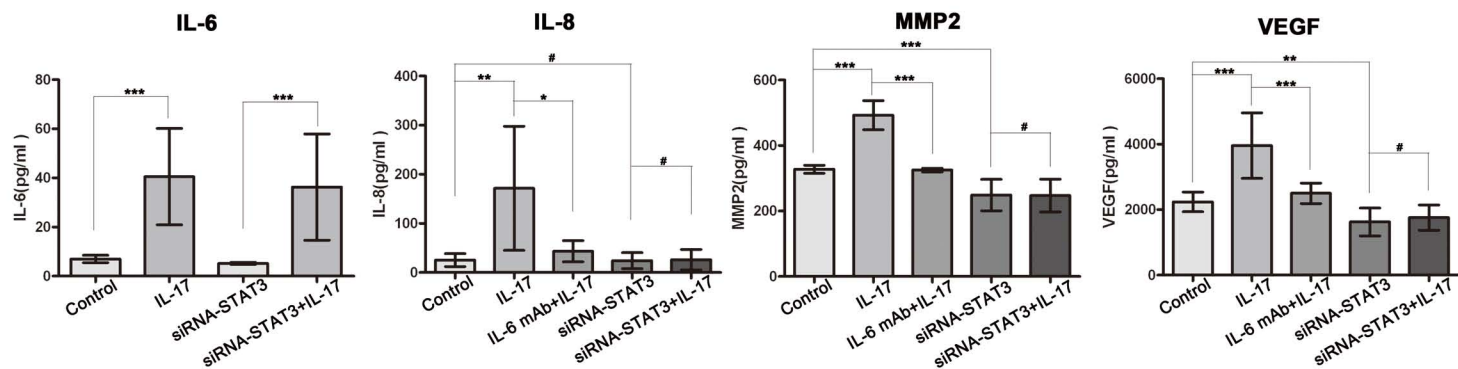
MMP2 and VEGF by tumor cells. Both IL-6 mAb and siRNA-STAT3 significantly

downregulated the expression of IL-8, MMP2 and VEGF, while IL-17-induced IL-6

upregulation was not altered. **(E)** IL-6 mAb reduced STAT3 activation, whereas AKT

activation by IL-17 was not affected. Data are expressed as mean  $\pm$  SD; Student's *t*

test; #  $p > 0.05$ ; \* $p < 0.05$ , \*\* $p < 0.01$ , and \*\*\* $p < 0.001$ .

**A****B****C****D****E**