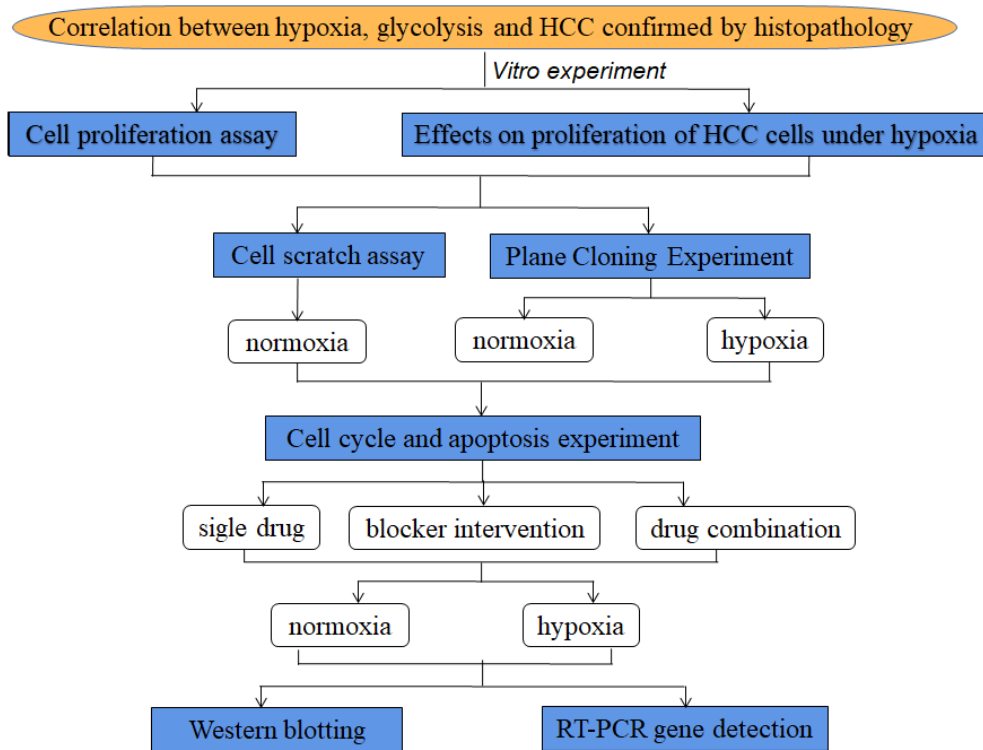
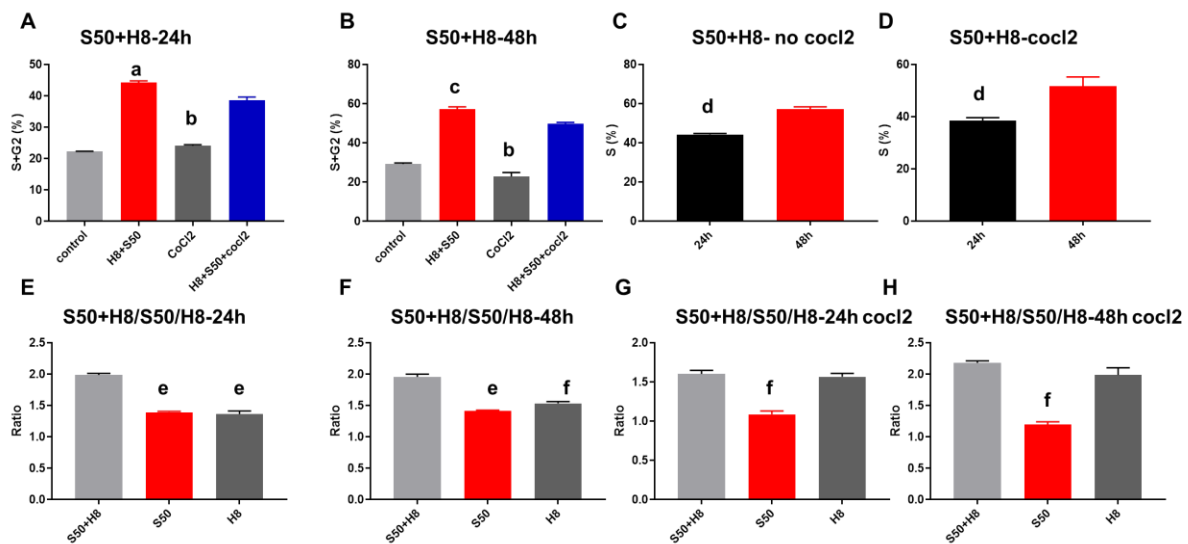


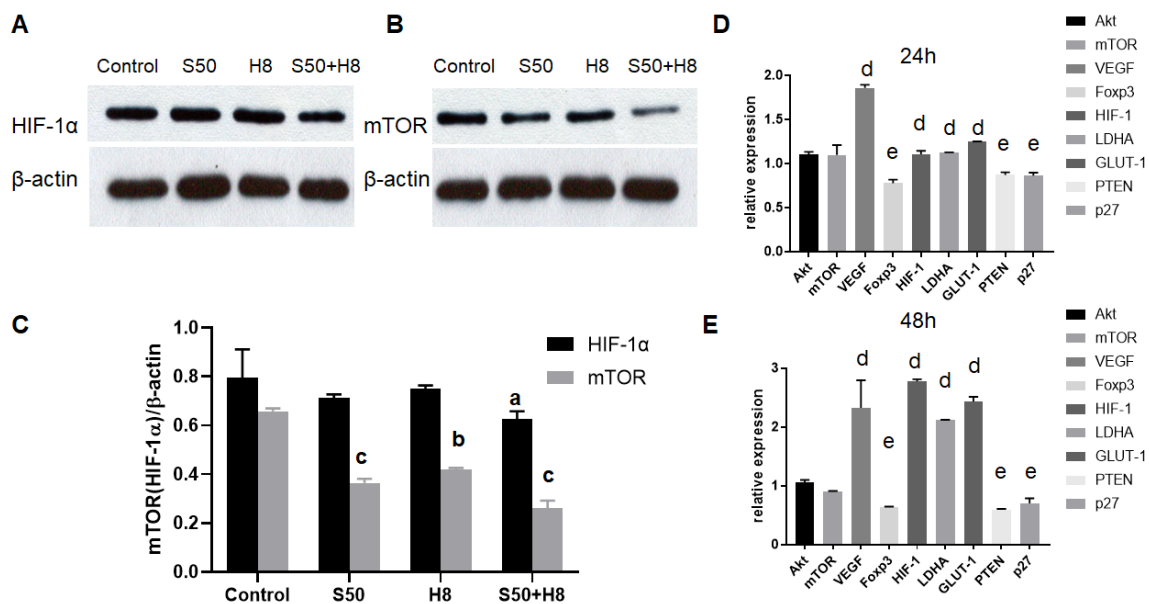
Supplementary material



Supplementary Figure 1 The experimental flow chart. HCC: hepatocellular carcinoma. RT-PCR: Reverse transcription-polymerase chain reaction.

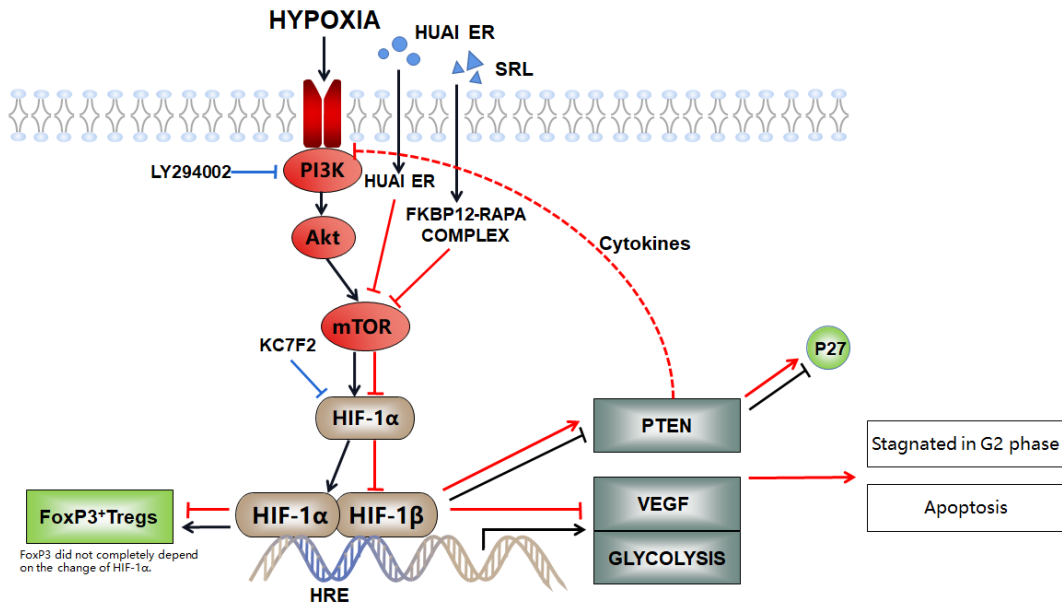


Supplementary Figure 2 Effect of Sirolimus combined with Huai Er on HepG2 cell cycle with normal oxygen and hypoxia. A: Effects of Sirolimus (SRL) combined with Huai Er on HepG2 cell cycle under oxygen and hypoxia at 24h analyzed by two-way ANOVA; B: Effects of SRL combined with Huai Er on HepG2 cell cycle under oxygen and hypoxia at 48h analyzed by two-way ANOVA; C: SRL combined with Huai Er on HepG2 cell cycle under oxygen analyzed by *t* student test; D: SRL combined with Huai Er on HepG2 cell cycle under hypoxia analyzed by *t* student test; E-H: Cell cycle ratio of drug treatment/control analyzed one-way ANOVA. ^a*P* < 0.01 vs H8+S50+cocl2; ^b*P* < 0.001 vs H8+S50+cocl2; ^c*P* < 0.05 vs H8+S50+cocl2; ^d*P* < 0.01 vs 48h; ^e*P* < 0.001 vs S50+H8; ^f*P* < 0.01 vs S50+H8. H8: 8 mg/mL for Huai Er; S50: 50 nM for SRL.



Supplementary Figure 3 Effect of Sirolimus and Huai Er on the expression of protein and target gene mRNA expression under hypoxia. A: The protein expression of HIF-1α with Sirolimus (SRL), Huai Er and combined treatment; B: The protein expression of mTOR with SRL, Huai Er and combined treatment; C: The relative value of HIF-1α and mTOR analyzed by one-way ANOVA; D: The mRNA level of mTOR signal way and glycolytic pathway under hypoxia at 24h analyzed by one-way ANOVA; E: The mRNA level of mTOR signal way and glycolytic pathway under hypoxia at 48h analyzed by

one-way ANOVA. ^a*P* < 0.01 *vs* control; ^b*P* < 0.05 *vs* control; ^c*P* < 0.001 *vs* control; d, down-regulated, ^d*P* < 0.05 *vs* control; e, up-regulated, ^e*P* < 0.001 *vs* control. mTOR: Mammalian target of rapamycin; VEGF: Vascular endothelial growth factor; FoxP3: Forkhead Box P3; HIF-1 α : Hypoxia inducible factor-1 α ; LDHA: Lactate dehydrogenase A; GLUT-1: Glucose transporter 1; PTEN: Phosphatase and tensin homolog deleted on chromosome ten; H8: 8 mg/mL for Huai Er; S50: 50 nM for SRL.



Supplementary Figure 4 Signaling pathways involved in this research. SLR: Sirolimus; mTOR: Mammalian target of rapamycin; VEGF: Vascular endothelial growth factor; FoxP3: Forkhead Box P3; Tregs: Regulatory T cells; HIF-1 α : Hypoxia inducible factor-1 α ; HIF-1 β : Hypoxia inducible factor-1 β ; LDHA: Lactate dehydrogenase A; PTEN: Phosphatase and tensin homolog deleted on chromosome ten; HRE: Hypoxia responsive element.

Supplementary Table 1 The name of the primer and the corresponding sequence information

GAPDH	TCGGAGTCAACGGATTTGGT
	TTCCCGTTCTCAGCCTTGAC
mTOR	CTTAGAGGACAGCGGGGAAG
	GACTCATCTCTCGGAGTTCCAT
Akt	TTGAGGGTTGTCTCCGTGCT
	GGTCTCCAGGCAGGACTGAT
HIF-1 α	AGGTGGATATGTCTGGGTTG
	AAGGACACATTCTGTTTGTTG
FoxP3	TACTTCAAGTTCCACAACATGCGACC
	CGCACAAAGCACTTGTGCAGACTCAG
GLUT1	CGGGCCAAGAGTGTGCTAAA
	TGACGATACCGGAGCCAATG
LDHA	GGTCCTTGGGGAACATGGAG
	TAGCCCAGGATGTGTAGCCT
P27	CCCTGAACGGAGCTGAAGTC
	CTAGTCTCCAGGGAGGTGCT
PTEN	CTGCAGAAAGACTTGAAGGCG
	TGCTTTGAATCCAAAAACCTTACT
VEGF	CTTGCCTTGCTGCTCTACCT
	ACGCGAGTCTGTGTTTTTGC

Objective gene mRNA= $2^{-\Delta\Delta CT}$, $\Delta\Delta$ = (The CT value of the target gene of the sample to be tested - the CT value of internal reference gene in the sample to be tested) - (The CT value of the target gene of the control sample - the CT value of internal reference gene in control sample).