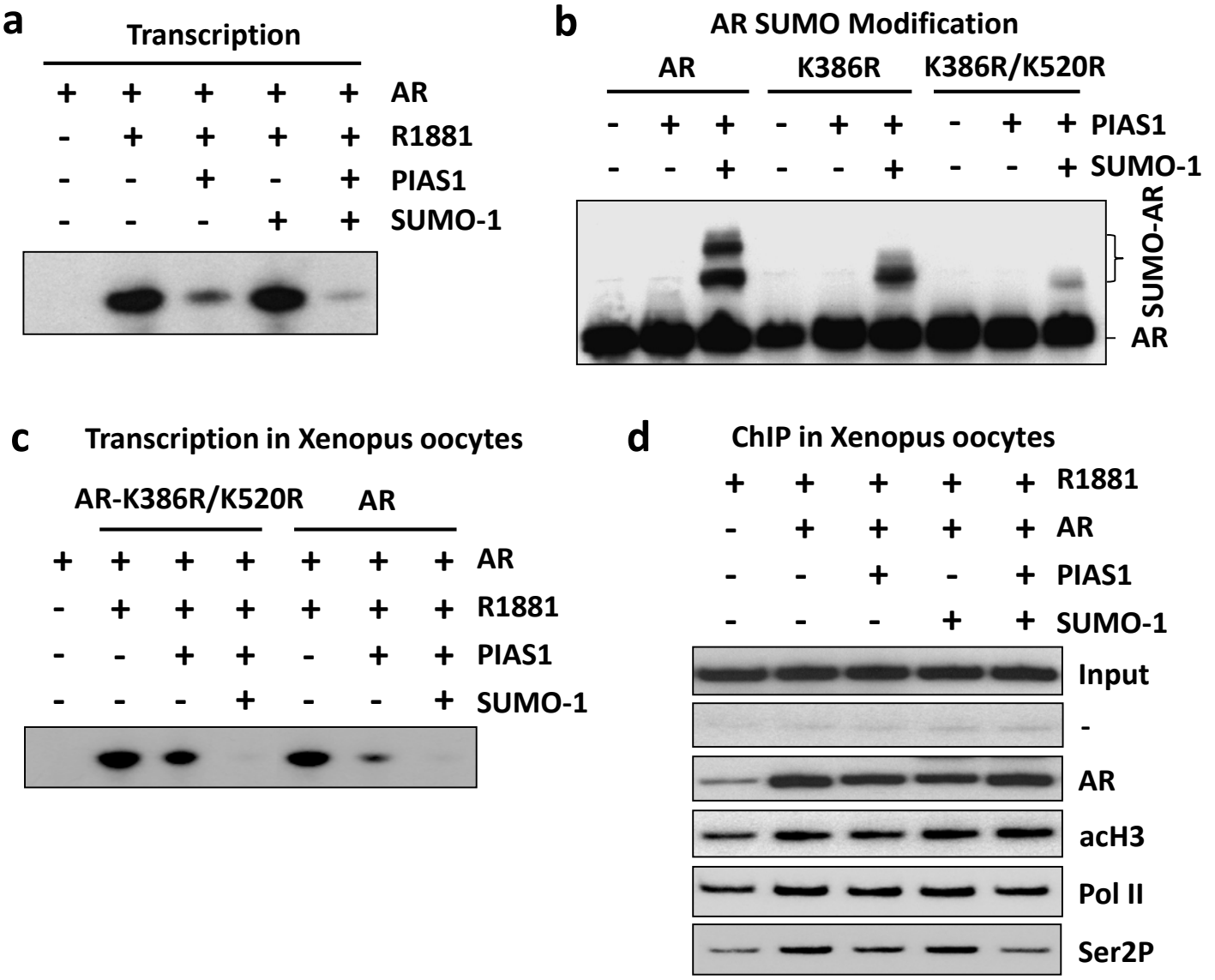

Supplementary Figure 1. SUMO represses transcriptional activation by AR independent of AR sumoylation. (**a**) SUMO repressed AR- and ligand-dependent transcriptional activation of MMTV reporter in Xenopus oocytes. Xenopus oocytes were injected with single-stranded (ssDNA) MMTV-Luc reporter and in vitro-synthesized mRNAs encoding Flag-AR, HA-PIAS1 and 6xHis-SUMO-1 as indicated. The injected ssDNA underwent one round of DNA synthesis and converted to minichromosomes via replication coupled-chromatin assembly. The oocytes were then treated with or without 10 nM R1881, a synthetic AR agonist, overnight. The levels of transcription from MMTV-Luc were determined by primer extension as described [38]. (**b**) Detection of sumoylation on the wild-type and K386R and K386R/K520R mutants in Xenopus oocytes. K386 and K520 are major sumoylation sites on AR as reported [35]. Xenopus oocytes were injected with mRNAs encoding wild-type or mutant AR together with or without PIAS1 or SUMO-1 as indicated. After overnight incubation the oocytes were harvested and WB analysis was performed to check the status of AR sumoylation. (**c**) Co-expression of PIAS1/SUMO-1 repressed transcriptional activation of MMTV-luc reporter by both wild-type and sumoylation-impaired mutant AR. Xenopus oocyte injection and transcriptional analysis were as in (**a**). (**d**) Xenopus oocytes were injected with ssDNA of MMTV-Luc and indicated mRNAs as in (**a**). The oocytes were then processed for ChIP assay analyzing the binding of AR, histone acetylation, recruitment of pol II, and Ser2P of pol II CTD on the MMTV-LTR using antibodies as indicated. The ChIP assay was performed essentially as described [38].