

SUPPLEMENTARY DATA

The ubiquitin specific protease 19 (USP19) regulates the hypoxia inducible factor 1 α (HIF-1 α) during hypoxia

Mikael Altun, Bin Zhao, Kelly Velasco, Haiyin Liu, Gerco Hassink, Julia Paschke, Teresa Pereira and Kristina Lindsten

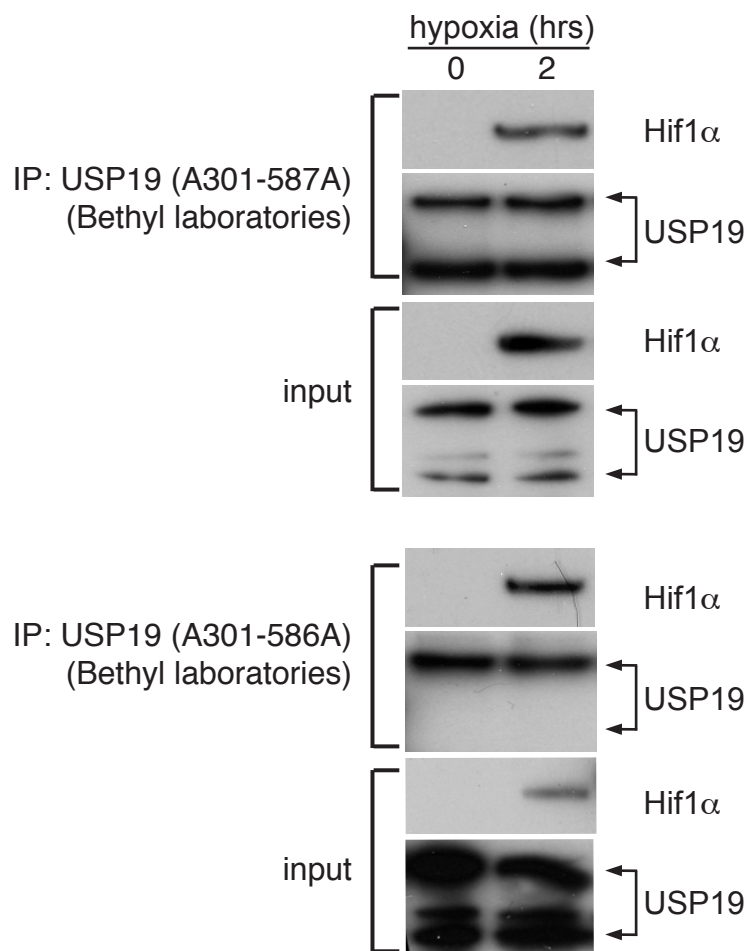


FIGURE S1. Interaction between endogenous USP19 and HIF-1 α .

A) HeLa cells were exposed to hypoxia for 2 hrs to accumulate detectable levels of endogenous HIF-1 α . Subsequent immunoprecipitations with two different antibodies against USP19, USP19(A301-586A) and USP19(A301-587A) (Bethyl laboratories, Montgomery, TX, USA), reveal an interaction with HIF-1 α . All USP19 western blots were probed with the USP19(A301-587A) antibody since this works for western blot. The different forms of USP19 detected after immunoprecipitation is likely due to limited binding to the target epitope under the native conditions of the immunoprecipitations.

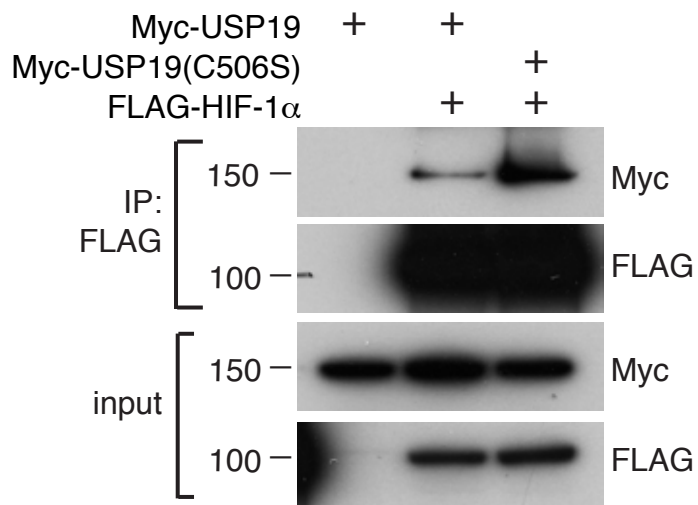


FIGURE S2. *USP19 catalytic activity is not required for interaction with HIF-1 α .* Co-immunoprecipitation from lysates of 293T cells overexpressing wild-type and catalytically inactive USP19 together with FLAG-HIF-1 α . Immunoprecipitation was performed using FLAG(M2) affinity gel and western blots were probed as indicated.

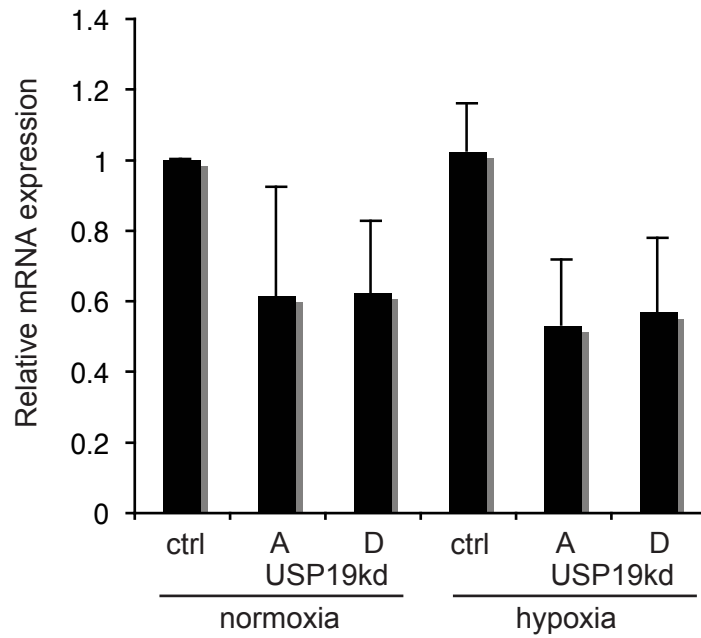


FIGURE S3. *USP19 knock-down assayed by qPCR.*

Same samples as in Figure 5E assayed for level of USP19 mRNA knock-down. Values show an approximate 50% reduction of USP19 mRNA achieved by transfection in HeLa cells of plasmids expressing two different shRNA against USP19, pRETRO-SUPER-USP19A/D.