Supplementary Figure 1. Conventional purification of the human PAF complex. (A) The purification scheme for human PAF complex. (B) Silver stain and Western blot of the purified human PAF complex. Fractions were from the 1ml MonoP column. The fraction numbers are shown on top of the panel. The upper panel is the silver stain of the complex. The five subunits of the human PAF complex are marked with black dots and labeled on the right side. The lower panel shows the Western blot with antibodies against three of the human PAF complex subunits.

Supplementary Figure 2. Sequence alignments for human PAF subunits, Rtf1, SKI subunits versus their yeast homologues.

Supplementary Figure 3. hSki8 is a novel higher eukaryote specific subunit of hPAF complex and hRtf1 is not a integral subunit of hPAF complex. (A) Western blots for the same materials shown in Fig. 1A. (B) Silver stain of the affinity purified hRtf1 complex. Fractions were from a 2ml Smart Superdex 200 gel filtration column. The fraction numbers are shown on top of the panel. Flag-hRtf1 is indicated on the side. Asterisks indicate commonly contaminating, non-specific proteins such as Hsp70, tubulin and actin. (C) Silver stain of the hPAF depleted Flag-hSki8, which was loaded onto a 2ml Smart Superose 6 gel filtration column. The fraction numbers are shown on top of the panel.

Supplementary Figure 4. Interaction assay for hPAF complex, hRtf1 and RNA polymerase II. The Co-IP experiments were performed with HeLa nuclear extracts. The 0.15M KCl wash used here is typical for a Co-IP experiment, but less stringent compared to the affinity purification performed in Fig. 1.

Supplementary Figure 5. hSki8 localizes to both cytoplasm and nucleus of HeLa cells, while hPaf1 is predominantly nuclear.

Supplementary Figure 6. RNAi against hCtr9 does not affect MAGE-A1 gene induction.