

Supplementary Figure 1. Removing one copy of *Rb1* gene promotes expression levels of proteins related to autophagy and angiogenesis in the *Vhl*KO retina. A. Representative Western blots of indicated proteins in retinas of indicated genotypes. B. The quantification of total protein level relative to β -actin in A. Error bars represent SD of measurements from three retinas (n=3) and asterisks indicate significant differences between WT and indicated genotypes (*p<0.05, **p<0.01, one-way ANOVA followed by Bonferroni correction).

Supplementary Figure 2. The ganglion cells, bipolar cells and horizontal cells in *Rb1/Vhl* DKO and *Rb1/Rb1/Vhl* TKO retinas. Horizontal P18 retinal section of indicated genotype was stained for nuclear (DAPI, blue), vascular endothelial cells (IB4, green), rod bipolar cells (Pkc α , red), ganglion cells (Pou4f2, red), horizontal cells (Onecut2, red). ONL: outer nuclear layer. INL: inner nuclear layer. IPL: inner plexiform layer. GCL: ganglion cell layer. RAP: Lesions of retinal angiomatous proliferation. Scale bar is 50 μ m.

Supplementary Figure 3. Deleted cell types in *Rb1/Vhl* DKO and *Rb1/Rb1/Vhl* TKO specified normally. P0 Retinas of the indicated genotype were stained for DAPI (blue) and transcription factor determinants/markers (green) of ganglion (Pou4f2), photoreceptor or bipolar (Crx), or cone (Thrb2) precursors. NBL, neuroblast layer; GCL, ganglion cell layer. Scale bar 50 μ m.

Supplementary Figure 4. *Vhl*KO suppresses the amacrine cell proliferation but induces constant proliferation of Müller cells in the *Rb1*KO and *Rb1/Rb1*DKO retina. A, C: P8 or P18 horizontal retinal sections of indicated genotypes were stained for nuclear (DAPI, blue), vascular endothelial cells (IB4, green, A), cell proliferation (Mki67, red, A, C; PH3, red, A), amacrine cells (Ap2a, green, C), Müller cells (Sox9, green, C), horizontal cells (Onecut2, red, C). B, D: Quantification of Mki67⁺, PH3⁺ cells (B), dividing amacrine cells, Müller cells and horizontal cells (D). Error bars represent SD of measurements from three retinas (n=3) and asterisks indicate significant differences between WT and indicated genotypes (*p<0.05, **p<0.01, one-way ANOVA followed by Bonferroni correction). ONL: outer nuclear layer; INL: inner nuclear layer; GCL: ganglion cell layer. RAP: Lesions of retinal angiomatous proliferation. Scale bar is 50 μ m.

Supplementary Figure 5. CoCl2 induces Hif-1 α accumulation in mouse retina. A. Representative Western blot of indicated proteins in retinas of indicated treatments. B. The quantification of total protein level relative to β -actin in A. Error bars represent SD of measurements from three retinas (n=3) and asterisks indicate significant differences between control and CoCl2 treatment group (*p<0.05, **p<0.01, one-way ANOVA followed by Bonferroni correction).

Supplementary Figure 6. *Ccnd1* null does not affect retinal cell differentiation and lamination.

Horizontal P18 *Ccnd1*^{-/-} retinal sections were stained for nuclear (DAPI, blue), rod (Rho, green), cone (Arr3, red), rod bipolar cells (Pkcα, green), amacrine (Pax6, green), horizontal cells (Calb1, green), Müller cells (Glul, red) and ganglion cells (Pou4f2, red). ONL: outer nuclear layer. INL: inner nuclear layer. IPL: inner plexiform layer. GCL: ganglion cell layer. Scale bar is 50μm.

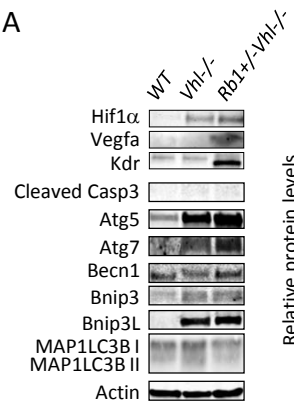
Supplementary Figure 7. *Rb1* knockout in the retina does not affect the body weight of *Ccnd1*^{-/-} mice.

The body weight of the indicated genotypes was measured by an electric balance from P0-P28. Error bars represent SD of measurements from nine animals from three litters (n=9) and asterisks indicate significant differences between *Ccnd1*^{-/-} mice and *WT* or *α-Cre; Rb1*^{fl/fl}; *Ccnd1*^{-/-} mice (**p<0.01, one-way ANOVA followed by Bonferroni correction).

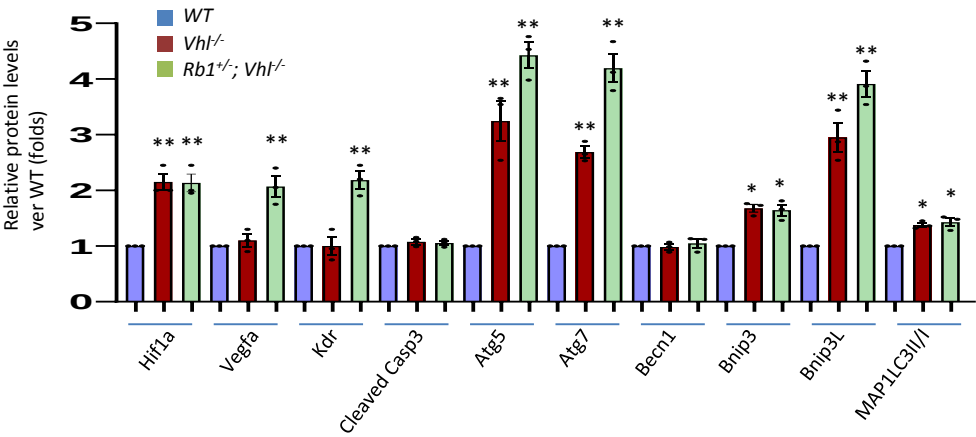
Supplementary Figure 8. Ectopic dividing cells at early time point of *Rb1/Rbl1/Vhl* TKO retinal cells gradually exit cell cycle when they fully developed later. P18 or P138 horizontal retinal sections of indicated genotypes were stained for nuclear (DAPI, blue), vascular endothelial cells (IB4, green), cell proliferation (Mki67, red). INL: inner nuclear layer; GCL: ganglion cell layer. RAP: Lesions of retinal angiomatous proliferation. Scale bar is 50μm.

Supplementary video 1. The new blood vessels linked to hyaloid vessels and DVP of Rb1/Vhl DKO retina. 40 z-stack images were taken using the Nikon C1si confocal microscope, and the Image J software was used to convert these z-stack images into a 3D video.

A

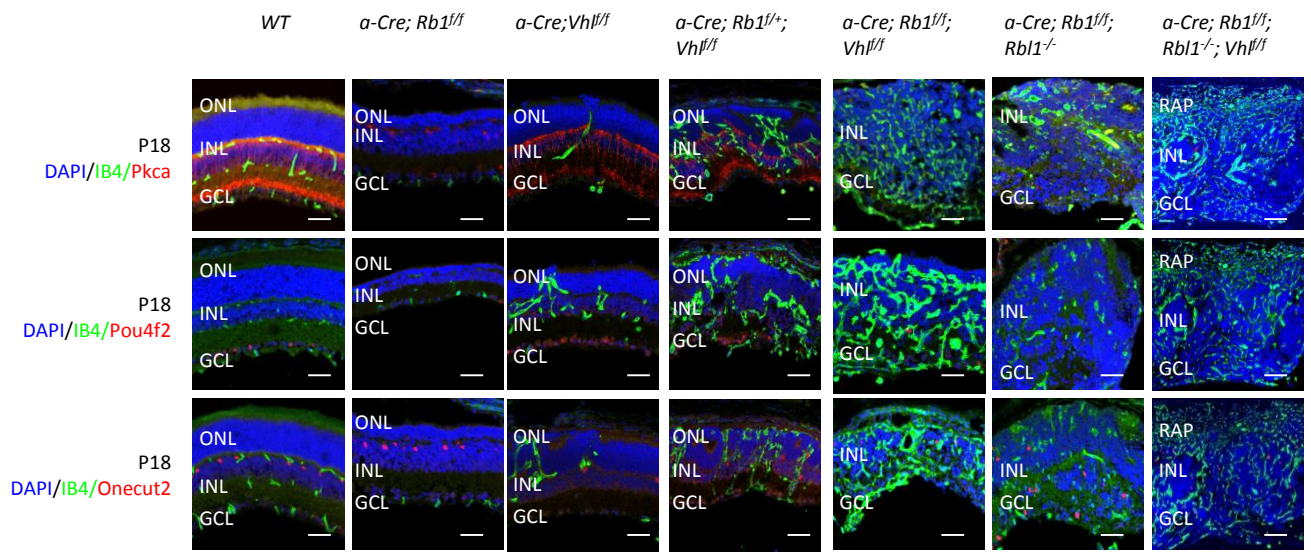


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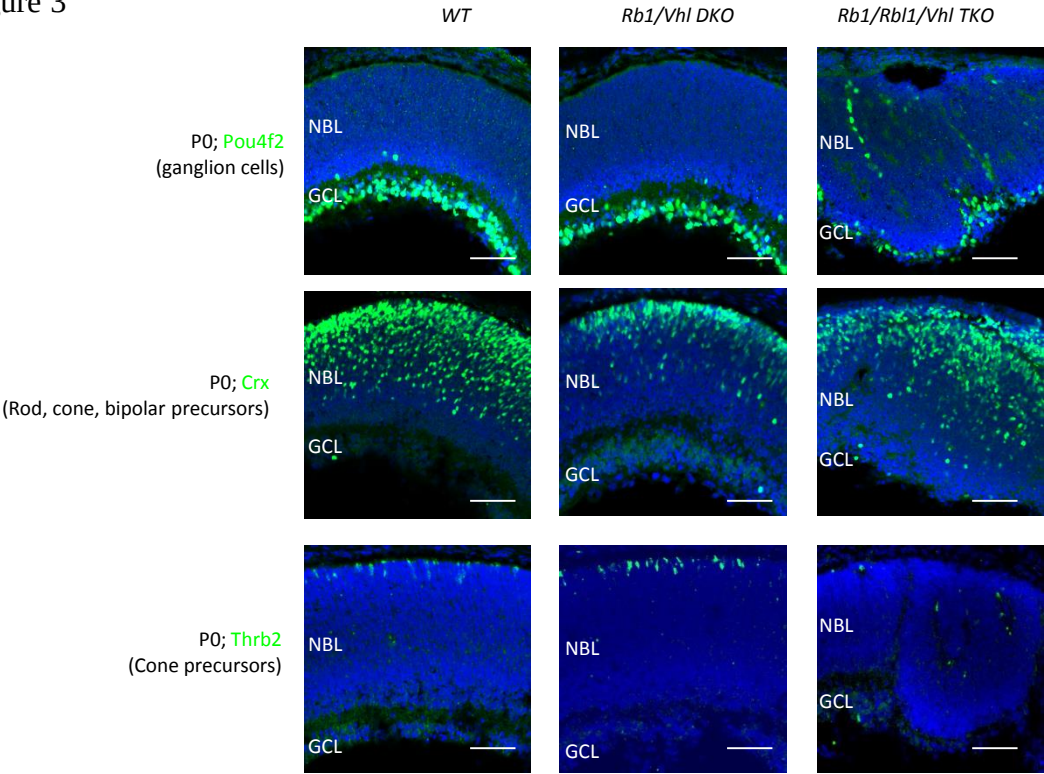
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Supplementary Figure 2



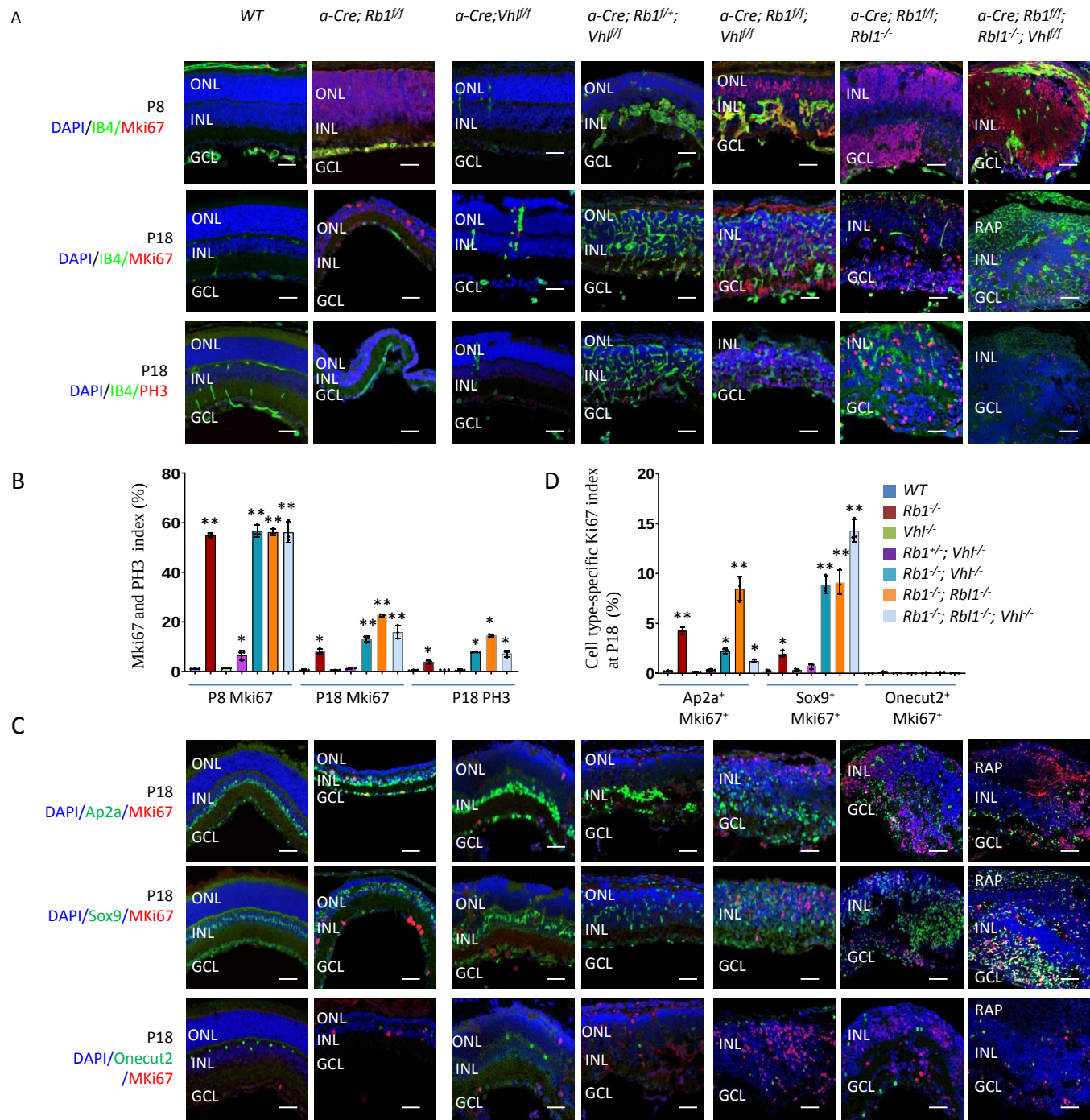
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Supplementary Figure 3

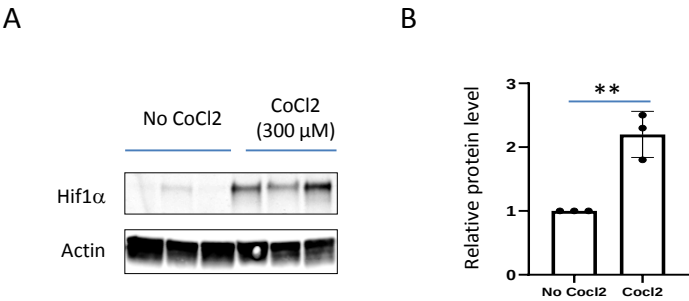


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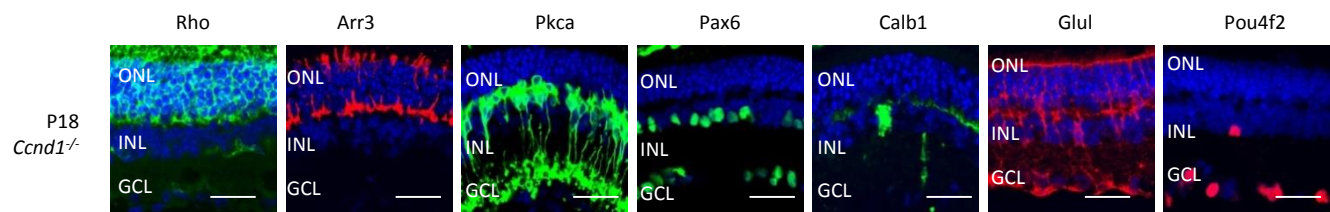
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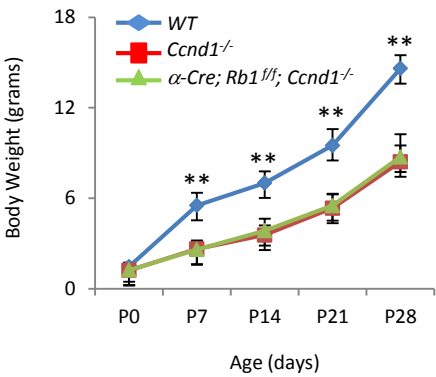


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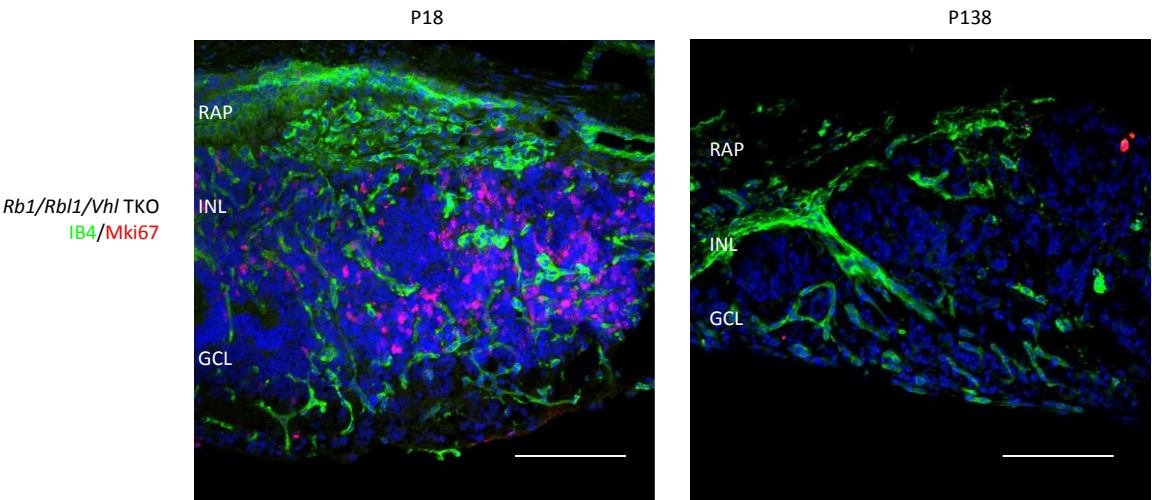


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Supplementary Figure 7



Supplementary Figure 7. *Rb1* knockout in the retina does not affect the body weight of *Ccnd1*^{-/-} mice. The body weight of the indicated genotypes were measured by an electric balance from P0-P28. Error bars represent SD of measurements from nine animals from three litters (n=9) and asterisks indicate significant differences between *Ccnd1*^{-/-} mice and WT or *α-Cre; Rb1^{f/f}; Ccnd1*^{-/-} mice (**p<0.01, one-way ANOVA followed by Bonferroni correction).



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Supplementary Table 2: RT-PCR primer sequences

	Genes	Forward primer (5' to 3')	Reverse primer (5' to 3')
1	<i>Actb</i>	ACCACCACAGCTGAGAGGGA	GCCATCTCCTGCTCGAAGTC
2	<i>Angpt2</i>	CTTAAGCCTGCACCGCTAAC	CTGAACTCCCACGGAACATT
3	<i>Bax</i>	AGGCCTCCTCTCCTACTTCG	CTCAGCCCATCTTCTTCCAG
4	<i>Bbc3</i>	GGGGGTCTGTGAAGAGCATA	CTGGGCACTGGGTAAAGAAG
5	<i>Becn-1</i>	ACCATGGCACTTCTCCTGTC	CAGGCTTCAAGGCTCTCCTA
6	<i>Bnip3</i>	AGGCGTCTGACAACTTCCAC	CCAAGGACCATGCTAGCTCT
7	<i>Ccne1</i>	CTCGGGTGTTGTAGGTGCT	CTGTTGGCTGACAGTGGAGA
8	<i>Cdk2</i>	TCCTCTGAGAGCAGTGATGCA	TTCCCCCAATGACCTAACCAG
9	<i>Cdk2a</i>	CGCAGGTTCTTGGTCACTGT	TGTTACAGAAAGCCAGAGCG
10	<i>E2f1</i>	CTGCAGCAACTGCAGGAGAG	CTCCGAAAGCAGTTGCAGC
11	<i>Egln3</i>	TTCCCTTTAACGGTTCATCG	TGCAGTCAAGGATCAAGACG
12	<i>Epo</i>	CTCCACTCCGAACACTCACA	CCTCTCCCGTGACAGCTTC
13	<i>Fzd4</i>	GCCAATGTGCACAGAGAAGA	GGCAAACCCAAATTCTCTCA
14	<i>Id2</i>	AGGTGGAGCGTGAATACCAG	CAGCATTCAGTAGGCTCGTG
15	<i>Kdr</i>	AGTGGCTCTGTCCTCCAAGA	TCTCACCCTCCTCAACACA
16	<i>Lgals3</i>	TCGTGACTGCTAGGCAAGTG	CAAGTGAGCAGGCACCTGTA
17	<i>Myc</i>	CAGATCAGCAACAACCGCAA	GACGTTGTGTGTCCGCCTCT
18	<i>Mycn</i>	ACATCCTGGAGCGTCAACG	TCTTCACCAGCTCAGGCACAT
19	<i>Rb1</i>	ACAACCCAGCAGTGC GTTATC	ACCAGGTCATCTTCCATCTGT
20	<i>Tek</i>	GAGGCCGAACATTCCAAGTA	ATTGTCACATGGCCAAACAA
21	<i>Tyms</i>	CGTGCAGGATAGGGTGAAGT	TGGAGCCTTCCTCCCTAGAT
22	<i>Vegfa</i>	TCTTCCTGCCCACCATCTAC	TGGTAACCCATGACCAGGAT
23	<i>Vhl</i>	CAGGAGACTGGACATCGTCA	TCCTCTCCAGGTGCTGACT

Supplementary Table 3: ChIP primer sequences

Promoter name	Forward primer (5' to 3')	Reverse primer (5' to 3')
<i>Bnip3</i> promote	AGTCCCAAGGCTCAGACCTC	GTGCCTGAGGCTGGAGTAAC
<i>Epo</i> promoter	GGGGTGTGGCTCAGAAGTAG	TGCAGTGCTGAGACTGGAAC
<i>Kdr</i> promoter	GGCCAAAGCACCATAAAACA	CAGTTGGGAGCATGAAGACA
<i>Tek</i> promoter	GGCGATCTGGGTGTAAGAAA	GGAAGGCAATCAATTTGAGG
<i>Vegfa</i> promoter	ATTCCTGGGAAAGGGAATTG	AACTGGGCTCAGGAACACAC