

Tubular STAT3 limits renal inflammation in autosomal dominant polycystic kidney disease.

Supplementary Material

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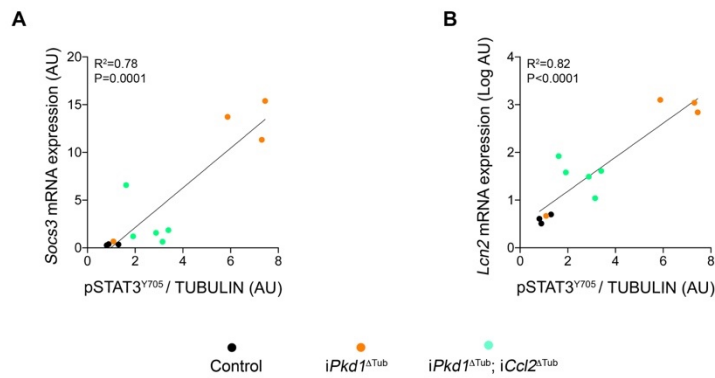
Supplementary Figure 1: Correlation between STAT3 phosphorylation and *Soes3* and *Lcn2* mRNA in kidneys from 13 old control, *iPkd1*^{ΔTub} and *iPkd1*^{ΔTub}; *iCcl2*^{ΔTub} mice.

Supplementary Figure 2: Primary cilia promote STAT3 phosphorylation in tubular cells *in vitro*.

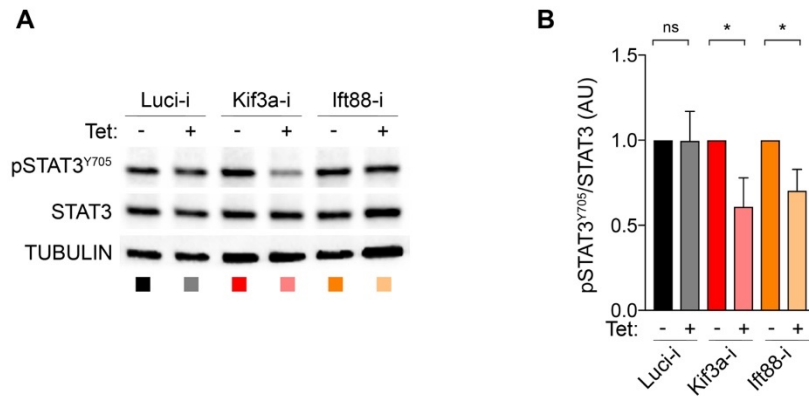
Supplementary Figure 3: STAT3 ablation does not reduce cell proliferation or increased apoptosis.

Supplementary Figure 4: STAT3 disruption has a selective impact on renal inflammation in ADPKD.

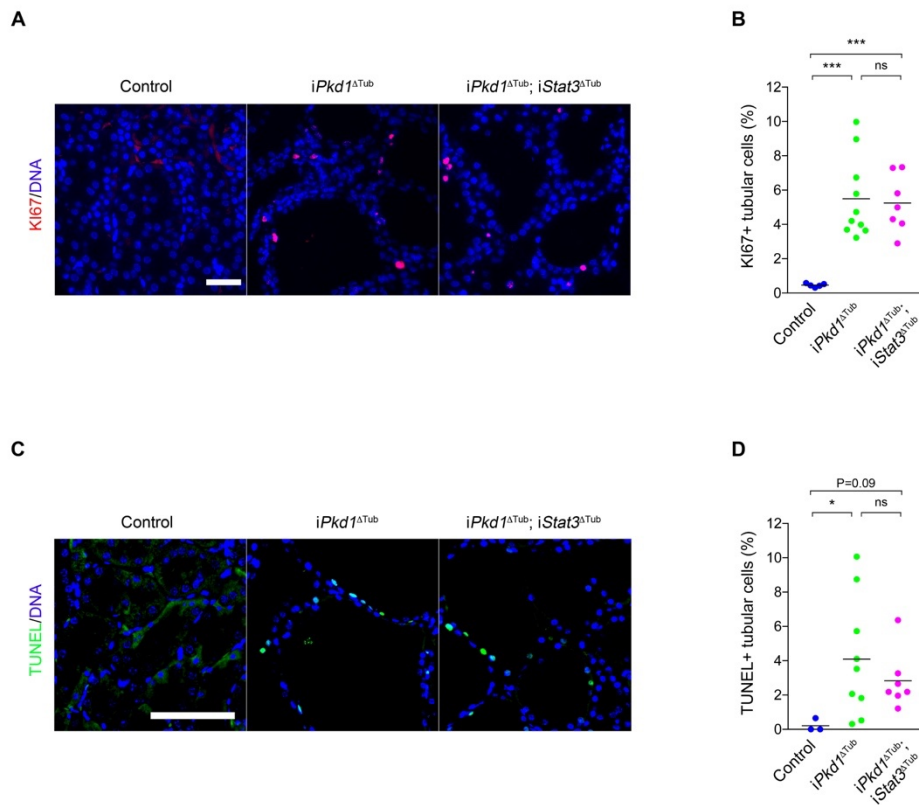
Supplementary Table 1: Primers used for qRT-PCR.



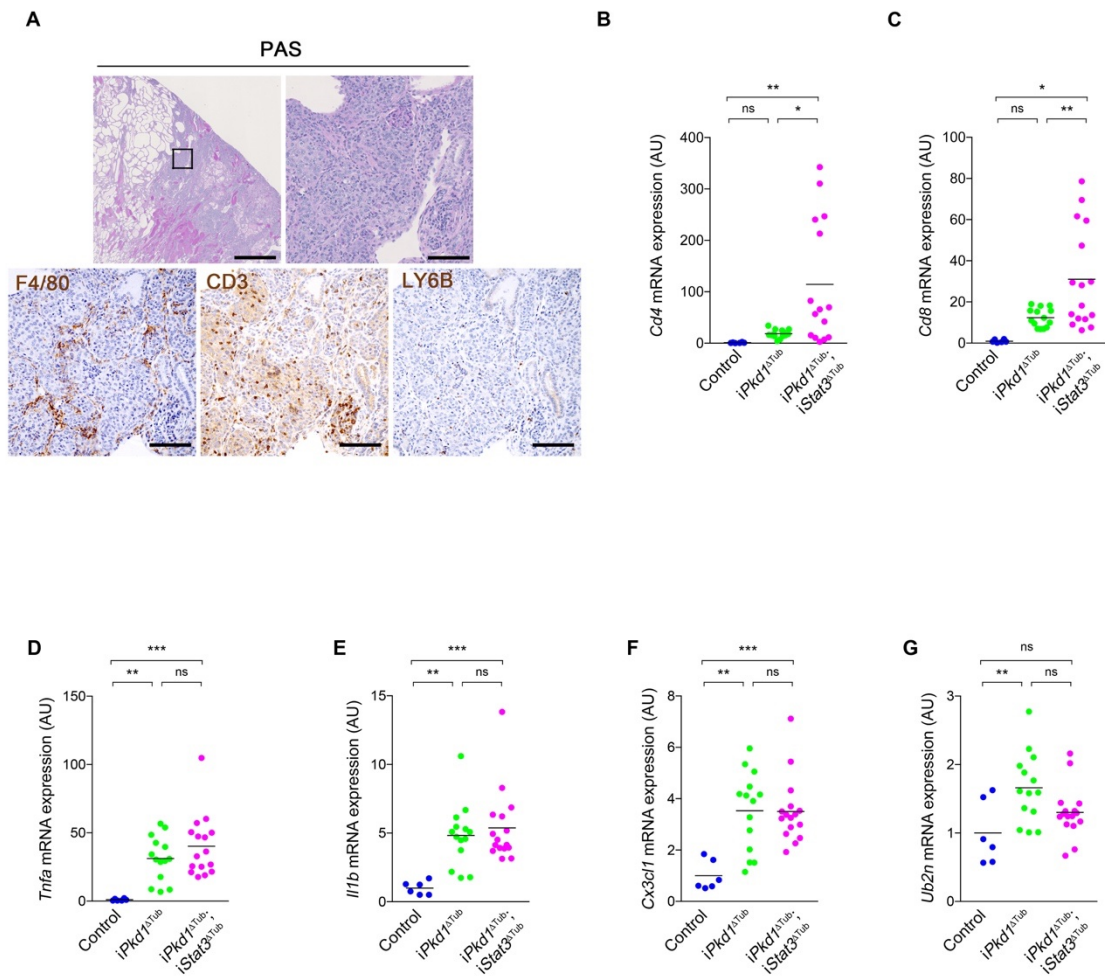
Supplementary Figure 1: Correlation between STAT3 phosphorylation and *Socs3* and *Lcn2* mRNA in kidneys from 13 old control, *iPkd1*^{ΔTub} and *iPkd1*^{ΔTub}; *iCcl2*^{ΔTub} mice. (A- B) Correlation between pSTAT3^{Y705} amount and the expression of *Socs3* (A) and *Lcn2* (B) mRNA in kidneys from 13 weeks old control, *iPkd1*^{ΔTub} and *iPkd1*^{ΔTub}; *iCcl2*^{ΔTub} mice. AU: arbitrary units.



Supplementary Figure 1: Primary cilia promote STAT3 phosphorylation in tubular cells *in vitro*. (A, B) Representative western blot (A) and quantification (B) of STAT3 phosphorylation in whole cell lysates from MDCK cells expressing inducible shRNA against *Kif3a* (Kif3a-i), *Ift88* (Ift88-i) or the non-relevant control luciferase (Luci-i) after 10 days of tetracyclin treatment (+ Tet). Bars are mean \pm SEM of 4 independent experiments. Paired *t* test, ns: not significant, **P*<0.05.



Supplementary Figure 3: STAT3 ablation does not reduce cell proliferation or increased apoptosis. (A-B) Representative image (A) and quantification (B) of KI67 staining of kidneys from 18 weeks old *iPkd1^{ΔTub}* and *iPkd1^{ΔTub}; iStat3^{ΔTub}* mice. **(C-D)** Representative image (C) and quantification (D) of TUNEL staining of kidneys from 18 weeks old *iPkd1^{ΔTub}* and *iPkd1^{ΔTub}; iStat3^{ΔTub}* mice. Each dot represents one individual mouse. One-way ANOVA followed by Tukey-Kramer test, ns: not significant, * $P < 0.05$, *** $P < 0.001$. Scale Bars 50 μ m.



Supplementary Figure 4: STAT3 disruption has a selective impact on renal inflammation in ADPKD. (A) Representative images of PAS, F4/80, CD3 and LY6B immunostaining of kidney sections from 18 weeks old $iPkd1^{\Delta Tub}$; $iStat3^{\Delta Tub}$. Scale bars: 1mm for upper left panel and 0.1mm for other panels. (B-G) Quantification of *Cd4* (B), *Cd8* (C), *Tnfa* (D), *Il1b* (E), *Cx3cl1* (F), and *Ub2n* (G) mRNA abundance in kidneys from control, $iPkd1^{\Delta Tub}$ and $iPkd1^{\Delta Tub}; iStat3^{\Delta Tub}$ mice at 18 weeks. Each dot represents one individual mouse. One-way ANOVA followed by Tukey-Kramer test, ns: not significant, * $P < 0.05$, ** $P < 0.01$, *** $P < 0.001$.

Supplementary Table 1: primers used for RT-qPCR

	Forward Primer (5' to 3')	Reverse Primer (5' to 3')
Primers for <i>Canis Lupus</i>		
<i>Ccl2</i>	CTGCTGCTATACACTCACCAATA	TCAGCACAGATCTCCTTGTTTAG
<i>Gapdh</i>	CATGTTTGTGATGGGCGTGAACCA	TTTGGCTAGAGGAGCCAAGCAGTT
Primers for <i>Mus Musculus</i>		
<i>Ccl2</i>	AGTAGGCTGGAGAGCTACAA	GTATGTCTGGACCCATTCCTTC
<i>Ccl5</i>	GCCCACGTCAAGGAGTATTT	CTTGAACCCACTTCTTCTCTGG
<i>Ccr2</i>	GCTCTACATTCACTCCTTCCAC	ACCACTGTCTTTGAGGCTTG
<i>Cd3e</i>	AAGCCTGTGACCCGAGGAA	TGCGGATGGGCTCATAGTCT
<i>Cd4</i>	CAGCATGGCAAAGGTGTATT	TGATGATGAGAGGAAACGATCC
<i>Cd8a</i>	CAGAGACCAGAAGATTGTCG	TGATCAAGGACAGCAGAAGG
<i>Cx3cl1</i>	GGAAAGAAACGTGGTCCAGA	GGAAAGAAACGTGGTCCAGA
<i>Cxcl10</i>	GGATGGCTGTCCTAGCTCTG	TGAGCTAGGGAGGACAAGGA
<i>Gapdh</i>	TGCACCACCAACTGCTTAG	TGGATGCAGGGATGATGTT
<i>Haver1</i>	GAGAGTGACAGTGGTCTGTATTG	CCTTGTAGTTGTGGGTCTTCTT
<i>Hprt</i>	GGCCAGACTTTGTTGGATTTG	CGCTCATCTTAGGCTTTGTATTTG
<i>Il1</i>	GAGGACATGAGCACCTTCTTT	GCCTGTAGTGCAGTTGTCTAA
<i>Il6</i>	CTCTGGGAAATCGTGGAAATG	AAGTGCATCATGGTTGTTCAT
<i>Lcn2</i>	GGACCAGGGCTGTCTGCTACT	GGTGGCCACTTGCACATTGT
<i>Ppia</i>	GGCTATAAGGGTTCCTCCTTTC	TTTCTCTCCGTAGATGGACCT
<i>Rpl13</i>	CTCATCCTGTTCCCCAGGAA	GGGTGGCCAGCTTAAGTTCTT
<i>Socs3</i>	CCACCCTCCAGCATCTTTGT	CAGGCAGCTGGGTCACCTTC
<i>Tnfa</i>	ATTCGAGTGACAAGCCTGTAG	TGAAGAGAACCTGGGAGT
<i>Ub2n</i>	CAGAACCAGTTCCTGGCATT	CAGTGCTGGGGACCACTTAT