Supplementary Figures

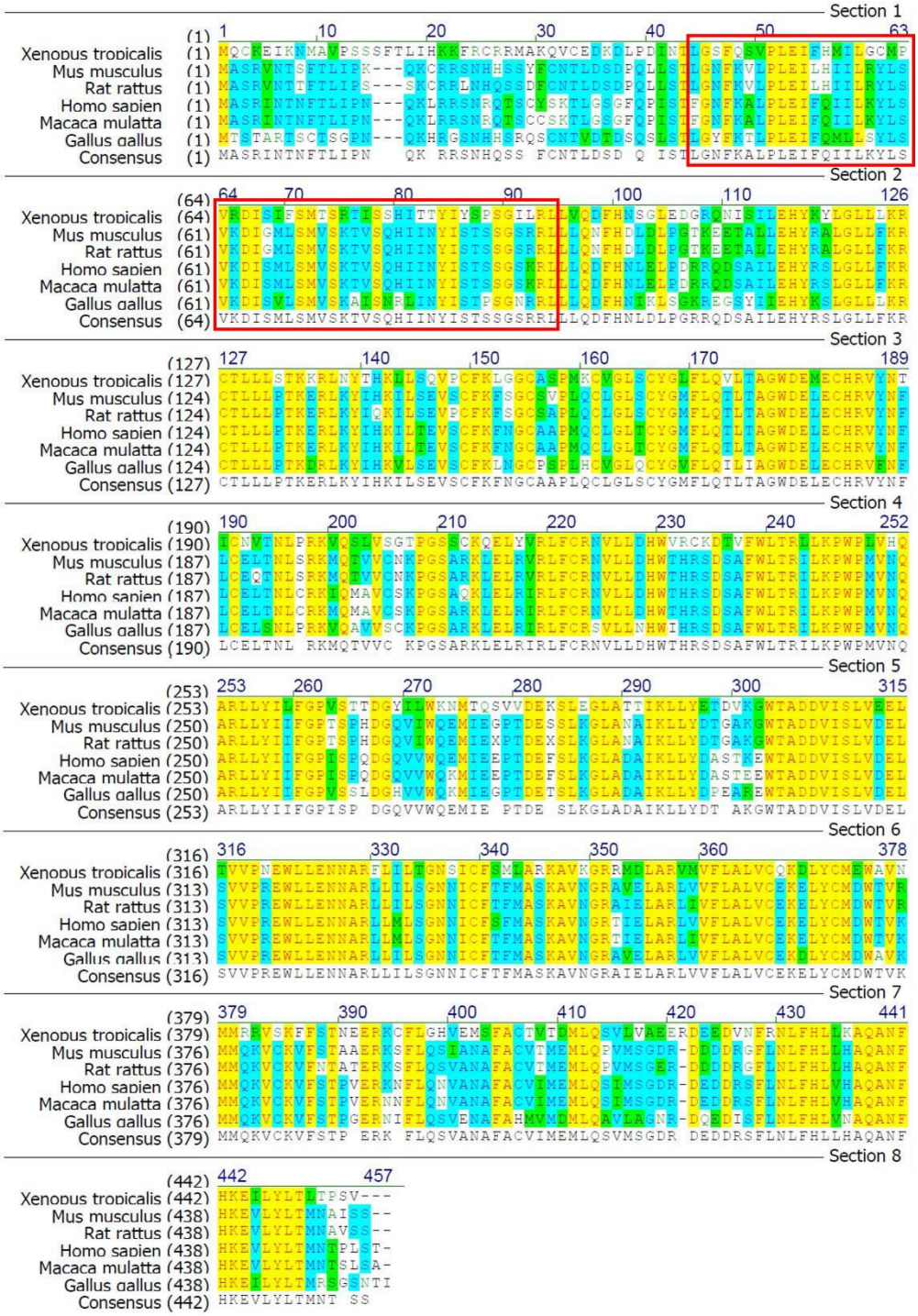

Fig. S1. *Fbxo47* is a highly conserved gene in multiple species. Sequence alignment of FBXO47 proteins from amphibians, birds, rodents, primates and humans. The sequence in the red box is the F-box domain.

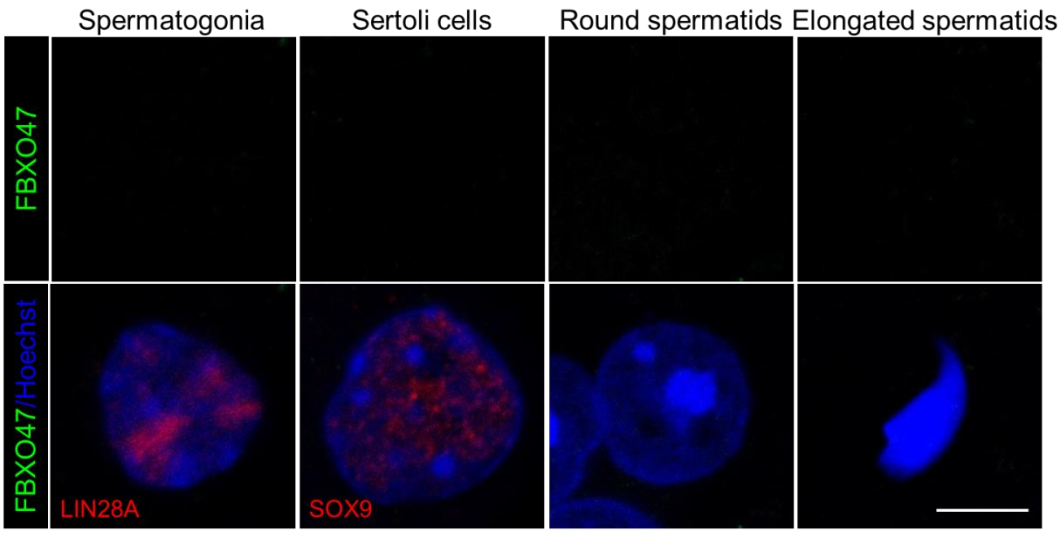

Fig. S2. Distribution pattern of FBXO47 in the testis suspensions. Testichular cells were stained with the indicated antibodies. No specific FBXO47 signal was detected in spermatogonia, Sertoli cells, round spermatids and elongated spermatids. The cells were prepared with a mild hypotonic treatment and then fixed in Triton X-100. LIN28A is a spermatogonia marker, and SOX9 marks Sertoli cells. Bars, 5 μm.

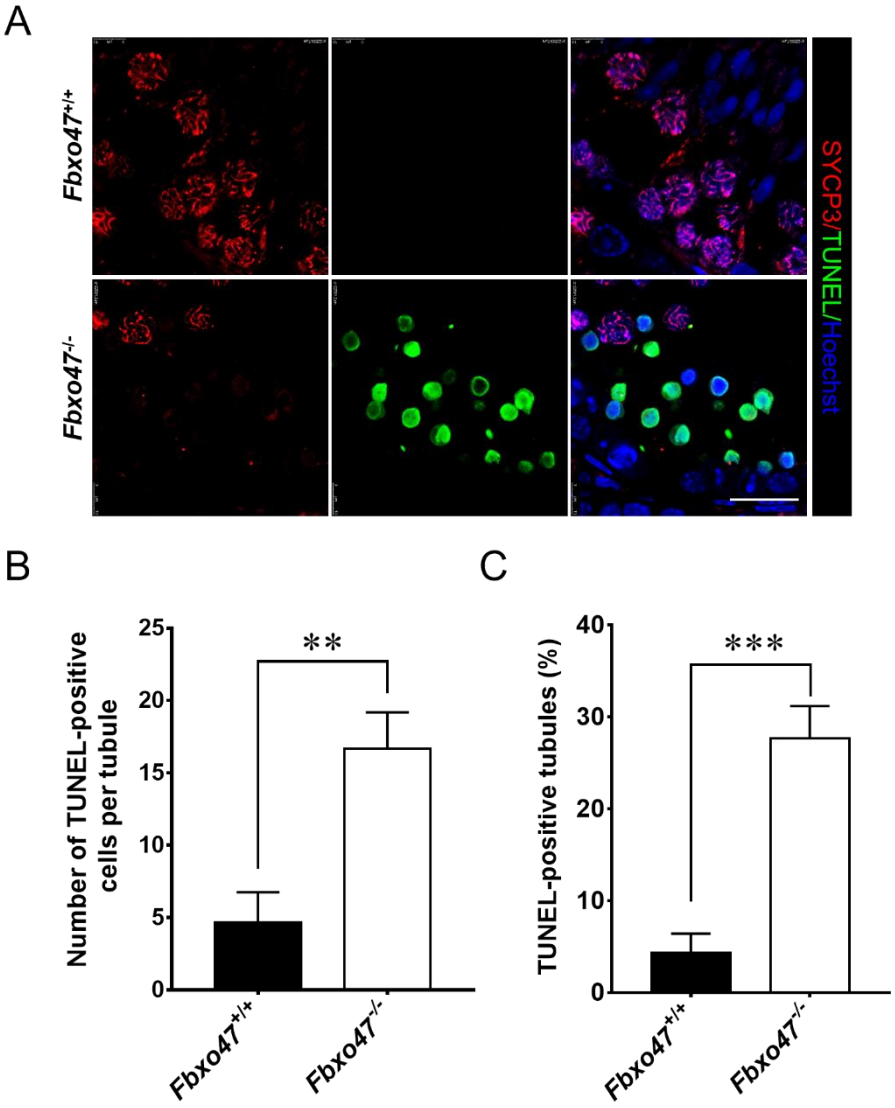

Fig. S3. Extensive apoptotic events account for the absence of spermatids and spermatozoa in *Fbxo47*-null testis.

(**A**) Paraffin-embedded testis sections from 2 months *Fbxo47*$^{+/+}$ and *Fbxo47*$^{-/-}$ mice stained with TUNEL and the indicated antibody. Scale bar, 10 μm.

(**B**) and (**C**) Graphs show the number of TUNEL-positive cells per tubule and the percentage of TUNEL-positive tubules in the sections. Error bars, SEM (n = 3); **P ≤ 0.01, ***P ≤ 0.001.

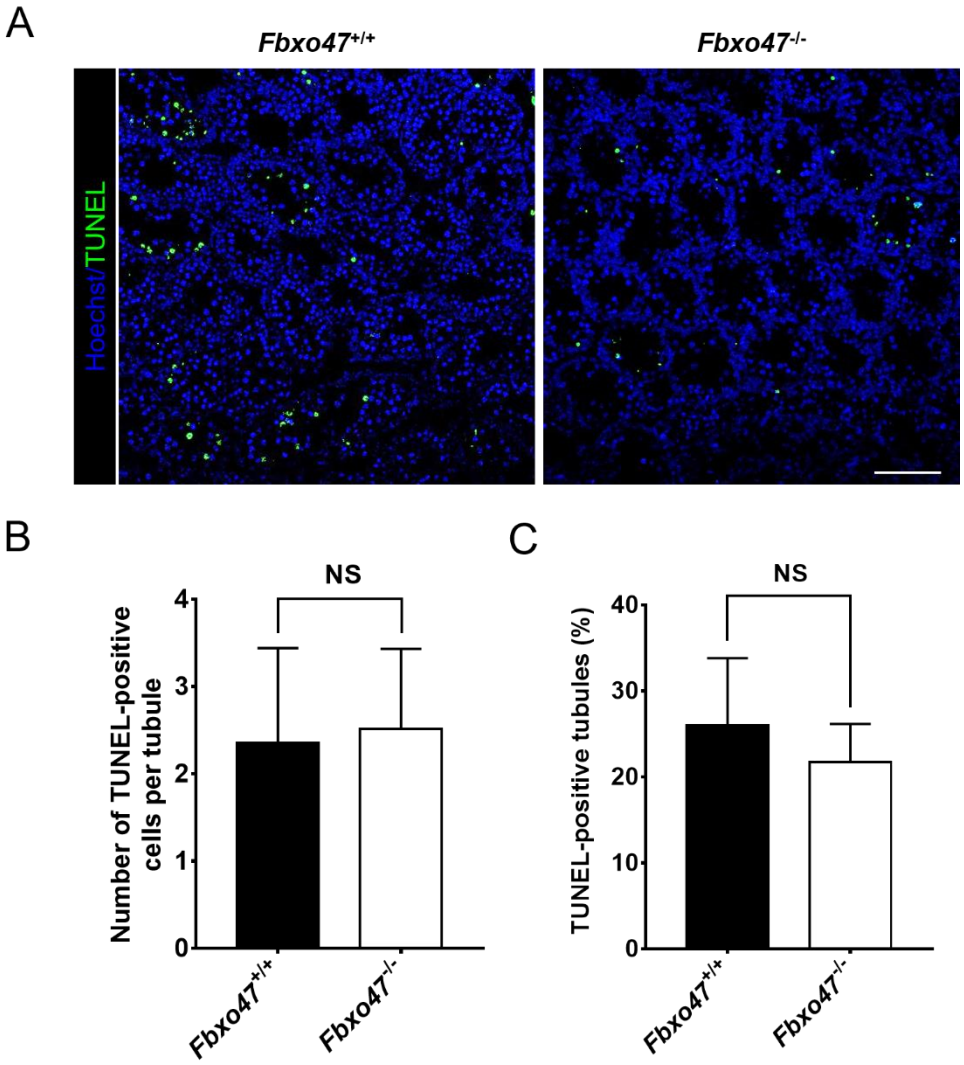

Fig. S4. Similar frequency of apoptotic events in juvenile (P14) *Fbxo47*-null and control testes.

(**A**) Paraffin-embedded sections of P14 *Fbxo47*$^{+/+}$ and *Fbxo47*$^{-/-}$ mice testes stained with TUNEL and counterstained with Hoechst. Scale bar, 100 μm.

(**B**) and (**C**) Graphs show the number of TUNEL-positive cells per tubule and the percentage of TUNEL-positive tubules in the sections. Error bars, SEM (n = 3); NS, no significance.

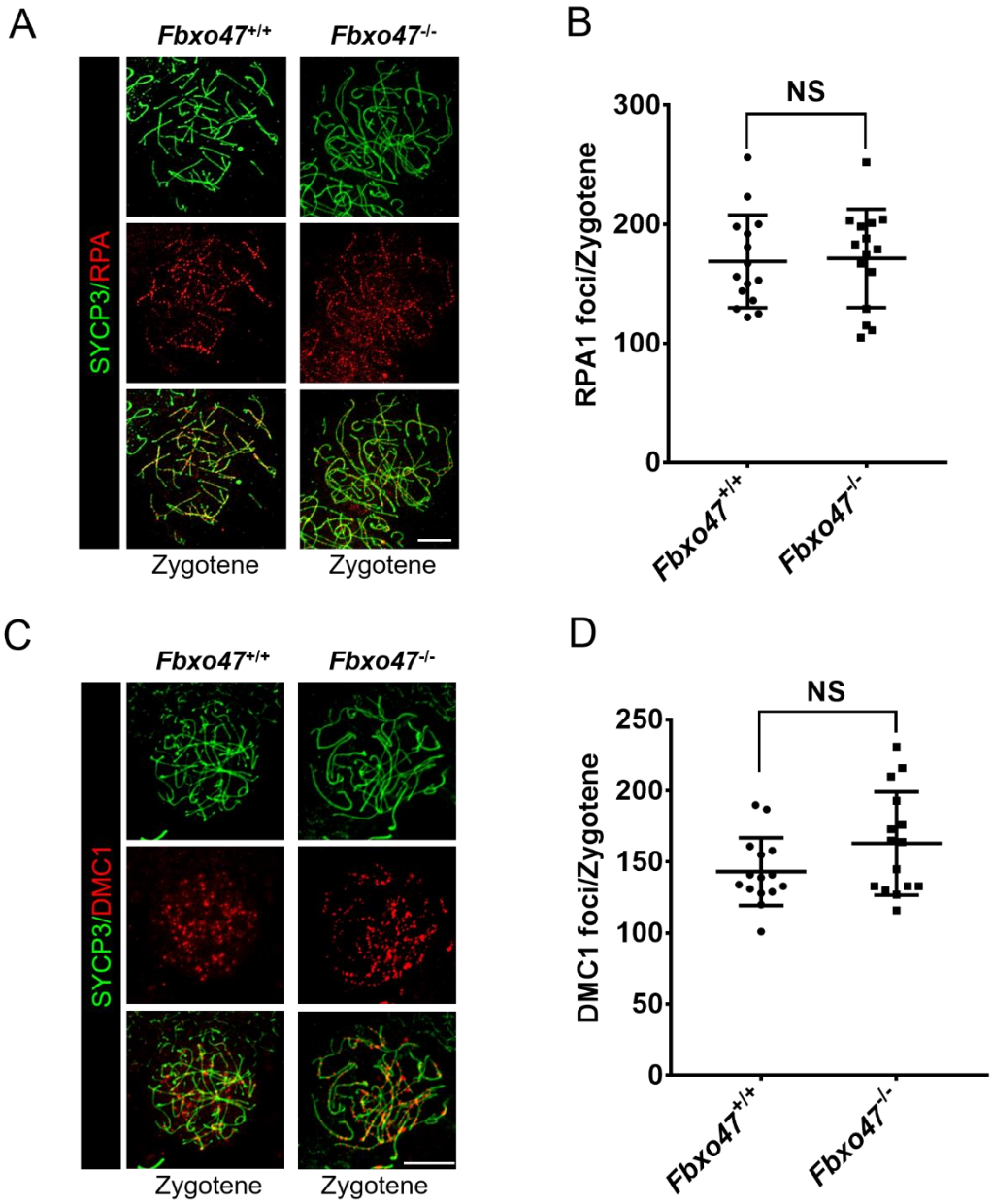

Fig. S5. Meiotic strand invasion is not impacted by the disruption of *Fbxo47*.

(**A**) and (**C**) Spread spermatocytes from *Fbxo47*$^{+/+}$ and *Fbxo47*$^{-/-}$ males were stained with anti-SCP3, anti-RPA1 and anti-DMC1 antibodies. Scale bars, 10 μm.

(**B**) and (**D**) Quantification of RPA1 and DMC1 foci in (**A**) and (**C**) (n >15 cells for each genotype). NS, no significance.

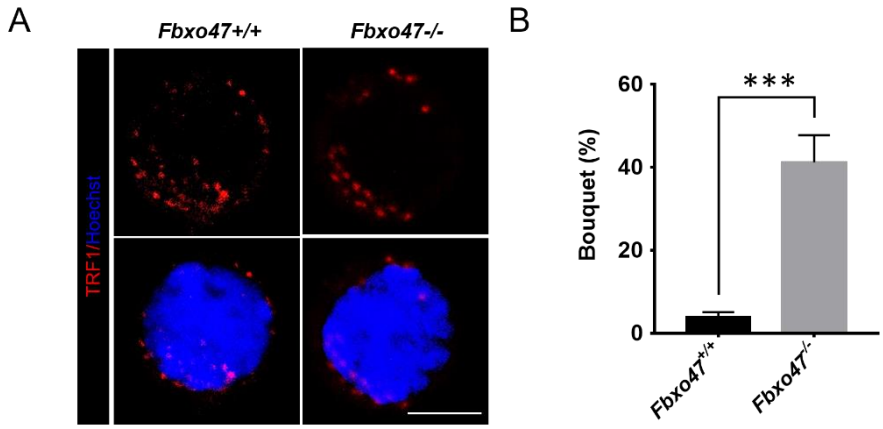

Fig. S6. Meiotic bouquet in *Fbxo47*$^{+/+}$ and *Fbxo47-/-* testes.

(A) Telomere distribution was visualized in testis suspension by immunofluorescence with indicated antibody (orthogonal z-stack projections). Scale bars, 10 μm.

(B) The proportion of cells with clustered telomeres (bouquets) in testis suspension of adult mice. This result suggests an extension of the duration of the transient bouquet telomere clustering during meiosis in *Fbxo47*$^{-/-}$ spermatocytes. Mean values of three independent experiments from three different mice are shown. Error bars, SEM (n = 3); ***$P \leq 0.001$ (Student's t-test).

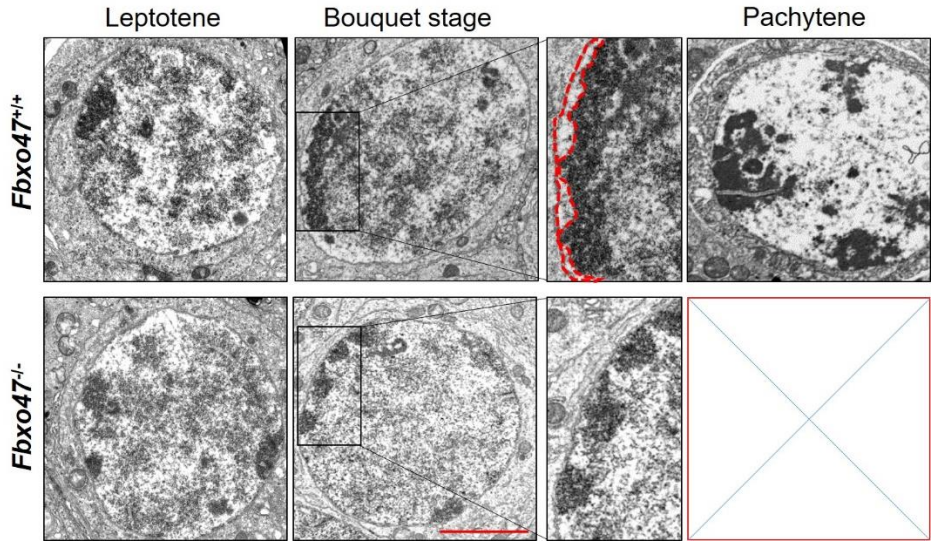

Fig. S7. FIB-SEM images of spermatocytes of the indicated genotypes. The dashed red line marks the jagged interspace. Scale bars, 4 μm.

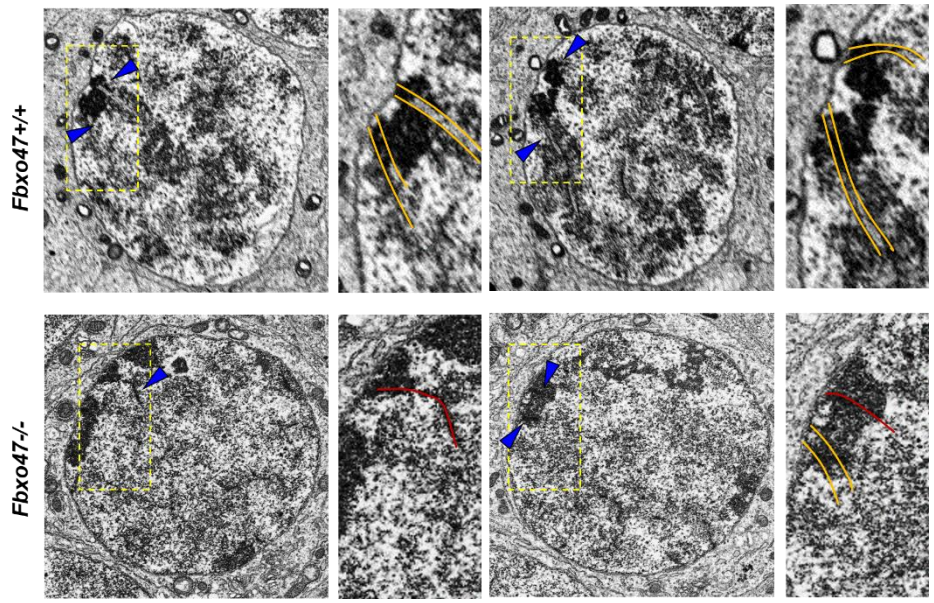

Fig. S8. Continuous electron microscopic images of sections selected from FIB-SEM images. Blue arrowheads indicate the synaptonemal complex. Red lines, unpaired lateral elements; Yellow lines, lateral elements of the synaptonemal complex.

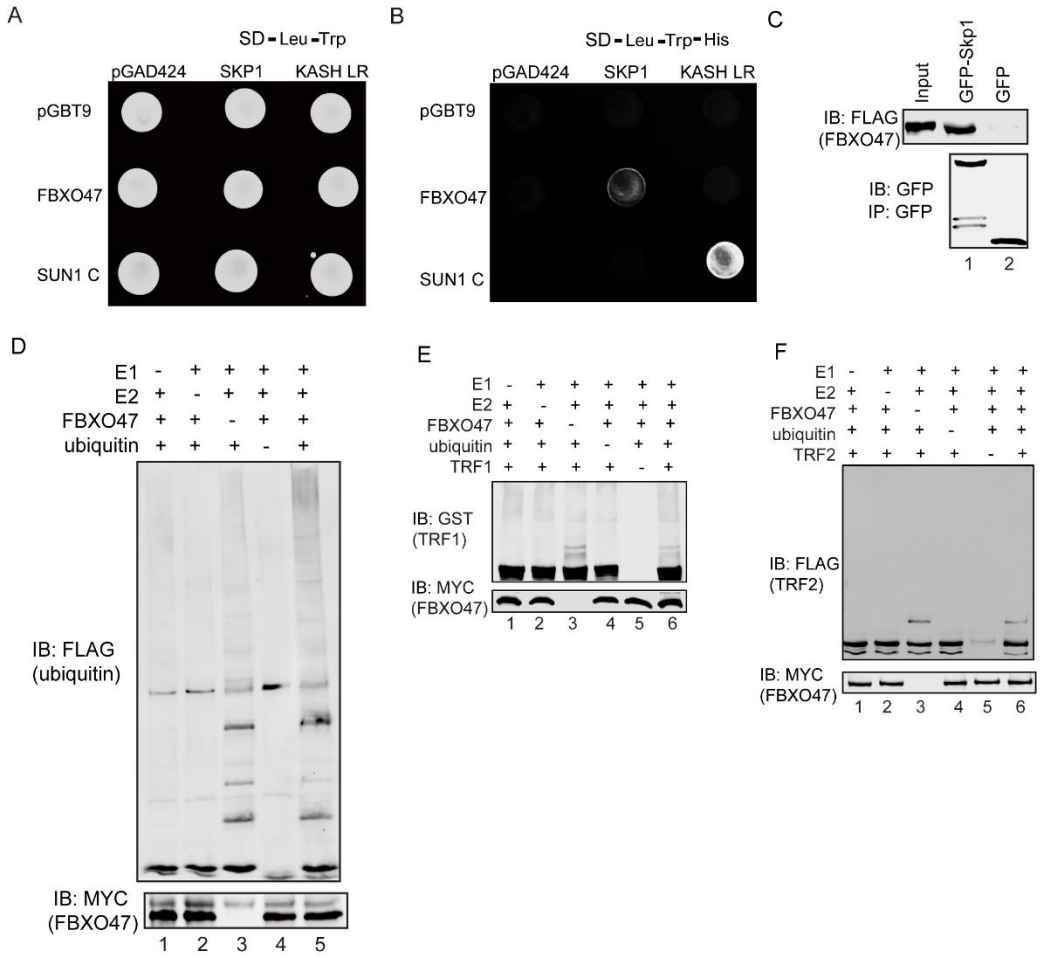

Fig. S9. FBXO47 does not appear to directly promote the ubiquitination of TRF1 and TRF2.

(A) and **(B)** FBXO47 was co-transformed with SKP1 in the Y2H assay, and yeast cells were spotted onto control (SD-Leu-Trp) and selective (SD-Leu-Trp-His) plates.

(**C**) FBXO47 interacts with SKP1. *pRK-Flag-Fbxo47* and *pEGFP-Skp1* were co-transfected into HEK293T cells. After 24 h, the cells were collected for immunoprecipitation (IP) with an anti-GFP antibody and analyzed with FLAG and GFP antibodies.

(**D**) FBXO47 functions as a component of SCF to catalyze ubiquitin chain formation. *pCS2-Myc-Fbxo47* was transfected into HEK293T cells. After 24 h, the cells were

collected for immunoprecipitation (IP) with an anti-MYC antibody. E1, E2, ubiquitin, ATP and Ufd2p were then added to the mixture. The reaction products were detected using anti-FLAG and anti-MYC antibodies**.**

(**E**) and (**F**) FBXO47 does not appear to directly promote the ubiquitination of TRF1 and TRF2. GST-TRF1 (**C**) and FLAG-TRF2 (**D**) were added to the *in vitro* FBXO47 ubiquitination system, which contained E1, E2, ubiquitin, ATP and the MYC-FBXO47 immunoprecipitate. The reaction products were detected using anti-FLAG, anti-GST and anti-MYC antibodies**.**