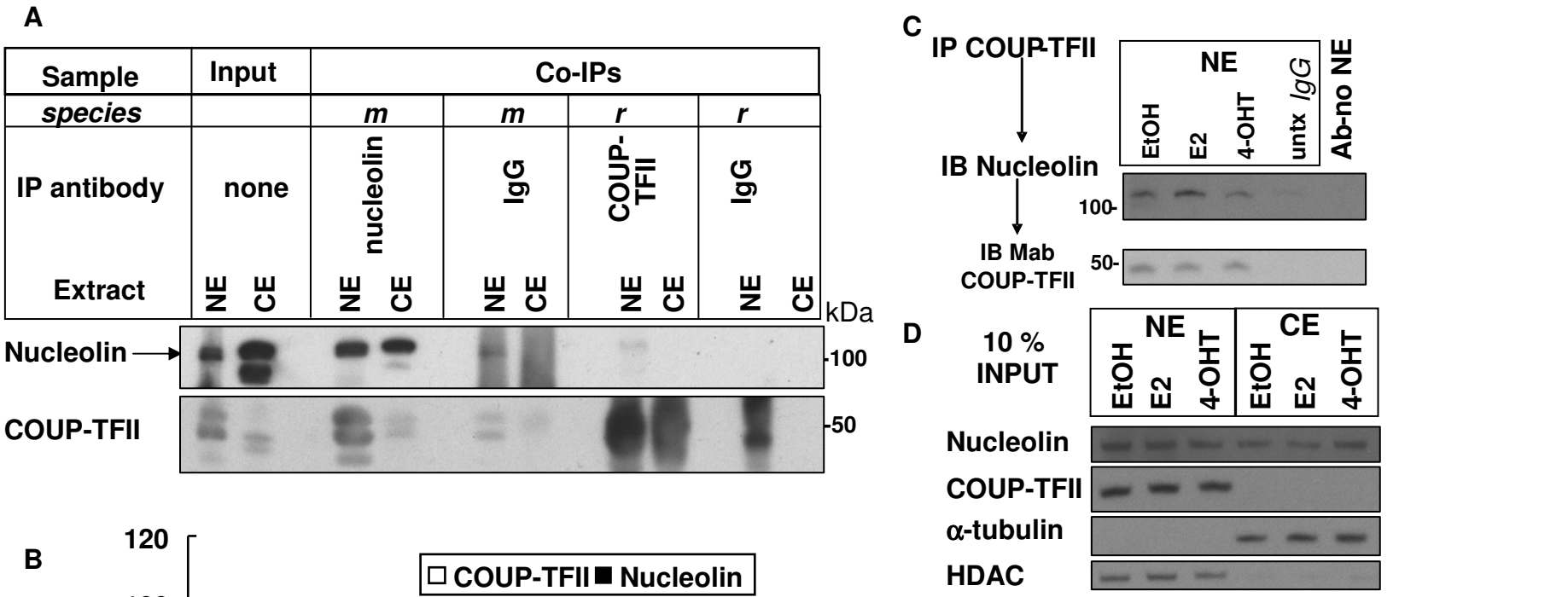

Figure S4: Endogenous nuclear nucleolin-COUP-TFII interaction in MCF-7 cells. A, Equal amounts (100 μg) of protein of CE and NE from MCF-7 cells were immunoprecipitated with nucleolin mAB (lanes 3 and 4), mouse (m) IgG (negative control for mAB, lanes 5 and 6), COUP-TFII antisera (lanes 7 and 8), or rabbit (r) IgG (negative control for IPs using COUP-TFII polyclonal antiserum, lanes 9 and 10), followed by western blot for nucleolin and COUP-TFII. 20% (20 μg) input NE and CE serve as loading controls (lanes 1 and 2). B, The relative amount of nucleolin and COUP-TFII in the nucleolin IP was plotted relative to expression of each protein in the input (set to 100). COUP-TFII in rabbit IgG IPs was not graphed because of the contamination of the heavy IgG chain (lanes 7 and 8, COUP-TFII blot). Western blots demonstrate that: 1) nucleolin interacts with COUP-TFII in the NE of MCF-7 cells (lane 7); 2) nucleolin is not IP'ed with rabbit IgG (lanes 9 and 10); 3) more COUP-TFII interacts with nucleolin in NE IP'ed with nucleolin antibody than with mouse IgG (lane 3 versus lane 5). C, MCF-7 cells were treated with EtOH, 10 nM E2, or 100 nM 4-OHT for 1 h prior to separation of NE and CE. 200 μg of NE or CE were IP'ed with polyclonal COUP-TFII antibody and immunoblotted with a monoclonal antibody (mAB) against nucleolin. The blot was stripped and re-probed with mAB against COUP-TFII. D, 10% input for NE and CE used in IP in part C.