

A

PINCR_FL-FP: Starts at position 1.

PINCR_FL-FP: ATTCAATCTGTGAGGTGGATGCGTGTTGGAAGGA

PINCR_FL-RP: Starts at position 1864

PINCR_FL-RP: AAGGGTATAACAATCACCCGGAGTCAGACAATTG

35 cycles

98 C 30s
98 C 10 s
60 C 10s
72 C 1.5 min
72 C 5 min
4 C forever

Primer: 10 uM mix of FP and RP = 2.5 ul

Template: For a 25 ul reaction with 2x Phusion NEB

pCB6-PINCR = 10 ng

cDNA = 2 ul out of a 10 ul RT reaction (iScript RT mix)

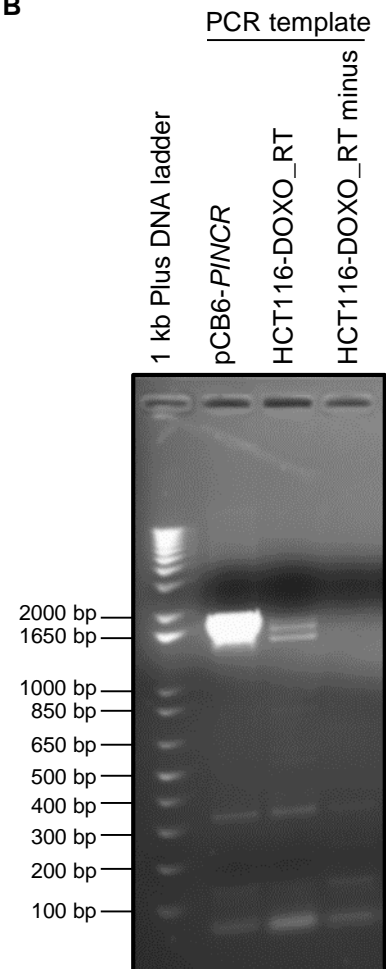
B

Figure 1 - figure supplement 4. RT-PCR for *PINCR* was performed using cDNA prepared from HCT116 DOXO-treated total RNA or pCB6-*PINCR*. The forward primer starts at the 5' end of the annotated *PINCR* RNA and the reverse primer is located near the 3' end of *PINCR* RNA. Location of the primers and conditions for PCR are shown in (A). Picture of the agarose gel after electrophoresis is shown in (B). pCB6-*PINCR* was used as positive control. RT minus refers to a control reaction in which the total RNA was used as template for PCR.