Figure S6
Lack of NPM involvement in transcriptional regulation of another AP-2α target, TGFα

To address the issue of whether NPM is involved in expression of other AP-2α targets that are not responsive to RA signaling, we examined one of the reported target genes of AP-2α, transforming growth factor-α (TGF-α) (Wang et al., 1997). AP-2α has been implicated in positively controlling the transcription of the TGF-α gene. As shown in Fig. S6A, ChIP analysis was performed using control antibodies (IgG, lane 2) or antibodies against AP-2α (lane 3) or NPM (lane 4). Products from final PCR analysis using primers specific to the NPM promoter were resolved by agarose gel. DNA input is shown in lane 1. Consistent with the findings in the above report, ChIP experiment has pinpointed AP-2 in the regulatory region of the gene. However, binding of NPM was not detected. In addition, RNA was isolated from control (Fig. S6B, lane 1) or NPMRNAi (lane 2) HeLa cells. The mRNA level of the TGFα gene (upper panel) was examined by RT-PCR. Expression level of β-actin (lower panel) was used as a loading control. This result indicates that knockdown of NPM expression does not affect the expression of TGF-α mRNA. Taken together, these findings suggest that the NPM-AP-2α association is selective and pathway-specific.