論文 / 著書情報
Article / Book Information

題目(和文)	RNAポリメラーゼIIの転写伸長段階を制御する新規ヒトタンパク質Rtf1に関する研究
Title(English)	Sutudy on Rtf1, an uncharacterized human protein that controls the elongation phase of RNA polymerase II transcription
著者(和文)	曹青福
Author(English)	Seifuku Sou
出典(和文)	学位:博士（理学）, 学位授与機関:東京工業大学, 報告番号:甲第10020号, 授与年月日:2015年11月30日, 学位の種別:課程博士, 審査員:山口　雄輝,徳永　万喜洋,十川　久美子,川上　厚志,立花　和則
Citation(English)	Degree:, Conferring organization: Tokyo Institute of Technology, Report number:甲第10020号, Conferred date:2015/11/30, Degree Type:Course doctor, Examiner:,,,,
学位種別(和文)	博士論文
Type(English)	Doctoral Thesis

Table of contents

CHAPTER 1: GENERAL INTRODUCTION

1.1 Transcription

Transcription is the first step of gene expression, a biochemical reaction to synthesize RNAs. From the molecular view, RNAs are polymerized nucleotide monophosphates (NMPs). They are synthesized by RNA polymerases based on the sequence of DNA templates. Many different types of RNAs are synthesized in our bodies, and these RNAs can be classified into two clusters: coding RNAs and noncoding RNAs. Eukaryotes have three to five different types of RNA polymerases, each responsible for the synthesis of a distinct set of RNAs. In this study, I focused on RNA polymerase II (pol II). The major products of RNA pol II are coding RNAs.

RNA pol II-mediated transcription is largely divided into three steps: initiation, elongation, and termination. These steps are not strictly distinguished because some of the events are associated with two adjacent steps. Major events in the initiation step include formation of the preinitiation complex, abortive initiation, and promoter clearance or promoter escape. Splicing, 5' capping, and promoter-proximal pausing are hallmarks in the elongation step. The most important events in termination are 3' processing (a.k.a. cleavage and polyadenylation). In this study, I focused on the step of transcription elongation.

For many years, transcription initiation was the most attractive field in transcription study because this step was considered to be the rate-limiting step and most important for successful RNA synthesis. For example, promoter escape, an event that occurs in the initiation step, is a rate-limiting step in RNA pol II transcription (15). It was considered that once transcription is initiated, RNA pol II progresses without obstacles until it reaches the 3' end of a gene (14). Recently, however, it was revealed that the elongation step is also highly regulated by various mechanisms. Moreover, some diseases are caused by misregulation of transcription elongation factors. For instance, mutation of human (h) Cdc73, a component of the human transcription elongation factor Paf1 complex, has been connected with breast, renal, and gastric cancer (37). Loss of hCtr9 and over-expression of hPaf1, both being components of the Paf1 complex, are also associated with pancreatic cancer (16). Increasing evidence suggests that regulation of transcription elongation is critical for precise control of gene expression. However, the current understanding on the mechanism of transcription elongation is not enough to explain some of the detailed questions on gene expression. Henceforth, I studied the step of transcription elongation for the most part in my research.

1.2 Transcription elongation factors

The regulation of transcription is a complicated and well-regulated process. In the pure *in vitro* transcription system, the purified RNA pol II enzyme can transcribe DNA nonspeecifically using naked DNA as a template. However, things are not so simple in the cells. First, DNA in the

cell nucleus is wrapped around histone proteins to form nucleosomes and condensed into the chromatin structure. Nucleosomes can serve as obstacles for transcription elongation by RNA pol II. Second, although RNA pol II has the enzymatic activity to synthesize RNA, it cannot be recruited specifically to target genes without any help of transcription factors. Third, RNA pol II would transcribe endlessly if there were no signal to terminate transcription. Fourth, the proper level of mRNA cannot be maintained without proper regulation of transcription initiation and elongation.

As is shown in Table 1-1, over a dozen transcription elongation factors have been reported in humans. These transcription factors promote or repress RNA pol II transcription elongation by different mechanisms. TFIIF is in fact a general transcription initiation factor for RNA pol II that is also capable of affecting transcription elongation by directly interacting with RNA pol II. Unlike with TFIIF, FACT promotes RNA poll II movement on chromatin during elongation by changing the chromatin structure. Handa and his colleagues originally identified transcription elongation factors, DSIF, NELF, and HDAg (57, 64). Especially, DSIF has a dual function; it promotes or represses RNA pol II elongation, in cooperation with NELF in the latter case. P-TFFb, another transcription elongation factor, is the key regulator to switch the function of DSIF by its phosphorylation from a repressor to an activator. Handa and his colleagues also identified the Paf1 complex and Tat-SF1 as factors that coactivate transcription elongation with DSIF (5). Unlike other elongation factors, the Paf1 complex seems to promote transcription elongation through multiple mechanisms: by interacting with RNA pol II directly and by facilitating histone modifications. The Paf1 complex has already been studied for a long time. However, there are still many questions about how the Paf1 complex regulates transcription elongation. The Paf1 complex is my starting point in the following study.

1.3 The Paf1 complex and Rtf1

1.3.1 Identification of the Paf1 complex

The Paf1 (RNA Polymerase II associated factor 1) complex (PAF1C) was originally identified in budding yeast and is composed of the proteins called Paf1, Ctr9, Cdc73, Rtf1 and Leo1 (3, 31). Paf1, the core component of PAF1C, was first found in search for yeast proteins associated with RNA pol II by immobilizing an antibody directed against the C-terminal domain (CTD) of RNA pol II. General transcription factors TFIIF, TFIIB, and the elongation factor TFIIS (Dst1/SII) were isolated with no surprise (36). In addition, a new uncharacterized protein was found and named Paf1. Cdc73 was also found together with Paf1 (36). Later, proteins associated with Cdc73-TAP were isolated by chromatography, and Ctr9, Rtf1, and Leo1 were identified (31). Thus, Paf1, Ctr9, Cdc73, Rtf1, and Leo1 are considered as canonical subunits of PAF1C. Similar studies in humans led to the identification of Ski8 as a metazoan-specific component of PAF1C (68). Since Ski8 is known as a subunit of the SKI complex, which is involved in the degradation of RNA, this protein seems to participate in two distinct complexes.

1.3.2 Identification of Rtf1 and its structural relationship with the Paf1 complex

Restores TBP (TATA box-binding protein) function 1 (Rtf1) was identified as a suppressor of a TBP mutant in *Saccharomyces cerevisiae* (53). Subsequent genetic and biochemical studies in budding yeast have shown that Rtf1 functions as a component of PAF1C (31, 49, 50, 52).

Each component of PAF1C is conserved in eukaryotes from yeasts to humans. Rtf1 is a bona fide subunit of PAF1C in *S. cerevisiae*. However, as shown in Table 1-2, biochemical studies on PAF1C homologs in various species showed that the physical interaction of Rtf1 with the remaining components of PAF1C is not stable in other species, such as *Schizosaccharomyces pombe* (fission yeast), *Caenorhabditis elegans* (nematode), *Drosophila melanogaster* (fly), and *Danio rerio* (zebrafish). Coimmunoprecipitation studies of Rtf1 in *S. pombe* and zebrafish could not detect the other components of PAF1C (23, 30). Immunostaining of *C. elegans* PAF1C suggested that Paf1, Ctr9, Cdc73, and Leo1 form a complex that is transported to the nucleus in an interdependent manner, while Rtf1 is localized to the nucleus independently of other PAF1C components (22). On the other hand, immunostaining of *Drosophila* polytene chromosome with antibodies against Paf1 and Rtf1 showed that these proteins are localized together over the chromosome in a similar pattern. However, the two proteins did not form a complex with a 1:1 stoichiometry (1). In humans, three groups (7, 45, 68) reported that Rtf1 does not interact with the remaining components of PAF1C, and one group (18) on the contrary reported that the N-terminally deleted Rtf1, a variant of Rtf1 probably generated by alternative translation initiation, can interact with Paf1 and/or Ctr9 to form a 6-subunit complex. To indicate the species-specific differences of PAF1C subunits, only Rtf1 from *S. cerevisiae* is referred to as a PAF1C subunit in this study.

Table 1-2. physical interaction of Rtf1

Species	Interaction Rtf1 vs PAF1C
S.serevisiae	Stable
S. pombe	Unstable
Drosophila	Unstable
Danio rerio	Unstable
Homo sapiens	?

1.3.3 The functional similarity and difference of Rtf1 and the Paf1 complex

(1) Organismal level

PAF1C is a multifunctional protein complex. Inhibition of Rtf1 and PAF1C subunits causes similar developmental defects in epidermal morphogenesis in *C. elegans* and in somitogenesis and cardiomyocyte development in zebrafish (2, 22, 23). In mouse embryos, Ctr9 and Rtf1 are required

during preimplantation development (66).

In budding yeast, deletion of *PAF1* or *CTR9* showed a severe growth defect, and deletion of *CDC73* showed a mild growth defect. However, deletion of *RTF1* or *LEO1* did not affect the cell growth (52). Moreover, *paf1*Δ and *ctr9*Δ mutants showed shared pleiotropic deficiencies, but only restricted phenotypes were observed in an *rtf1*Δ strain (4). Also, Mueller and Jaehning showed that many of the phenotypes associated with *paf1*Δ or *ctr9*Δ are not enhanced, but rather suppressed by simultaneous deletion of *RTF1* in *S. cerevisiae* (31).

(2) Transcription

PAF1C plays multiple roles in the regulation of transcription. PAF1C is involved in transcription elongation, 3' processing, 3' end formation of snoRNA, and telomere silencing (8, 13, 17, 35, 45, 47, 55, 56, 61). PAF1C is considered to regulate transcription elongation largely in two different manners: regulation of histone modifications and direct regulation of RNA pol II activity.

(a) Regulation of histone modifications

The post transcriptional modification of histones has a profound impact on transcription. In budding yeast, PAF1C facilitates histone modifications, such as histone H2B monoubiquitination (H2Bub) at K123 and histone H3 methylation at lysines 4 and 79 (K4 and K79), and Rtf1 is also critical for these modifications (20, 33, 34, 41, 60, 61, 62). These modifications are known to occur around the promoter and coding regions of actively transcribed genes, and accumulating evidence suggests that these modifications are important for efficient transcription initiation and elongation (26). The roles of PAF1C and Rtf1 in histone modifications are conserved in *Drosophila* and zebrafish (2, 23, 54). However, little is known whether human PAF1C and Rtf1 play similar roles in the regulation of histone modifications.

(b) Direct regulation of RNA pol II activity

PAF1C is also important for direct regulation of RNA pol II. In *in vitro* transcription assays using *S. cerevisiae* whole cell extracts and a naked DNA template, Rondon *et al.* showed that *paf1*Δ and *cdc73*Δ whole cell extracts were defective in elongation, whereas *rtf1*Δ and *leo*Δ whole cell extracts were not (44). In humans, *in vitro* transcription assays showed that human PAF1C cooperates with DSIF and Tat-SF1 to promote transcription elongation on a naked DNA template (5). PAF1C lacking Rtf1 was fully active in this transcription system, and the inclusion of Rtf1 had no discernible effect on the synergistic action of PAF1C, DSIF, and Tat-SF1 (our unpublished data). On the other hand, human PAF1C promotes transcription elongation together with TFIIS on a reconstituted chromatin template. Again, the presence or absence of Rtf1 did not affect the transcriptional activation potential of PAF1C (18). These data raised doubts about whether human Rtf1 is involved in transcription elongation directly.

1.4 Purpose of this study

Rtf1 is generally considered as a component of PAF1C and shares various functions with PAF1C subunits in some species. However, as mentioned above, Rtf1 is not stably associated with

PAF1C in most of the species examined, and studies *in vitro* suggest that human Rtf1 is not required for the transcriptional activation potential of PAF1C.

The first question of this study is to unveil whether human Rtf1 is involved in the regulation of transcription. If this is the case, several questions need to be answered. How does it regulate transcription? Does it function independently of PAF1C? How many genes are controlled by human Rtf1 in living cells?

1.5 Outline of this study

This thesis consists of 6 chapters as follows:

Chapter 1: General introduction

In chapter 1, general knowledge about transcription and transcription factors related to this study were introduced. Then, the purpose of this study was stated. The purpose of this study is to understand (i) whether human Rtf1 is involved in the regulation of transcription and, if so, (ii) how human Rtf1 regulates transcription.

Chapter 2: Identification of transcriptional activation potential of human Rtf1 *in vitro*

In chapter 2, I examined whether human Rtf1 is involved in the regulation of transcription. It was found that human Rtf1 is a nuclear protein that is essential for efficient transcription elongation in *in vitro* transcription assays.

Chapter 3: Identification of coactivator activity required for human Rtf1-mediated transcriptional activation

In chapter 3, the mechanism of action of human Rtf1 in the regulaton of transcription elongation was studied. I reached an unexpected finding that human Rtf1 requires “Rtf1 coactivator” activity, which is most likely unrelated to PAF1C, DSIF, Tat-SF1, and TFIIS.

Chapter 4: Structure-function analysis of human Rtf1

In chapter 4, human Rtf1 deletion mutants were prepared to examine whether human Rtf1 is involved in the regulation of histone modifications. To further understand how human Rtf1 regulates transcription, mutational studies were performed. Finally, the physical interaction of human Rtf1 and PAF1C was also examined by using deletion mutants.

Chapter 5: The roles of human Rtf1 and PAF1C in transcriptional regulation in HeLa cells

In chapter 5, to extend my findings to living cells, gene expression changes after knockdown of Rtf1, Paf1, and Ski8 were studied by RNA-seq analysis. In addition, chromatin immunoprecipitation analysis was performed to study the recruitment of Pol II, Rtf1, and PAF1C to target genes.

Chapter 6: Discussion

In chapter 6, based on the results obtained from chapters 2 to 5, I discussed the functions of human Rtf1 from three aspects: (i) PAF1C-dependent and -independent functions of human Rtf1; (ii) PAF1C-controlled, Rtf1-controlled, and H2Bub-controlled genes; and (iii) functional similarities and differences of Rtf1 proteins among species.

Table1-1. Human Transcription Elongation Factors

Elongation Factors	Subunits	Properties
TFIIF	RAP30, RA74	One of general transcription factor, promotes elongation
SII/TFIIS		Releases arrest, increases transcription fidelity, promotes chromatin elongation
SIII/Elongin	A,B,C	promotes transcription elongation
ELL		promotes transcription elongation
P-TEFb	Cdk9, CyclinT	releases repression of elongation, protein kinase activity
DSIF	Spt4, Spt5	induces pausing, promotes elongation, involves in mRNA processing
NELF	A,B,C/D,E	induces pausing, involves in mRNA processing
Tat-SF1		promotes transcription elongation
Paf1 Complex	Paf1, Ctr9, Leo1, Cdc73, Ski8	promotes transcription elongation, involves in histone modification
HDAg		D type lever virus protein
Spt6		promotes transcription elongation, facilitates chromatin elongation
FACT	Spt16, SSRP1	facilitates chromatin elongation
Elongator	Elp1, Elp2, Elp3	facilitates chromatin elongation, histone acetyl transferase activity

CHAPTER 2: IDENTIFICATION OF TRANSCRIPTIONAL ACTIVATION POTENTIAL OF HUMAN RTF1 IN VITRO

2.1 Introduction

How do transcription elongation factors directly regulate RNA pol II? This has been one of the most fundamental questions in the study of transcription. In the living cells, numerous transcription factors are involved in transcription. The mechanism of transcriptional regulation is very complicated because of spatiotemporally regulated transcription factors. To understand the mechanism of RNA pol II elongation along DNA templates in detail, it is necessary to simplify the transcription system used. Thus, *in vitro* transcription assays are idealized for mechanistic study. *In vitro* transcription assays can be classified into two types based on the templates used: naked DNA and reconstituted chromatin. The use of naked DNA template eliminates the effect of nucleosomes on transcription. Hence, naked DNA is useful for studying the direct effect of transcription elongation factors on RNA pol II.

Many studies in budding yeast showed that Rtf1 is an integral component of PAF1C facilitating histone modifications in a transcription-dependent manner (20, 33, 34, 41, 60, 61, 62). In humans, however, it is still unclear whether Rtf1 is involved in transcription or not (5, 18, and our unpublished data). In this chapter, an unexposed role of human Rtf1 in transcription was unveiled. Experiments were performed to investigate human Rtf1 from three aspects. (i) Where is human Rtf1 localized in HeLa cells? (ii) Does human Rtf1 have a transcriptional activation potential *in vitro*? (iii) Which step of transcription is controlled by human Rtf1?

2.2 Results

2.2.1 Localization of Rtf1 in human cells

To understand whether human Rtf1 directly regulates transcription, the subcellular localization of Rtf1 in HeLa cells was examined. If Rtf1 is a transcription factor, at least a fraction of Rtf1 molecules should be localized in the nucleus where transcription occurs. Immunostaining was performed to determine the localization of Rtf1 in HeLa cells (Fig. 2-1). Anti-Rtf1 antibody was used to stain Rtf1 molecules, and 4',6-diamidino-2-phenylindole (DAPI) was used to counterstain DNA. Immunofluorescence signals obtained with anti-Rtf1 antibody were perfectly overlapped with those of DAPI (Fig. 2-1). These data clearly indicated that human Rtf1 is a nuclear protein.

2.2.2 Identification of transcriptional activation potential of human Rtf1

Next, to determine whether Rtf1 regulates transcription directly, *in vitro* transcription assays were performed using Rtf1-depleted HeLa cell nuclear extracts (NE) and naked DNA templates. *In vitro* transcription is a cell-free system in which three main elements — the transcription machinery, template DNA, and nucleotide triphosphates (NTPs) — are utilized. Transcription reactions occur in microcentrifuge tubes by adding these factors sequentially and incubating for sometime (Fig. 2-2). HeLa NE, which contain RNA pol II and transcription factors, was used as a source of these protein factors while the plasmid pSLG402 (Fig. 2-2), which contains two G-free cassettes placed downstream of the adenovirus major-late promoter (MLP), was used as a template. In addition, four NTPs containing radioactive [α-^{32}P] UTP were used to label newly synthesized transcripts. Synthesized transcripts were treated with ribonuclease T1 (RNase T1), an endoribonuclease specific to guanosine, to liberate RNA fragments corresponding to G-free cassettes. Urea polyacrylamide gel electrophoresis (PAGE) was performed following phenol/chloroform extraction and ethanol precipitation of the transcripts. Radioactive signals were detected by autoradiography. As reported previously by our group (5), HeLa NE supported efficient transcription initiation and elongation from the pSLG402 template. After RNase T1 treatment, 83-nt and 377-nt fragments corresponding to the promoter-proximal and promoter-distal G-free cassettes were generated (Fig. 2-2). The former represents the efficiency of initiation and "early" elongation, and the distal-to-proximal ratio represents the efficiency of "late" elongation.

To examine the role of Rtf1 in transcription, *in vitro* transcription assays were performed using Rtf1-depleted HeLa NE. Immunodepletion was performed using anti-Rtf1 antibody as described in the Materials and methods section. Briefly, anti-Rtf1 antibody was immobilized to protein G agarose beads and incubated with HeLa NE. The supernatant was then collected by centrifugation. This process was repeated a few more times, and the final supernatant was regarded as Rtf1-depleted NE. As a result, Rtf1 was efficiently removed from the NE whereas the levels of all the subunits of PAF1C, Spt5 (the large subunit of DISF), Tat-SF1, and Rpb1 were negligibly affected by this procedure (Fig. 2-3A).

Strikingly, Rtf1-depleted NE (NEΔRtf1) exhibited severe transcription defects compared to IgG-depleted control NE (NEΔIgG) (Fig. 2-3C). Moreover, the addition of Flag-Rtf1 expressed in and purified from insect cells (Fig. 2-3B) almost fully restored the transcription defects (Fig. 2-3C). These results demonstrated that human Rtf1 is a transcription factor indispensable for transcription *in vitro*.

2.2.3 Determination of the transcription step controlled by human Rtf1

The step of transcription that was affected by Rtf1 depletion was not clear from the above results, because the reduction of promoter-proximal products can be caused by defects in initiation, early elongation, or both. To pinpoint the step of transcription defect,

primer extension assays were performed. Transcripts obtained from "cold run" *in vitro* transcription reactions using a plasmid template containing the adenovirus E4 promoter (E4P) and NEΔIgG or NEΔRtf1 were subjected to primer extension assays. To quantify transcripts of ≥ 29 nt, ≥ 69 nt, ≥ 98 nt, and ≥ 432 nt, four radiolabeled primers (Fig. 2-5A) were prepared and respectively hybridized to "cold" run-off products, followed by reverse transcription (RT) (Fig. 2-4). As shown in Figure 2-5B, the levels of ≥ 29 nt, ≥ 69 nt, and ≥ 98 nt transcripts were negligibly affected by Rtf1 depletion. By contrast, the level of ≥ 432 nt transcripts was significantly reduced by the immunodepletion and recovered by the addition of Flag-Rtf1 to NEΔRtf1. Since the stimulatory effect of Rtf1 was clearly dependent on transcript length, it was concluded that Rtf1 acts only on the elongation phase of transcription *in vitro*.

As a result of primer extension reactions with primers A, B, and C, the products with sizes significantly greater than expected were also obtained. Notably, the RT products of 200 to 300 nt were reduced by Rtf1 depletion and restored by its addition. How could this be explained? There are two potential sources of such products with heterogeneous sizes: (i) transcription initiation from multiple sites during *in vitro* transcription and (ii) primer mishybridization during reverse transcription. Since the patterns of longer-than-expected products seem to vary with the primers used, these products were most likely generated by primer mishybridization. The DNA template used here contains a 380-nt G-free cassette downstream of the adenovirus E4 promoter, and primers A, B, and C are designed to hybridize within the G-free cassette. These primers may have mishybridized to non-target sites within the cassette because of its low complexity. Given this assumption, RT products with heterogenous sizes can be equated with transcripts with the same sizes. Thus, it can be concluded that Rtf1 is particularly important for the synthesis of >200 nt transcripts under the conditions employed in primer extension assays.

2.3 Conclusions

The role of human Rtf1 in transcription was unknown. Past studies on human PAF1C and Rtf1 suggested that human Rtf1 is not likely to regulate transcription directly. In this chapter, I found that human Rtf1 is in fact very important for efficient transcription elongation *in vitro*.

First, human Rtf1 is a nuclear protein. Second, human Rtf1 is an essential transcription factor. It was found that human Rtf1 is indispensable for the synthesis of full-length transcripts *in vitro* using naked DNA templates. Third, human Rtf1 is a transcription elongation factor. Depletion of human Rtf1 from HeLa NE resulted in a severe transcription defect. This defect was not caused by codepletion of other important transcription factors or RNA pol II, as addition of recombinant Rtf1 restored transcription. It was concluded that human Rtf1 is critical for the "late" transcription elongation step.

2.4 Materials and methods

2.4.1 Immunofluorescence microscopy

HeLa cells were seeded to 35-mm dishes containing collagen-coated coverslips at 5 x 10^5 cells/dish and incubated overnight. Cells in 35-mm dishes were washed twice with PBS (137 mM NaCl, 2.7 mM KCl, 10 mM Na_2HPO_4, 1.76 mM KH_2PO_4) and fixed with 1 ml of 4% formaldehyde solution in PBS for 15 min at room temperature (r.t.). Cells were subsequently washed twice in PBS, incubated in 1 ml of 0.25% Triton X-100 solution in PBS for 20 min at r.t., washed twice in PBS and incubated in blocking solution (1% BSA, 0.02% azide in PBS) for 1 h at r.t. Coverslips were transferred onto a piece of parafilm, overlayed with 100 µl of anti-Rtf1 antibody (Bethyl, A300-179A) diluted in blocking solution and incubated overnight at 4 ºC. The coverslips were washed twice with 0.1% NP-40 solution in PBS, overlayed with 100 µl of anti-rabbit antibody conjugated with AlexaFluor-594 (Molecular Probes) solution for 30 min at r.t., washed twice in 0.1% NP-40 solution, washed twice with PBS, and mounted on slide glasses using 10 µl of the Vectashield mounting medium containing DAPI (Vector Laboratories). Stained cells were inspected under the fluorescence microscope BX51 (Olympus).

2.4.2 Preparation of nuclear extracts from HeLa cells

Nuclear extracts prepared according to Dignam et al. (10). Cultured HeLa cells were centrifuged at 3,000 rpm for 5 min. The cell pellets were washed once with PBS(+) (137 mM NaCl, 2.7 mM KCl, 10 mM Na_2HPO_4, 1.76 mM KH_2PO_4, 1 mM $MgCl_2$, 0.5 mM DTT) and suspended in buffer A (10 mM HEPES pH7.9, 10 mM KCl, 1.5 mM $MgCl_2$, 0.5 mM DTT). Cells were transferred to a Dounce homogenizer and disrupted by 20 strokes of the pestle. Samples were centrifuged at 3,000 rpm for 10 min at 4°C, and the cytoplasmic supernatant fractions were transferred to new tubes. Pellets were centrifuged again for maximal removal of the supernatants. Nuclear pellets were resuspended in buffer C (20 mM HEPES pH7.9, 25% glycerol, 420 mM NaCl, 1.5 mM $MgCl_2$, 0.2 mM EDTA, 0.5 mM PMSF, 0.5 mM DTT), lysed with a gentle agitation for 30 min, and centrifuged at 1,500 rpm for 10 min at 4°C. The supernatants, which represent nuclear extracts, were dialyzed against buffer D (20 mM HEPES pH7.9, 20% glycerol, 100 mM KCl, 12.5 mM $MgCl_2$, 0.2 mM EDTA, 0.5 mM PMSF, 0.5 mM DTT). After centrifugation, the supernatant was transferred to new tubes and stored at -80°C.

2.4.3 *In vitro* transcription assays

In vitro transcription assays were performed using pSLG402, which contained the adenovirus major-late promoter (25) and 2 µl of HeLa NE or 6 µl of P1.0 with or without 0.2 pmol of wild-type or mutant Rtf1. After preincubation of 22-µl reactions for 40 min, 3 µl of four NTPs (final concentrations of 30 µM ATP, 300 µM CTP, 300 µM GTP, and 2.5 µM [α-^{32}P] UTP) and 500 U of RNase T1 were added, and initiation/elongation was allowed to

proceed for 20 min unless stated otherwise. The reactions were stopped at the indicated time by the addition of 100 µl of stop buffer (10 mM Tris-HCl, pH 7.5, 0.3 M NaCl and 5 mM EDTA). Synthesized transcripts were then purified by phenol-chloroform extraction and ethanol precipitation and resolved by 8% urea PAGE. The amounts of radioactivity incorporated were quantified using the phosphorimager STORM 860 (Amersham Pharmacia Biotech).

2.4.4 Immunoblotting

Protein samples were loaded onto an SDS-polyacrylamide gel and electrophoresed using running buffer (25 mM Tris-HCl, 190 mM glycine, 0.1% SDS) at 30 mA/gel. Separated proteins were transferred to a polyvinylidene fluoride membrane (Millipore) using transfer buffer (25 mM Tris-HCl, 190 mM glycine, 20% methanol), and the membrane was incubated with blocking buffer (3% bovine serum albumin (BSA) in TBST (20 mM Tris-HCl pH7.5, 150 mM NaCl, 0.1% Tween 20)) for 30 min. Next, the blocked membrane was incubated with primary antibody solution (diluted in blocking buffer) for 1 h at r.t. or overnight at 4ºC. The membrane was washed three times for 5 min each with TBST and incubated in HRP-conjugated secondary antibody solution (diluted in TBST) for 30 min at r.t. The membrane was again washed three times for 5 min each with TBST and rinsed in TBS (20 mM Tris-HCl pH7.5, 150 mM NaCl). Immunoreactive bands were visualized by ECL (Amersham Biosciences).

2.4.5 Antibodies

Antibodies used in this chapter are summarized. Anti-Leo1, anti-Spt5, and anti-TatSF1 antibodies were produced in-house in rabbits (4). The following commercial antibodies were also used: anti-Ctr9 (Bethyl, A301-395A), anti-Rtf1 (Bethyl, A300-179A), anti-Paf1 (Bethyl, A300-173A), anti-Cdc73 (Santa Cruz, sc-33638), anti-Ski8 (Abcam, ab57840), anti-Rpb1 (8WG16).

2.4.6 Preparation of recombinant proteins

Human Rtf1 cDNA was prepared by RT-PCR from total RNA extracted from HeLa cells and cloned into pFastBac1 (Invitrogen) with the N-terminal Flag tag. Recombinant baculovirus was produced using pFastBac1/Flag-Rtf1. The resulting baculovirus was infected into insect Sf9 cells. Sf9 cells were harvested 3 days post-infection. The cell pellet was lysed with high-salt buffer (500 mM NaCl, 1.0% NP-40, 50 mM Tri-HCl pH8.0), and the supernatant was obtained by centrifugation. The lysate was incubated with anti-Flag agarose beads (Sigma) for 2 h at 4ºC. Next, agarose beads were washed with high-salt buffer three times and then equilibrated with NE (-) buffer (20 mM HEPES pH7.9, 20% glycerol, 100 mM KCl, 0.2 mM EDTA, 0.5 mM PMSF, 0.5 mM DTT) twice. Finally, the

Flag-Rtf1 protein was eluted with 0.1 mg/ml Flag peptide in NE (-) buffer.

2.4.7 Immunodepletion

Immunodepletion was performed as previously described (32, 64). Protein G-immobilized agarose beads were washed by high-salt buffer twice. Antibody was added to the protein G beads and incubated for 1 h at r.t. Antibody-immobilized beads were washed three times with high-salt buffer, followed by equilibrium of the beads with NE (+) buffer. An aliquot of the beads were incubated with HeLa NE at 4ºC for 2 h, and the beads were separated by centrifugation. The supernatant was incubated with another aliquot of antibody-immobilized beads, and the same process was repeated two to four more times. The supernatant of the final round was collected and stored at -80ºC.

2.4.8 Primer extension

First, *in vitro* transcription was performed using pTF3-6C2AT, the supercoiled plasmid containing the adenovirus E4 promoter, without radioactive nucleotides or RNase T1 treatment, as previously described (64). RNA was purified by phenol/chloroform extraction and ethanol precipitation. Purified RNA was dissolved in MilliQ water. On the other hand, the 5' end of primers were ^{32}P-labelled by using [γ-^{32}P] ATP and polynucleotide kinase. Next, reverse transcription was performed by using Superscript III, one of radiolabelled primers, and purified *in vitro* transcription products. The synthesized cDNA was purified by phenol/chloroform extraction and ethanol precipitation and loaded onto an 8% urea polyacrylamide gel. The amounts of radioactivity incorporated were quantified using the phosphorimager STORM 860 (Amersham Pharmacia Biotech). The sequences of primers are as follows:

A, 5′-GAATAATGAGGAAAGGAGAGT-3′;
B, 5′-GATGATAGATTTGGGAAATATAA-3′;
C, 5′-GGAGAGTAGGGTGGTATAG-3′;
D, 5′-AGGAAACAGCTATGACCATG-3′.

The expected lengths of the products were 29 nt, 69 nt, 98 nt, and 432 nt, respectively.

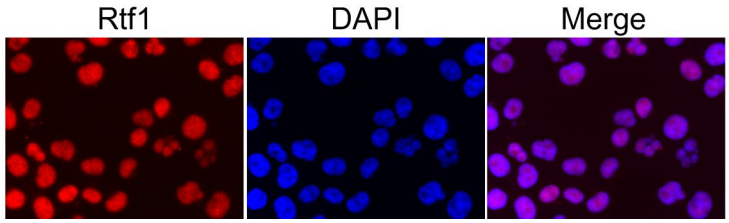

Figure 2-1. Human Rtf1 is a nuclear protein.
The subcellular localization of human Rtf1 was examined by immunofluorescence microscopy. HeLa cells were stained with anti-Rtf1 antibody and counterstained with DAPI.

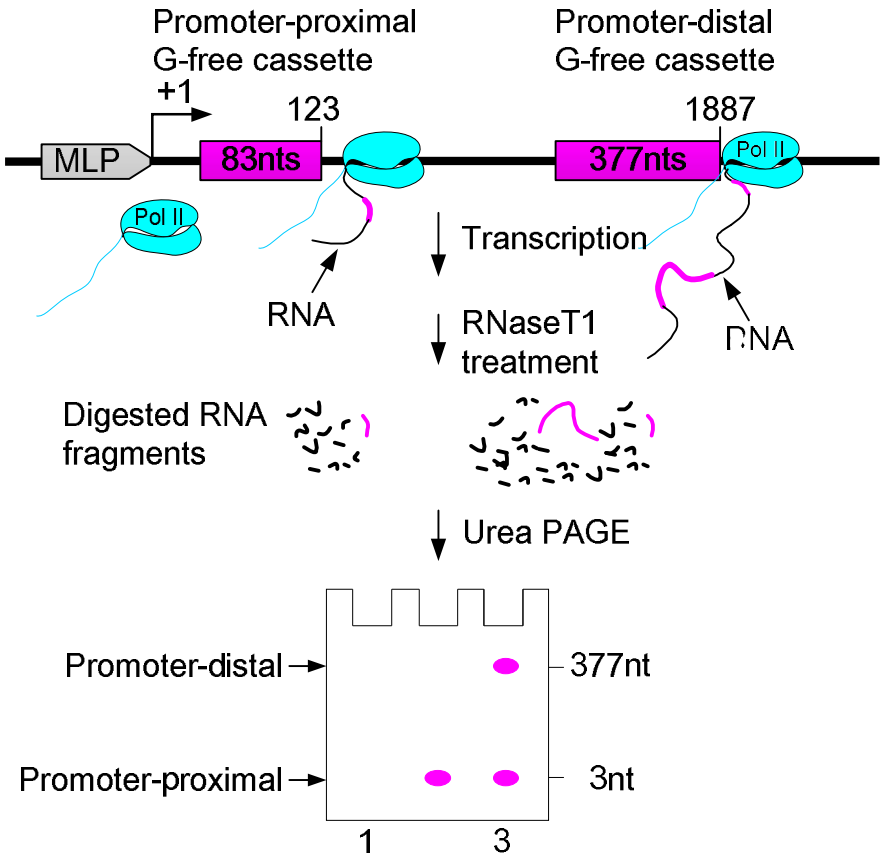

Figure 2-2. The scheme of *in vitro* transcription assay.

In vitro transcription was performed using pSLG402, HeLa NE or NE fractions, and the synthesized transcripts were treated by RNase T1. The digested RNA fragments were subjected to urea PAGE after phenol/chloroform extraction and ethanol precipitation. The DNA template pSLG402 contains two G-free cassettes. MLP, adenovirus major-late promoter. Promoter-proximal and -distal products are indicated with arrows.

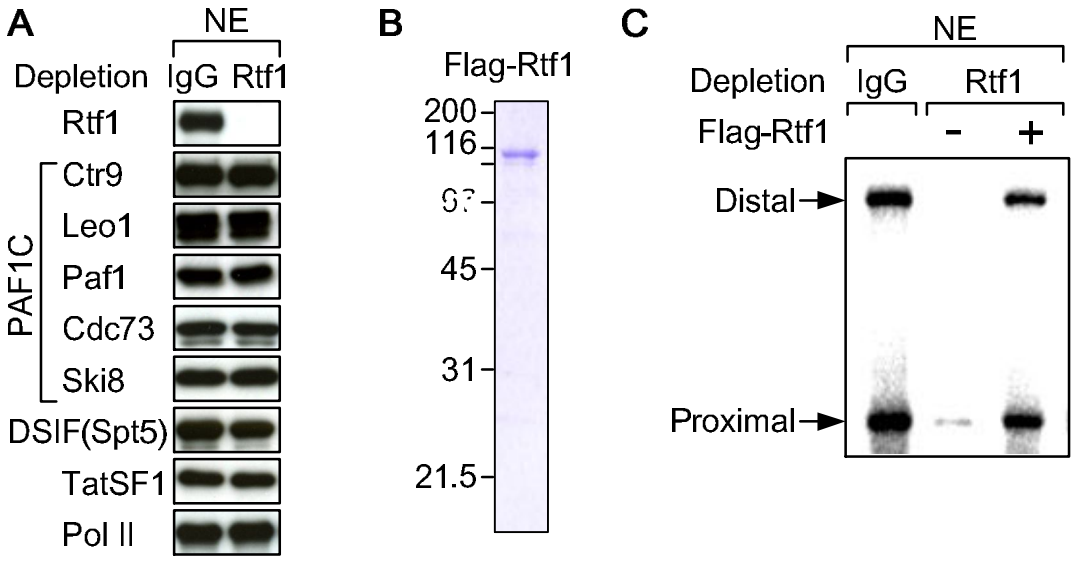

Figure 2-3. Human Rtf1 is essential for transcription *in vitro*.

(A) HeLa NE immunodepleted with control IgG or anti-Rtf1 antibody were analyzed by immunoblotting with the indicated antibodies. (B) Coomassie blue-stained Flag-Rtf1 purified from insect cells. (C) *In vitro* transcription was performed using pSLG402, one of the NEs shown in (A), and Flag-Rtf1. Promoter-proximal and -distal products are indicated with arrows.

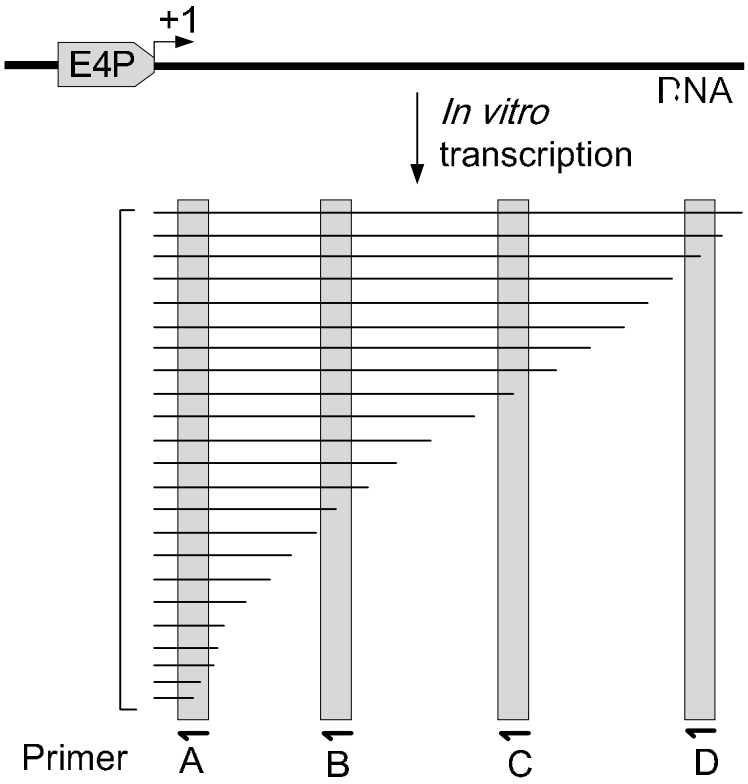

Figure 2-4. The scheme of primer extension assays.

In vitro transcription was performed using pTF3-6C2AT, containing the adenovirus E4 promoter (E4P), and NE without radioactive nucleotides or RNase T1 treatment. Purified transcripts were subjected to reverse transcription using Superscript III and one of [^{32}P]-labeled primers designed to hybridize to transcripts at different distances from the transcription start site.

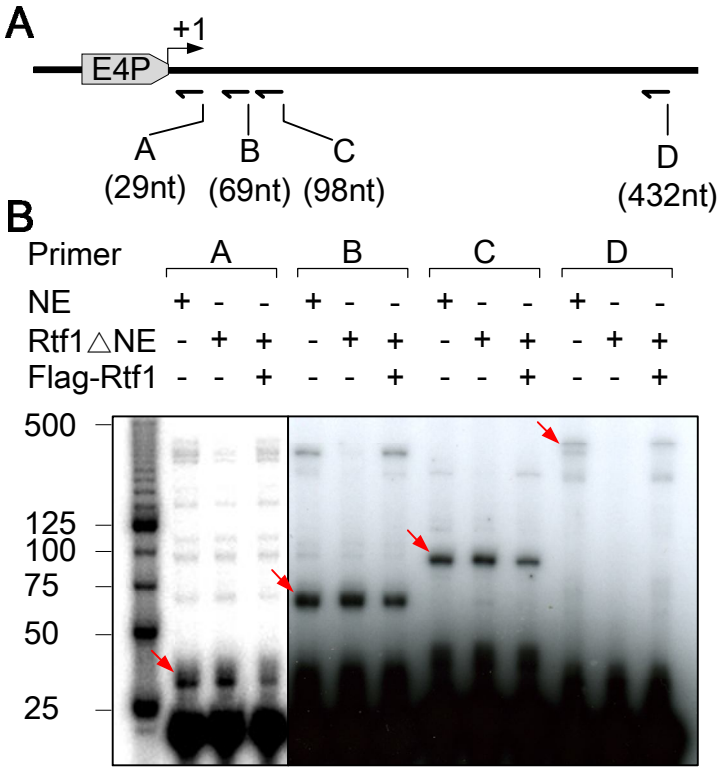

Figure 2-5. Human Rtf1 is a transcription elongation factor.

(A) Schematic structure of the DNA template and the primer-binding sites used in (B). E4P, adenovirus E4 promoter. (B) Primer extension analysis of the products obtained with HeLa NE immunodepleted with control IgG (NEΔIgG) or anti-Rtf1 (NEΔRtf1). Specific primer extension products are indicated with arrows.

CHAPTER 3: IDENTIFICATION OF COACTIVATOR ACTIVITY REQUIRED FOR HUMAN RTF1-MEDIATED TRANSCRIPTIONAL ACTIVATION

3.1 Introduction

In chapter 2, it was found that human Rtf1 is indispensable for transcription *in vitro*. Rtf1-depleted HeLa NE showed severe transcription defect, and the transcription defect was restored by the addition of recombinant Flag-Rtf1. However, it is still unclear how human Rtf1 promotes transcription elongation.

The first purpose of chapter 3 is to understand if human Rtf1 promotes transcription elongation alone or with a help of another factor(s)? Rtf1 is generally considered as a component of PAF1C. Therefore, in this chapter, various biochemical approaches were taken to understand if human Rtf1 requires PAF1C for the regulation of transcription elongation. Also, several other transcription factors, such as DSIF, TatSF1, and TFIIS, were examined if they are involved in Rtf1-mediated transcriptional activation *in vitro*.

3.2 Results

3.2.1 Requirement of coactivator activity in human Rtf1-mediated transcriptional activation

To further understand the mechanism of action of Rtf1, I investigated whether another factor is required for Rtf1-mediated transcriptional activation. To this end, I used a previously used strategy (5, 57, 64). Using a phosphocellulose P11 column, HeLa NE was fractionated into the flow-through fraction (P0.1), the 0.3 M KCl eluate (P0.3), and the 1.0 M KCl eluate (P1.0) (Fig. 3-1A). P1.0 contained most of the general transcription factors and RNA pol II and was sufficient for directing transcription initiation; however, because it lacked several transcription elongation factors, it did not support efficient transcription elongation alone under the conditions used (Figs. 3-1A–C; see also Ref. 5). Contrary to the observations obtained using NEΔRtf1 (Fig. 2-3C), the addition of Flag-Rtf1 to P1.0 did not facilitate the efficient synthesis of the promoter-distal region (Fig. 3-1C), suggesting that another factor is required for Rtf1-mediated transcriptional activation.

To search for such an "Rtf1 coactivator," a limited amount of P0.1 and/or P0.3 was added together with Flag-Rtf1 to P1.0. As shown in Figure 3-1B, Rtf1 was partitioned into P0.1 and P0.3 to a similar extent, whereas PAF1C was largely found in P0.3. In the absence of exogenous Flag-Rtf1, P0.1 and/or P0.3 did not enhance elongation at the concentrations used. To the contrary, P0.3 slightly repressed transcription, possibly because of the

enrichment of an inhibitory factor(s) (Fig. 3-1E). The combination of Flag-Rtf1 and P0.3 significantly increased the promoter-distal products. P0.1 showed a weak coactivation effect with Flag-Rtf1, whereas no further increase was observed in response to the combination of P0.1 and P0.3, suggesting that the same factor was responsible for the effects of P0.1 and P0.3 on Rtf1.

To ensure the presence of Rtf1 coactivator, P0.3 was then fractionated on a DEAE Sepharose column (Fig 3-1A), which yielded the flow-through fraction (D0.1) and three eluate fractions (D0.225, D0.3, and D1.0). At this step, Rtf1 and PAF1C were largely separated, with Rtf1 exclusively found in D0.225 and PAF1C mostly detected in D0.3 (Fig. 3-1D). With regard to other transcription elongation factors, DSIF and Tat-SF1 were largely removed from the active fraction at the phosphocellulose chromatography step (Fig. 3-1B), whereas a portion of DSIF and Tat-SF1 was cofractionated with PAF1C and found in D0.3 (Fig. 3-1D). At the functional level, only D0.3 exhibited strong coactivation with Flag-Rtf1 (Fig. 3-1E). Incidentally, D0.1 repressed the synthesis of the promoter-proximal region, suggesting that the inhibitory factor(s) found in P0.3 was fractionated into D0.1. These data undoubtedly indicated that Rtf1-meditated transcription requires a coactivator(s) and showed that the P0.3D0.3 fraction contains the coactivator(s). The reason P0.1 and P.3 did not promote transcription elongation alone (Fig. 3-1C) was probably because Rtf1 was diluted as a result of phosphocellulose column chromatography.

To identify the Rtf1 coactivator, P0.3D0.3 was subjected to heparin Sepharose chromatography (Fig. 3-2A). The resulting active fraction (H0.4) was further resolved on a mono S column, and the most active fractions (mono S#5) was assayed. As shown in Figure 3-2C, P0.3D0.3H0.4S#5 strongly coactivated the synthesis of the promoter-distal region with increasing concentrations of Flag-Rtf1. The transcription activation potential of Rtf1 seemed to be saturated at around 240 ng/reaction. Remarkably, in the absence of the coactivator fraction, Flag-Rtf1 did not induce the synthesis of promoter-distal products, even at the highest concentration examined (960 ng/reaction) (Fig. 3-2C). These data showed that human Rtf1 is critically dependent on the partially purified coactivator fraction.

To characterize the physical properties of the Rtf1 coactivator further, glycerol gradient sedimentation analysis was performed. The results indicated that the apparent molecular weight of the coactivator is 200 kDa (Fig. 3-5).

To understand the mechanism of human Rtf1-mediated transcriptional activation, the identity of the Rtf1 coactivator is a key. Despite various attempts, however, further purification and identification of the Rtf1 coactivator have not been successful to date.

3.2.2 Human Rtf1 promotes transcription elongation independently of DSIF

Based on past studies, the most likely candidate for the Rtf1 coactivator was PAF1C. Considering a recent study in *S. cerevisiae* showing that the Plus3 domain of Rtf1 interacts with the C-terminal region of Spt5, a large subunit of DSIF (59), DSIF might also be a

candidate for the Rtf1 coactivator. Given the finding that, at the phosphocellulose step, the Rtf1 coactivator activity was partitioned into P0.1 and P0.3 fractions to a similar level (Fig. 3-1C), it can be said that PAF1C and DSIF did not comigrate with the coactivator activity at this step. The possibility remained that these factors are responsible for the coactivator activity, however, since the detectable levels of PAF1C and DSIF still existed in the P0.3D0.3H0.4S#5 fraction (Fig. 3-2B), indicative of their weak physical interactions with Rtf1.

To explore this possibility, PAF1C was immunodepleted from HeLa NE using antibodies against PAF1C. As a result, Cdc73, Ctr9, Leo1, and Paf1, but not Ski8, were efficiently depleted by antibodies against the respective subunits (Fig. 3-3A). With these antibodies, other subunits of the complex were codepleted to varying degrees, with the Ski8 subunit showing the greatest resistance to codepletion. When anti-Leo1 antibody was used to deplete PAF1C from the P0.3D0.3 coactivator fraction, all the PAF1C subunits including Ski8 were codepleted to a satisfactory level (Fig. 3-3B). This apparent discrepancy was probably due to the presence of the SKI complex, another Ski8-containing complex. Since the SKI complex was fractionated into P0.1 (data not shown), Ski8 found in P0.3D0.3 was almost entirely incorporated into PAF1C and was codepleted with the other PAF1C subunits. With regard to DSIF, it was efficiently and specifically depleted from P0.3D0.3 using anti-Spt5 antibody (Fig. 3-3B).

Contrary to our expectations, the coactivator fraction from which Leo1 or DSIF was depleted (P0.3D0.3ΔLeo1 or P0.3D0.3ΔDSIF) was as efficient as the mock-depleted fraction in stimulating the synthesis of the promoter-distal region (Figs. 3-3C–E), suggesting that none of these proteins are responsible for the coactivator activity. To rule out the possibility that PAF1C and DSIF are functionally redundant and that either can suffice as a coactivator for Rtf1, we codepleted PAF1C and DSIF from P0.3D0.3 using the combination of anit-Leo1 and anti-Spt5 antibodies (Fig. 3-3B). As a result, codepletion of PAF1C and DSIF did not substantially affect Rtf1-mediated transcriptional activation (Figs. 3-3C–E), suggesting that these factors are not responsible for the coactivator activity of P0.3D0.3.

3.2.3 Human Rtf1 promotes transcription elongation independently of the Paf1 complex

Even after depletion using anti-Leo1 antibody, small amounts of Ctr9, Paf1, and Cdc73 remained in P0.3D0.3ΔLeo1 (Fig. 3-3B). To eliminate the possibility that the residual PAF1C subunits remaining in the depleted fraction supported the Rtf1-mediated transcriptional activation, I sought to deplete PAF1C subunits completely. After several attempts, the combination of anti-Leo1, anti-Cdc73, and anti-Paf1 antibodies was found to result in an almost complete depletion of PAF1C from P0.3D0.3, giving rise to P0.3D0.3ΔPAF1C (Fig. 3-4A). As shown in Figure 3-5B, P0.3D0.3ΔPAF1C coactivated the

synthesis of the promoter-distal region as efficiently as mock-depleted P0.3D0.3, suggesting that Rtf1 promotes transcription elongation independently of PAF1C *in vitro*.

3.2.4 Tat-SF1, TFIIS, and histones are not involved in human Rtf1-mediated transcriptional activation

I performed immunoblot analysis of other candidate coactivators. First, Tat-SF1 and TFIIS were considered because they are known to activate transcription in corporation with PAF1C *in vitro* (5, 18). While a small fraction of Tat-SF1 was found in P0.3D0.3 (Figs. 3-1B and D), further purification resulted in a separation of the coactivator activity and Tat-SF1 (Fig. 3-5). With regard to TFIIS, it was fractionated almost entirely into the P1.0 fraction (Fig. 3-6A). Since P1.0 was used as a source of the general transcription factors and Pol II in our transcription assays, The possibility cannot be excluded that TFIIS plays a role in the Rtf1-dependent activation. It can be said, however, that TFIIS is not responsible for the coactivator activity found in P0.3D0.3.

Considering the tight functional link between Rtf1 and chromatin (17, 29, 41, 58), histones could modulate the transactivation potential of Rtf1 *in vitro*. Immunoblot analysis showed, however, that P0.3D0.3 had an undetectable level of histones H2B and H3, and most of the histones were found depleted at the phosphocellulose column chromatography step (Fig. 3-6B). Probably, basic histone molecules were so tightly associated with phosphocellulose that they were barely eluted from the column even at the highest KCl concentration employed. In any case, these results suggest that histones are not involved in the coactivator's function.

3.3 Conclusions

A novel function of human Rtf1 to directly regulate transcription elongation *in vitro* was introduced in chapter 2. However, it was unknown exactly how human Rtf1 regulates transcription elongation. In chapter 3, it was shown that human Rtf1 regulates transcription elongation in a manner different from the known transcription elongation factors.

First, human Rtf1 requires coactivator to promote transcription elongation *in vitro*. The result that human Rtf1 alone cannot promote transcription elongation *in vitro* showed that the process of Rtf1-mediated transcription elongation is not so simple.

Next, human Rtf1 promotes transcription elongation independently of known transcription elongation factors, such as PAF1C, DSIF, TatSF1, and TFIIS. These results suggested that human Rtf1-mediated transcription is distinct from the known transcription regulation pathways. However, the identity of the Rtf1 coactivator is still unknown.

Lastly, human Rtf1 promotes transcription elongation *in vitro* in a chromatin-independent manner. Naked DNA templates were used throughout this study. Moreover, the lack of histone proteins in the *in vitro* transcription system employed showed that Rtf1 promotes transcription elongation in a manner independent of its function to

regulate histone modifications.

3.4 Materials and methods

3.4.1 Glycerol gradient sedimentation

Six solutions containing 100 mM KCl, 20 mM HEPES, and different concentrations of glycerol (53%, 48%, 43%, 38%, 33%, and 28%) were prepared, and 100 µl of each solution was used to form a gradient in ultracentrifuge tubes (Beckman). Then, 50 µl of samples were layered on the top. MWGF1000-1KT (Sigma-Aldrich) was used as molecular weight marker. After centrifugation at 50 krpm for 20 h at 4ºC using the XL-100 ultracentrifuge (Beckman). Totally 13 fractions, 50 µl each, were collected from the top to the bottom.

3.4.2 Antibodies

Antibodies used in this chapter are summarized below. Anti-Leo1, anti-Spt5, and anti-TatSF1 antibodies were produced in-house in rabbits (4). The following commercial antibodies were also used: anti-Ctr9 (Bethyl, A301-395A), anti-Rtf1 (Bethyl, A300-179A), anti-Paf1 (Bethyl, A300-173A), anti-Cdc73 (Santa Cruz, sc-33638), anti-Ski8 (Abcam, ab57840), anti-Rpb1 (8WG16), anti-TFIIS (Transduction Laboratories, S84820), anti-H2B (Abcam, ab1790), anti-H3 (Abcam, ab1791).

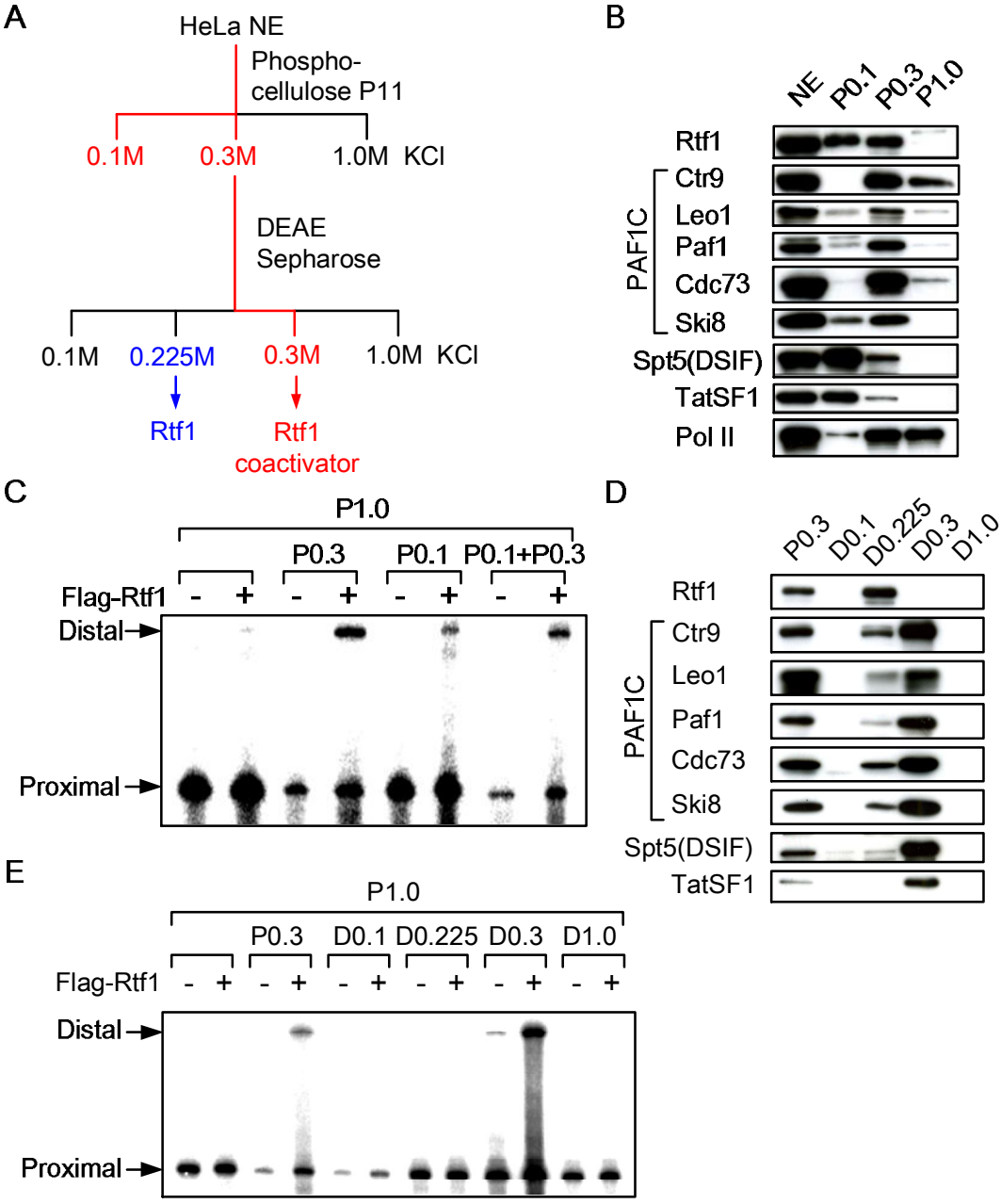

Figure 3-1. Human Rtf1 requires another factor to promote transcription elongation. (A) The separation scheme. (B) Immunoblot analysis of HeLa NE and phosphocellulose column fractions. (C) *In vitro* transcription assays of phosphocellulose column fractions. Reactions containing pSLG402 and the indicated phosphocellulose column fraction(s) were incubated with or without Flag-Rtf1. Promoter-proximal and -distal products are indicated with arrows. (D) Immunoblot analysis of DEAE Sepharose column fractions. P0.3 (input) was analyzed for comparison. (E) *In vitro* transcription assays of DEAE Sepharose column fractions. Promoter-proximal and -distal products are indicated with arrows.

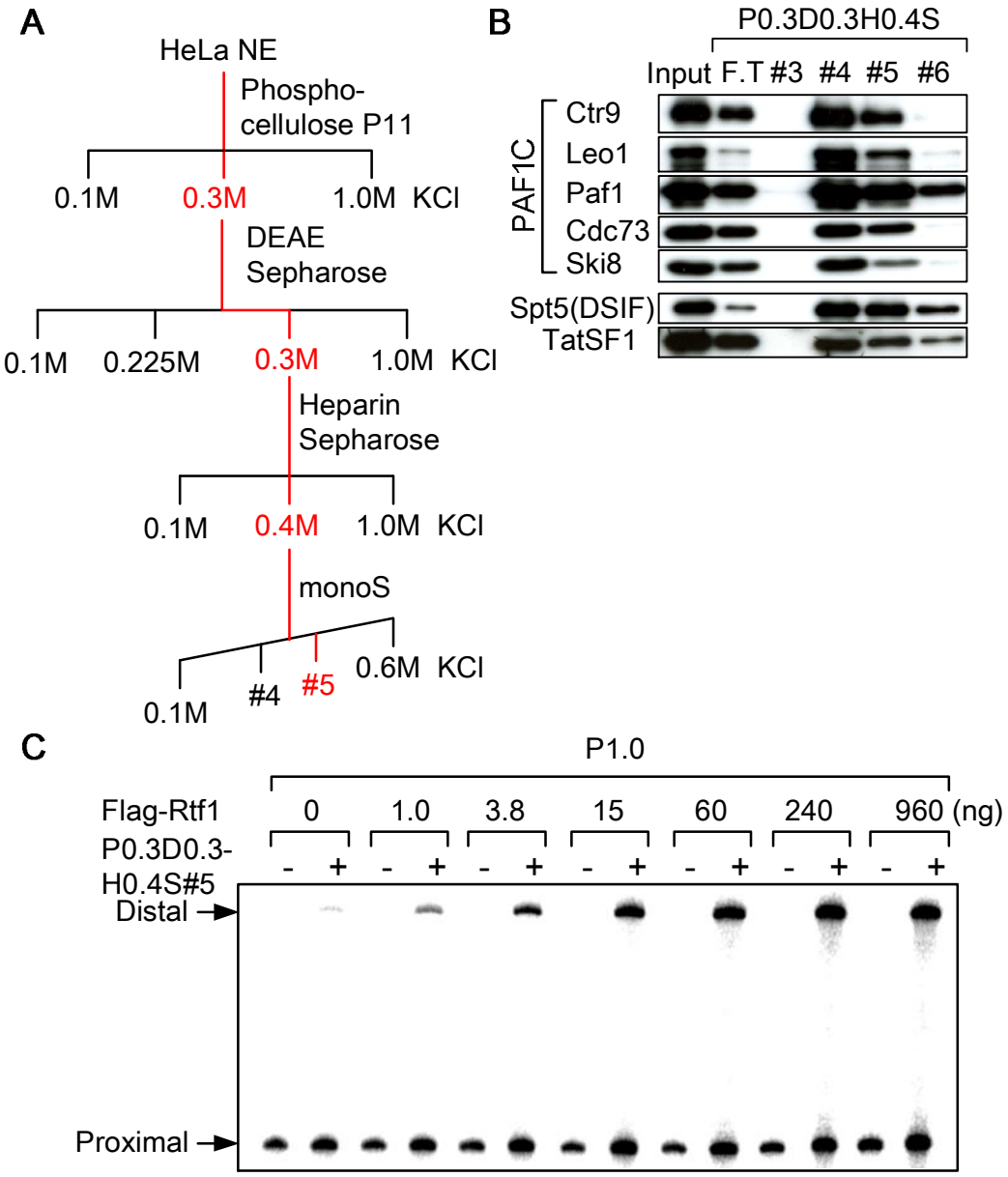

Figure 3-2. Flag-Rtf1 alone cannot promote transcription elongation.

(A) The separation scheme. (B) Immunoblot analysis of mono S column fractions derived from P0.3D0.3H0.4S. (C) A Rtf1 coactivator fraction P0.3D0.3H0.4S#5 was added together with increasing amounts of Flag-Rtf1 to *in vitro* transcription assays. Promoter-proximal and -distal products are indicated with arrows.

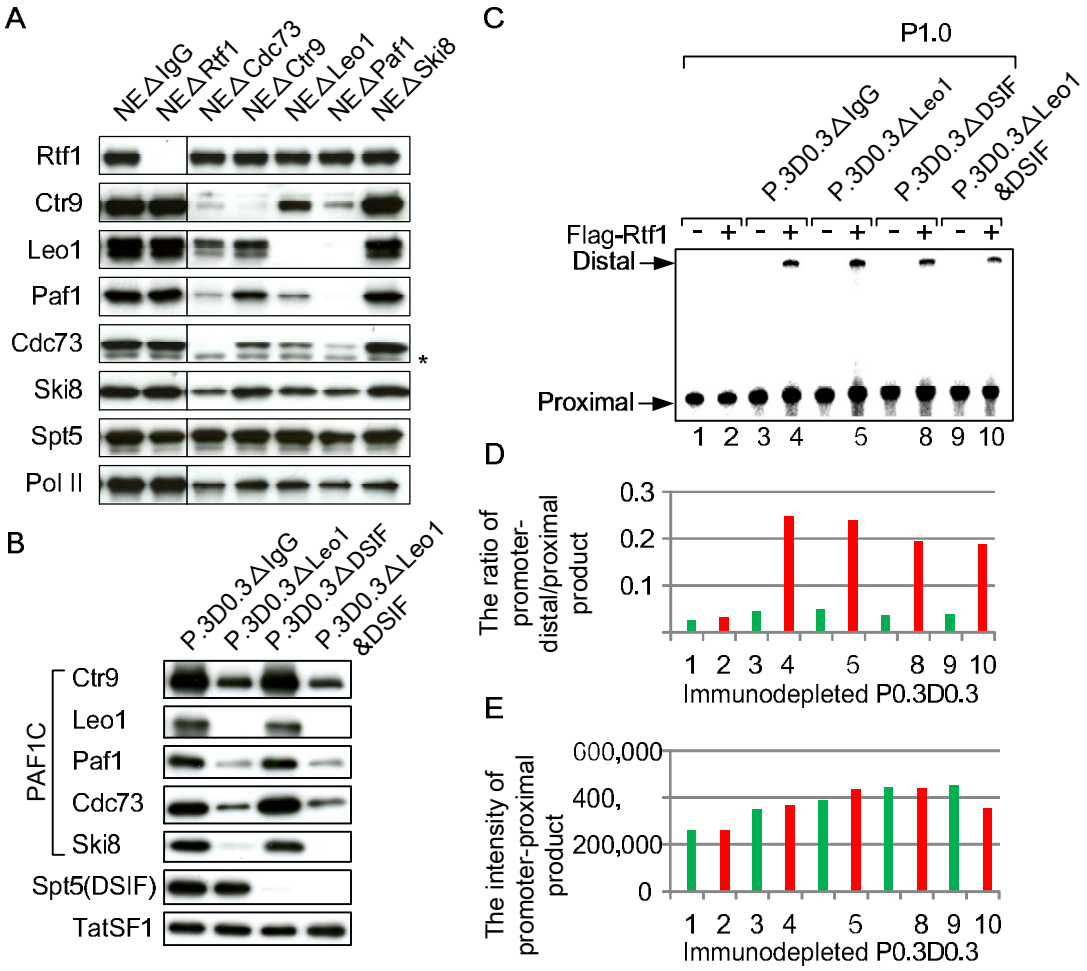

Figure 3-3. Human Rtf1 promotes transcription elongation independently of DSIF *in vitro*.

(A) Immunoblot analysis of HeLa NE after immunodepletion of one of the indicated factors. Mock-depleted NE (NEΔIgG) were analyzed as a control. An asterisk denotes a nonspecific signal. (B) Immunoblot analysis of the P0.3D0.3 fraction after immunodepletion of Leo1 and/or DSIF. Mock-depleted P0.3D0.3 (P0.3D0.3ΔIgG) was analyzed as a control. (C) *In vitro* transcription assays for immunodepleted P0.3D0.3 fractions. Promoter-proximal and -distal products are indicated with arrows.

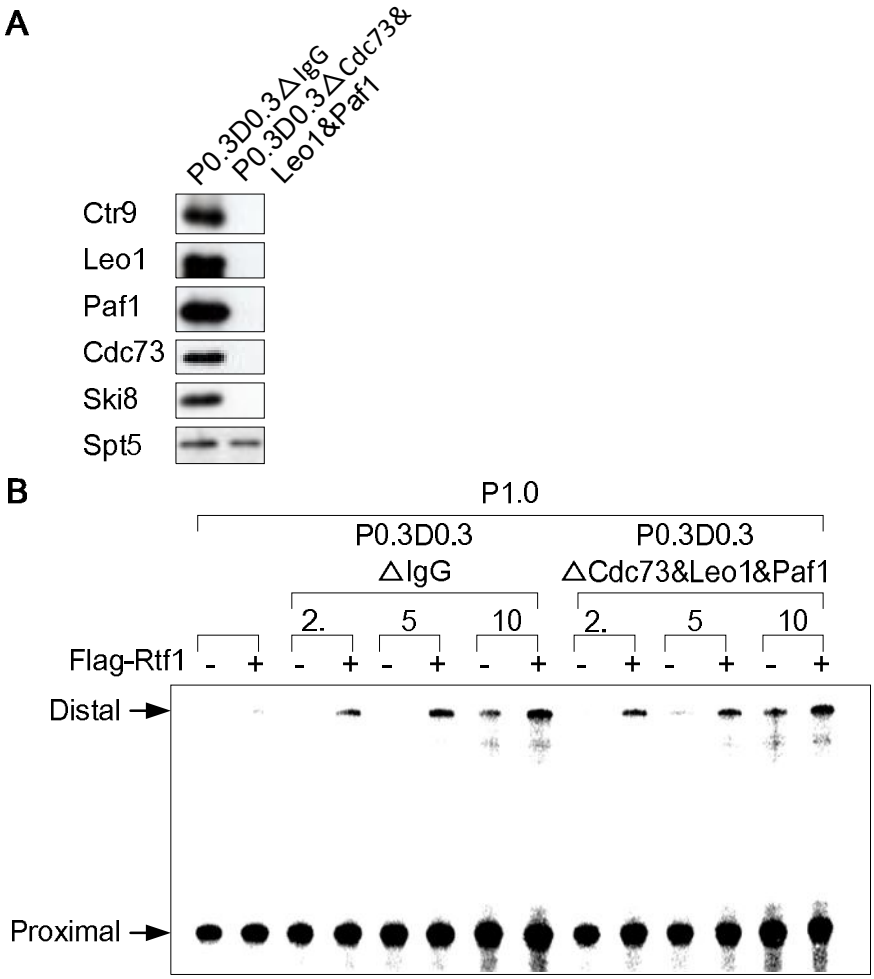

Figure 3-4. PAF1C is not required for Rtf1-mediated transcriptional activation.
(A) Immunoblot analysis of the P0.3D0.3 fractions immunodepleted with anti-Leo1, anti-Cdc73, and anti-Paf1 antibodies (P0.3D0.3ΔPAF1C) or with control IgG. (B) The indicated amounts of P0.3D0.3 fractions were added with or without Flag-Rtf1 to *in vitro* transcription assays.

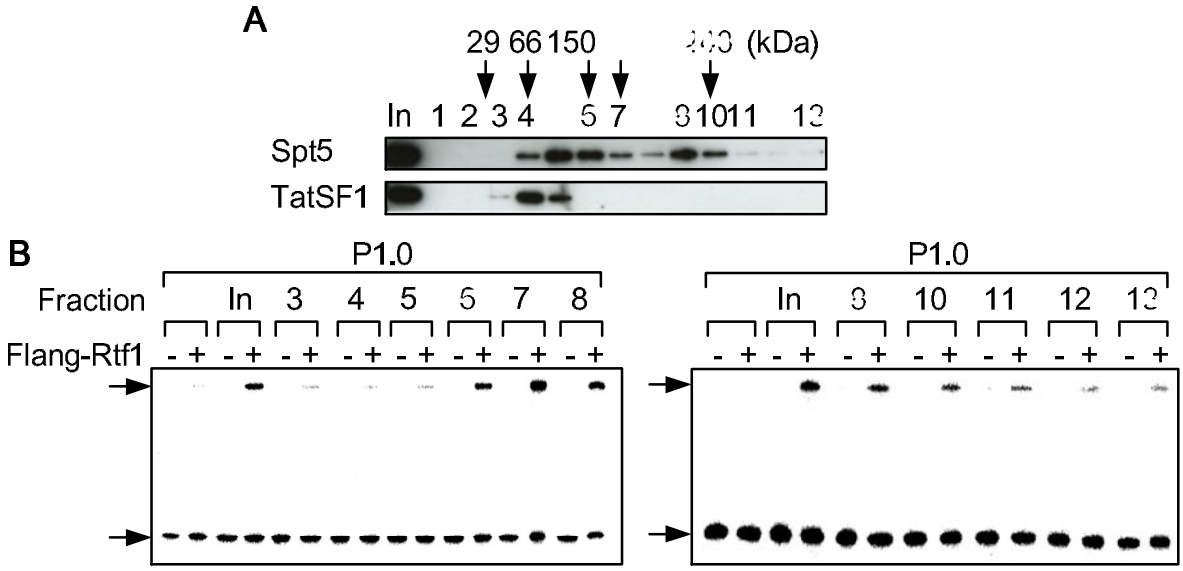

Figure 3-5. Tat-SF1PAF1C is not required for the Rtf1-mediated transcriptional activation elongation.

(A) Immunoblot analysis glycerol gradient sedimentation fractions of P0.3D0.3.; (B) In vitro transcription assays of the glycerol sedimentation fractions derived from P0.3D0.3. P0.3D0.3 (input) was analyzed for comparison.

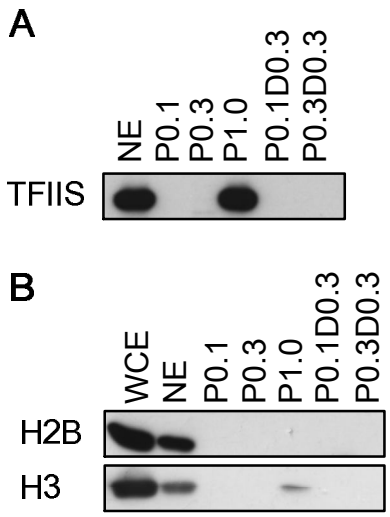

Figure 3-6. TFIIS and histones are not involved in Rtf1-meditated transcriptional activation.

(A) Immunoblot analysis of the NE, phosphocellulose P11 column fractions, and DEAE Sepharose column fractions of P0.1 and P0.3 with anti-TFIIS antibody. (B) Immunoblot analysis of the whole cell extract (WCE), NE, phosphocellulose P11 column fractions, and DEAE Sepharose column fractions of P0.1 and P0.3 with anti-H2B and anti-H3 antibodies.

CHAPTER 4: STRUCTURE-FUNCTION ANALYSIS OF HUMAN RTF1

4.1 Introduction

Post-transcriptional modification of histone proteins play important roles in the regulation of transcription. PAF1C is involved in the regulation of H2B monoubiquitination (H2Bub). In budding yeast, deletion of *RTF1*, *CTR9*, *PAF1*, or *CDC73* caused a decrease in the H2Bub level while deletion of *LEO1* did not (30, 34, 60, 62). Yeast Rtf1 plays facilitates H2Bub via its histone modification domain (HMD), an evolutionally conserved domain in eukaryotes (58). In contrast to the findings in yeast, Reinberg and colleagues showed that, humans, H2Bub established by the E2 ubiquitin-conjugating enzyme RNF20/40 and the E3 ubiquitin ligase UbcH6 is dependent on PAF1C lacking Rtf1 and the histone chaperone FACT *in vitro*. Once established, H2Bub cooperates with FACT to stimulate transcription elongation (39). In a subsequent study, Roeder and colleagues reconstituted human PAF1C using recombinant subunits to demonstrate that PAF1C activates the transcription of a chromatin template in cooperation with TFIIS *in vitro* (18). No difference was found between PAF1C and the PAF1C-Rtf1 complex in their chromatin transcription assays. Therefore, there is no compelling evidence that human Rtf1 regulates H2Bub.

In chapters 2 and 3, it was shown that human Rtf1 regulates transcription elongation directly. These results posed a few questions, such as whether human Rtf1 is required the H2Bub. If so, which domains are responsible for the functions of histone modification and direct regulation of transcription? Are these functions separable or is the same domain critical for both functions?

Rtf1 is evolutionally conserved in eukaryotes. the *S. cerevisiae* Rtf1 is a bona fide subunit of PAF1C and show strong interactions with Paf1 and Ctr9 (31). However, the interaction of Rtf1 with PAF1C is not stable in other species, such as *S. pombe*, *Drosophila*, and zebrafish (1, 23, 30). Also, Rtf1 was not detected in human PAF1C isolated by Zhu at al. (68), Rozenblatt-Rosen et al. (45), and Chu et al. (7). Only in Kim et al. (18), the human Rtf1-PAF1C complex was successfully reconstituted using the baculovirus coexpression system, in which the N-terminally deleted Rtf1 mutant was used. However, it is still unknown which domain of human Rtf1 is required for its interaction with PAF1C.

In this chapter, I performed structure-function analysis of human Rtf1 to understand the structural requirement for its coactivator-dependent function. In addition, I investigated whether the functional domains identified in *S. cerevisiae* are evolutionarily conserved. Finally, I investigated the physical interaction of human Rtf1 with PAF1C.

4.2 Results

4.2.1 Identification of the human Rtf1 domain essential for H2B monoubiquitination

Detailed structure-function analysis of *S. cerevisiae* Rtf1 has shown (i) that its histone modification domain (HMD) is required for H2B monoubiquitination and H3K4 and K79 methylation, (ii) that the Plus3 domain of Rtf1 is required for its interaction with the phosphorylated form of Spt5 and for the recruitment of other PAF1C subunits, and (iii) that the short C-terminal region of Rtf1 is required for its interaction with the other PAF1C subunits (58).

To determine domains required for the multiple functions of human Rtf1, six deletion mutants were constructed with a focus on the HMD, the Plus3 domain, and the C-terminal region (C-ter) (Fig. 4-1A). These constructs were designed for expression in mammalian and bacterial cells and contained an shRNA-resistant mutation to make knockdown–rescue experiments possible. Moreover, to avoid mislocalization of the deletion mutants, the nuclear localization signal from the SV40 large T-antigen was attached to the N-terminus of Rtf1. HeLa cells were sequentially infected with a lentiviral vector for shRNA targeting Rtf1 and then transfected with one of the Rtf1 deletion mutants. As expected, knockdown of Rtf1 significantly reduced its expression and the level of monoubiquitinated H2B (H2Bub) (Fig. 4-1B). Overexpression of full-length Rtf1 restored the H2Bub level. Similarly, Rtf1 Δ1, Δ5, and Δ6 restored the H2Bub level, whereas the other Rtf1 mutants did not. These results are consistent with previous findings in *S. cerevisiae* (58) and indicate the evolutionarily conserved role of the Rtf1 HMD in histone modifications.

4.2.2 Identification of the human Rtf1 domains important for its transcriptional activation potential

Next, Rtf1 deletion mutants were expressed in and purified from bacteria by tandem affinity purification using a histidine tag and a Flag tag (Fig. 4-2A), and the resulting recombinant proteins were examined by in vitro transcription assays using NEΔRtf1. As shown in Figure 4-2B, Rtf1 Δ1 and Δ2 fully restored elongation defects, whereas the other Rtf1 mutants did not, suggesting the possible involvement of the Plus3 domain and the C-terminal region in transcriptional activation. These findings prompted us to test two previously described Rtf1 point mutants in our assays (59). Single or double mutations of R366A and F441A, which reportedly disrupt the function of the Plus3 domain, were introduced into Rtf1 Δ1 (Fig. 4-3A). Rtf1 Δ1 and its two point mutants were also prepared by tandem affinity purification (Fig. 4-3A). The double point mutation severely impaired the transactivation potential of Rtf1 (Fig. 4-3B), indicating that the Plus3 domain is critical for the coactivator-dependent function.

4.2.3 Identification of the human Rtf1 domain important for its interaction with the Paf1 complex

To explore the physical interaction between human Rtf1 and PAF1C, Rtf1 mutants were absorbed onto anti-Flag agarose beads and then incubated with HeLa NE (Fig. 4-4A). Concordant with Kim et al. (18), Rtf1 Δ1 pulled down PAF1C from HeLa NE (Figs. 4-4B, C). Δ3 lacking the Plus3 domain pulled down PAF1C even more efficiently, but Δ5 did not. Therefore, despite differences in the binding affinity of Rtf1 to PAF1C between *S. cerevisiae* and humans, the same C-terminal region of Rtf1 is critical for its binding to PAF1C. Taken together, these results revealed different structural requirements for Rtf1 in H2B monoubiquitination, its coactivator-dependent function *in vitro*, and the physical interaction with PAF1C.

To evaluate the strength of physical interactions of Rtf1 with PAF1C, Flag-Paf1 and Flag-Rtf1 were overexpressed in HeLa cells and immunoprecipitated with anti-Flag antibody (Fig. 4-5A). Ctr9, Leo1, Cdc73, and Ski8 were efficiently coprecipitated with Flag-Paf1 whereas only a small amount of Ctr9 and Leo1 bound to Flag-Rtf1 (Fig. 4-5B). Moreover, the weak interaction was abolished by increasing the concentration of NaCl in wash buffer to 150 mM. These results showed that, although Rtf1 can interact with PAF1C, the strength is much weaker than the interaction between Paf1 and the remaining components of PAF1C. Together with the results of DEAE Sepharose column chromatography (Fig. 3-1D), it was confirmed that the interaction of Rtf1 with PAF1C is unstable.

4.3 Conclusions

Previous chapters showed that human Rtf1 regulates transcription elongation *in vitro* independently of PAF1C. Meanwhile, regulation of histone modifications is considered to be a major function of Rtf1 in many species. In this chapter, mutational analysis of human Rtf1 identified the domains required for the regulation of transcription elongation and for the physical interaction between human Rtf1 and PAF1C. The results obtained from the mutational analysis are summarized in Figure 4-6.

First, human Rtf1 is important for H2Bub in HeLa cells. The decreased level of H2Bub by the knockdown of Rtf1 showed the function of histone modification was also evolutionally conserved in the human.

Second, the HMD of human Rtf1 is essential for the regulation of H2Bub. The mutational analysis showed that at least the N-terminal 163 amino acids and the C-terminal 202 amino acids of human Rtf1 are dispensable for H2Bub.

Third, the Plus3 domain and the C-terminal region of human Rtf1 are important for its transcriptional activation potential *in vitro*. The HMD, on the other hand, is dispensable for Rtf1-mediated transcriptional activation. These results suggested the human Rtf1

regulates transcription elongation and histone modifications in mechanistically distinct manners through different domains.

Lastly, the C-terminal region is important for the physical interaction of human Rtf1 with PAF1C. Consistent with the results in *S. cerevisiae*, binding assays using deletion mutant of human Rtf1 showed that the C-terminal region plays an important role in its interaction with PAF1C. The comparison of the interactions of Flag-Rtf1 and Flag-Paf1 with the remaining components of PAF1C showed that the Rtf1–PAF1C interaction is much weaker than the Paf1–PAF1C interaction. It was suggested that most of the Rtf1 molecules in human cells are not present in a complex with PAF1C.

4.4 Materials and methods

4.4.1 shRNA-mediated knockdown and RNA analysis

Double-stranded oligonucleotides for shRNAs against Rtf1 and Cdc73 were cloned into pBluescript-U6 as previously described (63). After functional validation, cassettes including a mouse U6 promoter were excised and subcloned into pLenti6 (Life Technologies). shRNAs against Ctr9, Leo1, Paf1, and Ski8 were directly cloned into the lentiviral vector pRSI9. Recombinant lentiviruses were produced and concentrated prior to use according to standard procedures. In knockdown experiments, HeLa cells were infected with recombinant lentivirus expressing no shRNA or expressing one of the shRNAs and selected in the presence of 5 μg/ml Blasticidin for pLenti6 or 1 μg/ml Puromycin for pRSI9. Ski8 knockdown cells were harvested 4 days post-infection, and all the other cells were harvested 7 days post-infection. Cells were lysed with high-salt buffer (50 mM Tris-HCl, pH 7.9, 500 mM NaCl, 1% NP-40) for immunoblotting or with Sepasol RNA I Super G (Nacalai Tesque) for RNA analyses. The following 21-nucleotide sequences were used as shRNA targets.

Rtf1 #2: 5′-AAGAAUUGAAUCGGGUUCGAU-3′;
Ctr9 #3: 5′-AAGCAGAAGCGGAACAUGAUG-3′;
Leo1 #4: 5′-AAGAGGCAGUGAUAGUGAAGA-3′;
Paf1 #5: 5′-AACCAGUUUGUGGCCUAUUUC-3′;
Cdc73 #1: 5′-AAGUAUAGACAGAAGCGCUCC-3′;
Ski8 #3: 5′-AGUGGAGCCAUAGAUGGAAUC-3′.

4.4.2 Immunoprecipitation

Immunoprecipitations were performed as previously described (32, 64). Flag-Paf1 and Flga-Rtf1 expression plasmids or an empty vector were transiently transfected into HeLa cells. Cells were harvested 3 days post-transfection. Lysates were prepared in a buffer containing 100 mM NaCl, 50 mM Tris-HCl (pH8.0), and 0.1% of NP-40 and incubated with Flag antibody-immobilized agarose beads at 4°C for 2 h. Beads were washed 6 times in a

buffer containing 50 mM Tris-HCl (pH8.0), 0.1% of NP-40, and 100 to 150 mM NaCl. The proteins bound to beads were eluted by boiling with 2x loading dye.

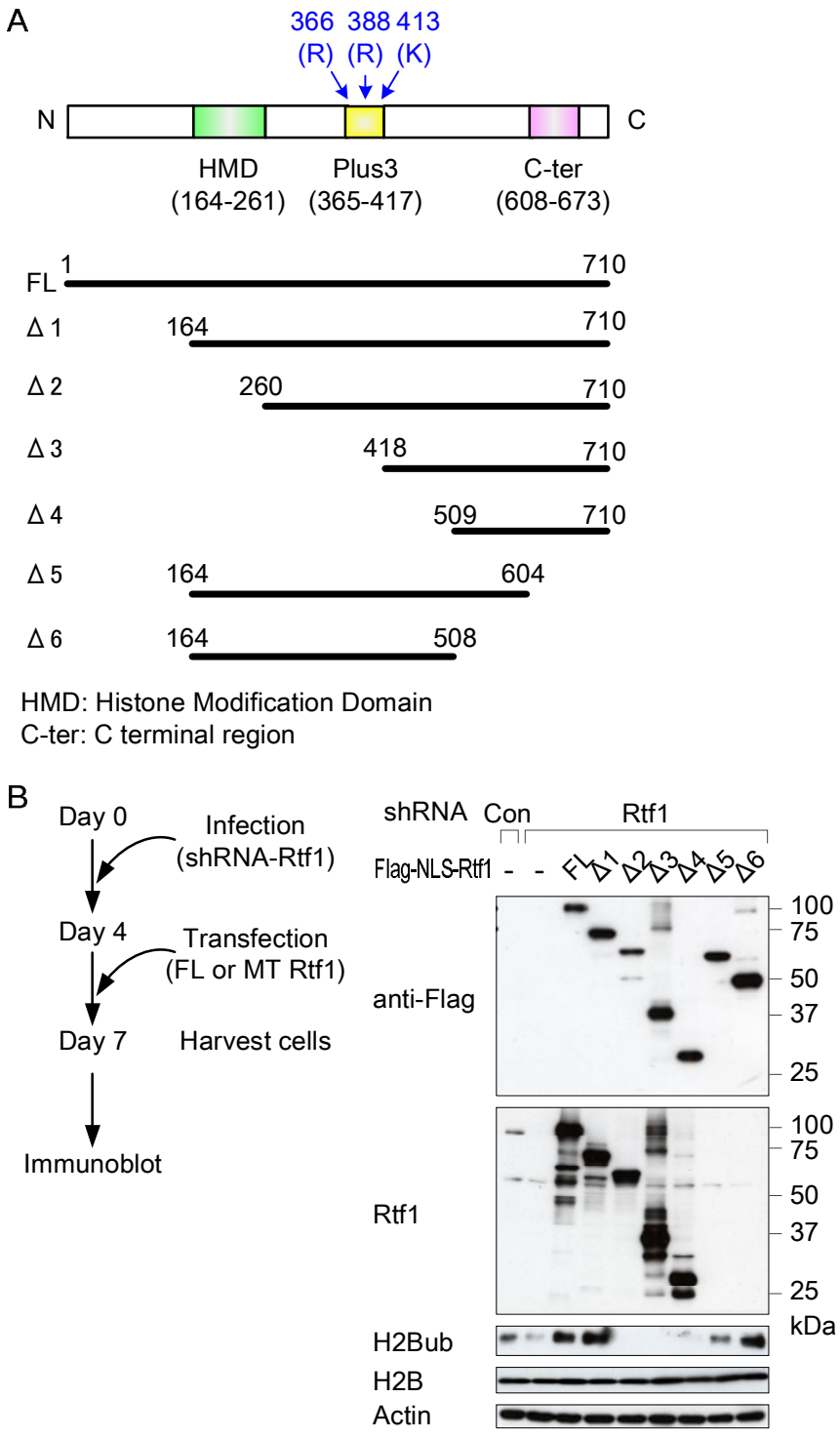

Figure 4-1. The HMD is responsible for H2B monoubiquitination in human cells.
(A) Schematic structures of human Rtf1 deletion mutants. (B) Knockdown–rescue experiments. HeLa cells were sequentially infected with a lentiviral vector for shRNA targeting Rtf1 and transfected with one of the Flag-NLS-Rtf1 expression vectors carrying shRNA-resistant mutations. Cells were harvested 7 days post-infection (3 days post-transfection) and subjected to immunoblot analysis.

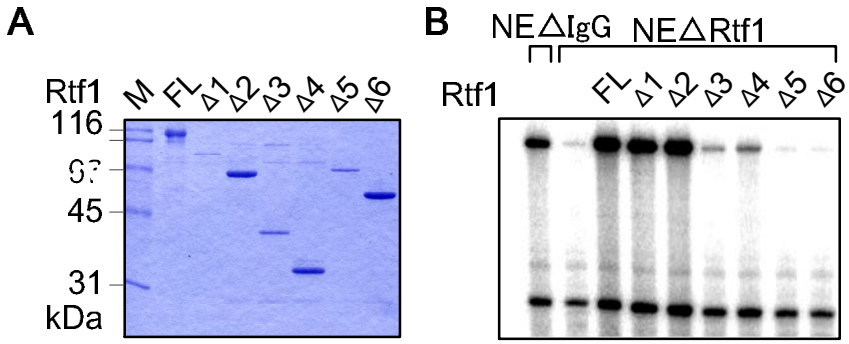

Figure 4-2. The Plus3 domain and the C terminal region are important for the transcriptional activation potential of human Rtf1.

(A) Purity of full-length (FL) Flag-Rtf1 prepared from insect cells and His-Rtf1-Flag deletion mutants prepared from bacteria was examined by Coomassie blue staining. (B) The transactivation potential of Rtf1 mutants was examined by *in vitro* transcription assays using NEΔRtf1.

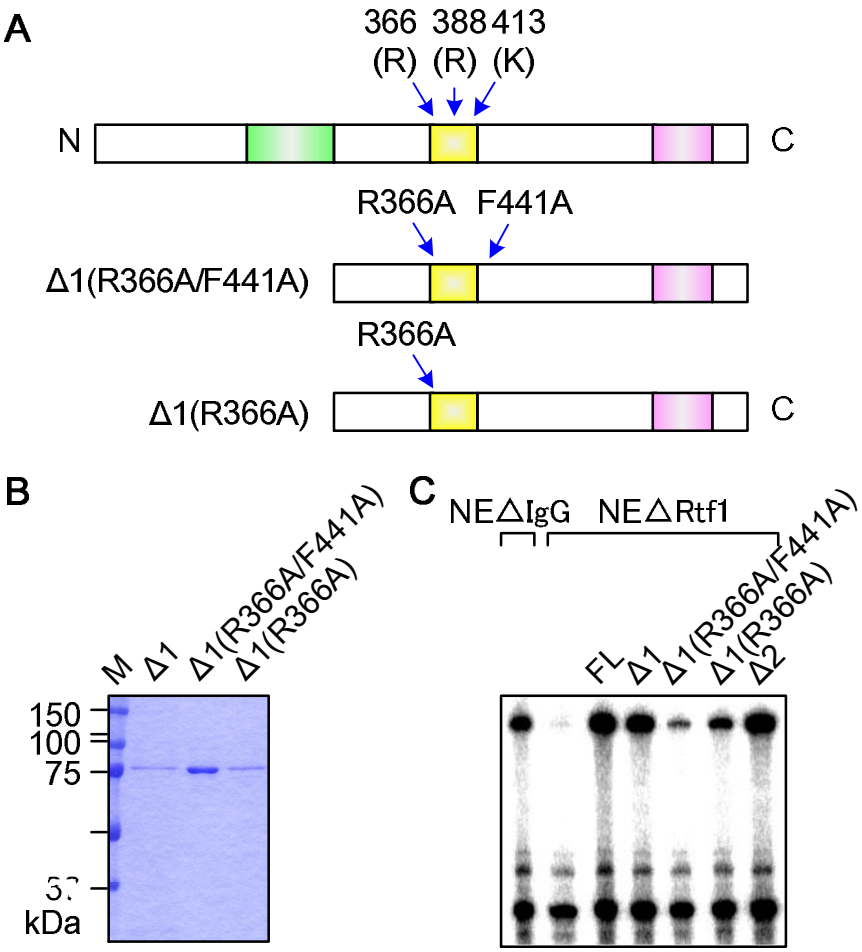

Figure 4-3. The Plus3 domain of human Rtf1 is important for promoting transcription elongation *in vitro*.

(A) Schematic structures of human Rtf1 point mutants. (B) Purity of His-Rtf1-Flag proteins prepared from bacteria was examined by Coomassie blue staining. (C) The transactivation potential of Rtf1 mutants was examined by *in vitro* transcription assays using NEΔRtf1.

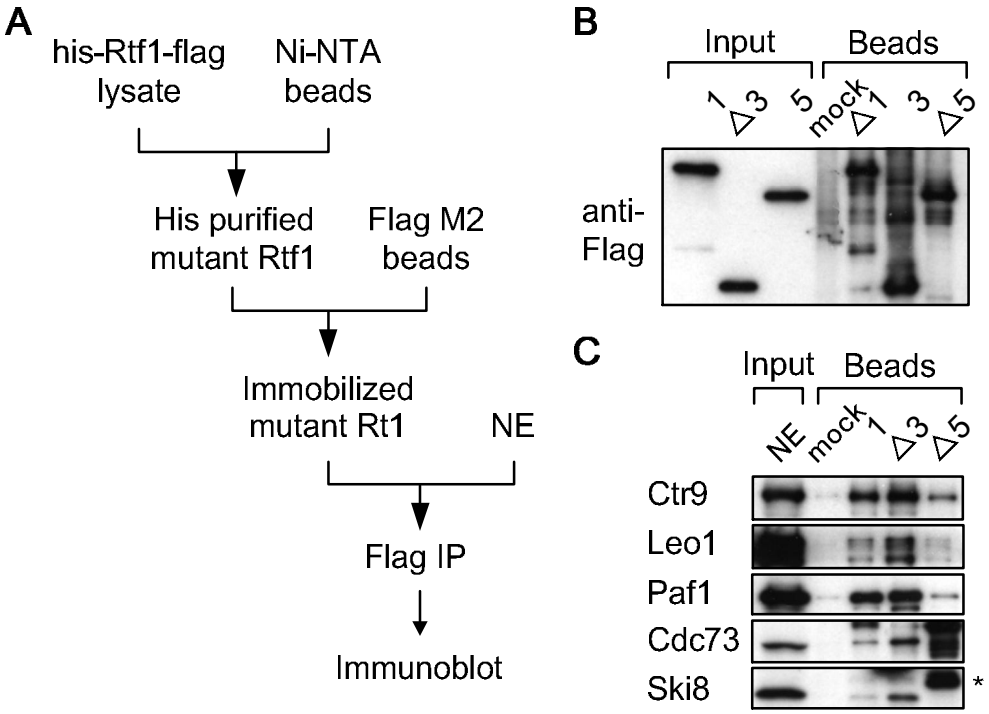

Figure 4-4. Identification of the PAF1C-interacting domain of human Rtf1.
(A) Scheme of immunoprecipitation. (B, C) Input and bound fractions were analyzed by immunoblotting. Asterisks denote nonspecific signals.

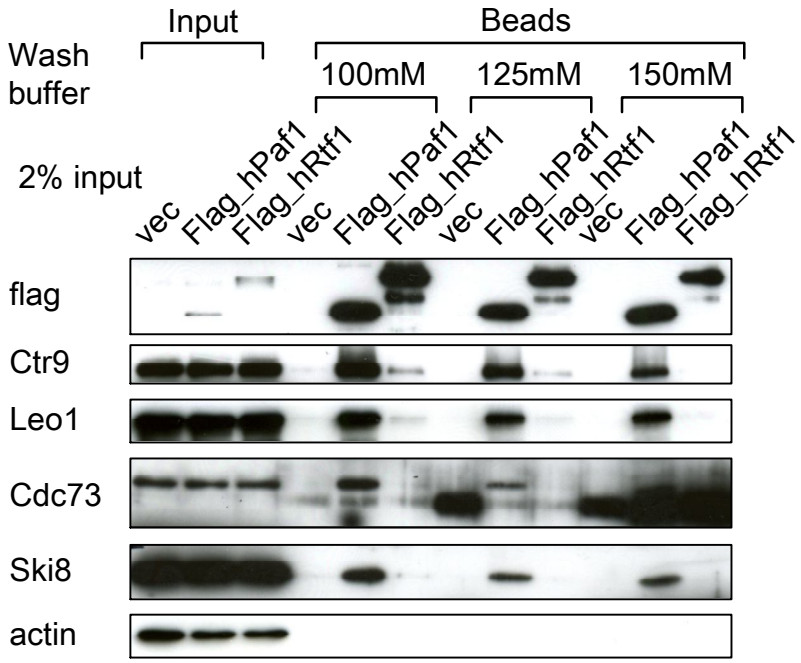

Figure 4-5. Comparison of the interactions of human Rtf1 and Paf1 with the remaining components of PAF1C.

HeLa cells were transfected with the indicated expression vectors and harvested 3 days post-transfection for immunoprecipitation with anti-Flag agarose beads. During the washing step, wash buffers containing 100mM, 125mM, and 150mM of NaCl were used as indicated. The input and bound proteins were analyzed by the immunoblotting.

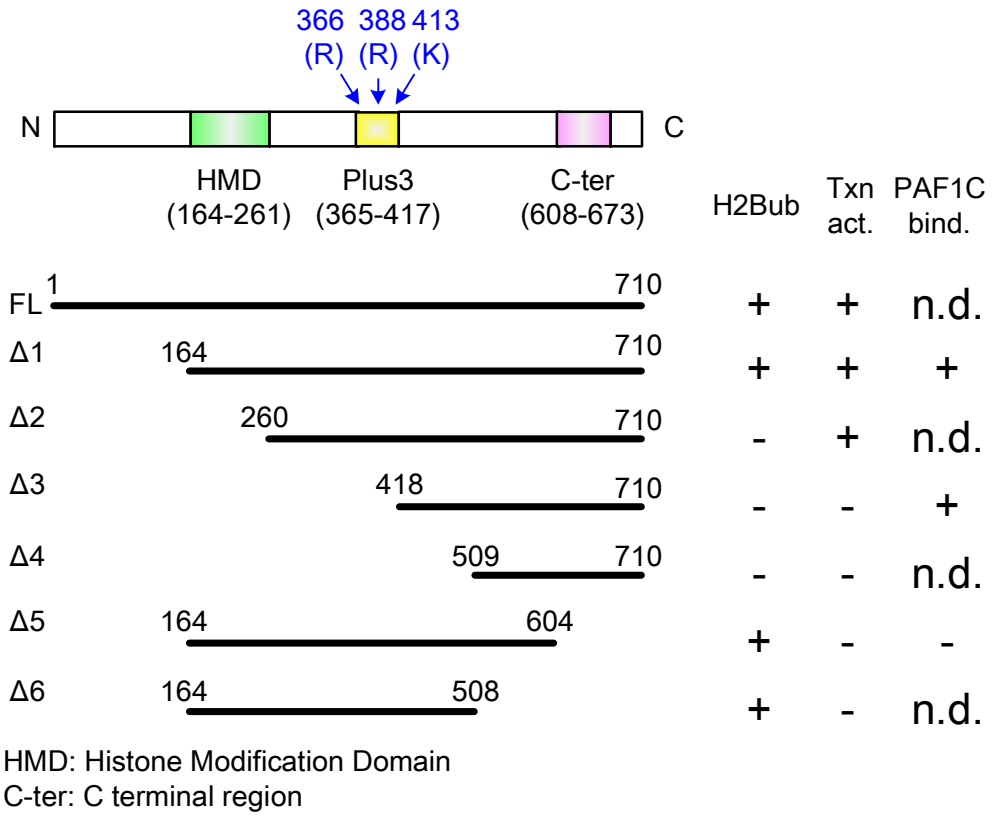

Figure 4-6. The summary of the structural analysis of human Rtf1.

Schematic structures of human Rtf1 deletion mutants. The results obtained from chapter 4 were summarized to the right. Txn act., transcriptional activation; PAF1C bind., PAF1C binding; n.d., not determined.

CHAPTER 5: NON-OVERLAPPING FUNCTIONS OF RTF1 AND THE PAF1 COMPLEX IN HUMAN HELA CELLS

5.1 Introduction

In budding yeast, deletion of *RTF1* and *LEO1* did not affect cell growth whereas growth defect was found in *paf1*Δ, *ctr9*Δ, and *cdc73*Δ strains (52). Unlike the findings in yeast, inhibition of Rtf1 in other species caused various developmental defects. In *Drosophila* and zebrafish, for example, inhibition of Rtf1 affected the Notch signaling pathway. A defect in wing development was caused by the knockdown of *Drosophila* Rtf1 (11). Deletion of *Rtf1* or *Ctr9* homologs in zebrafish caused a disruption of neural crest cells and a defect in somitogenesis (2). These results showed that the physiological role of Rtf1 varies among species. In humans, there is no report on the physiological role of Rtf1.

Rtf1 plays a key role in the regulation of H2B monoubiquitination in budding yeast and humans (30, 62, and chapter 4). Similarly, the components of yeast PAF1C are required for H2Bub in yeast except for Leo1 (30, 62), and human Cdc73 is also required for H2Bub (5). Thus, the functions of Rtf1 and PAF1C in histone modifications seem to be conserved between budding yeast and humans.

As transcription factors, both Rtf1 and PAF1C seem to be important for transcriptional regulation of cellular genes. However, deletion of *PAF1* or *CTR9* in yeast influenced expression of only a small subset of genes (40). It will be interesting to investigate how many genes and what kind of genes are affected by the knockdown of human Rtf1 or PAF1C.

The recruitment of transcription factors to target genes has a close relationship with transcriptional regulation. In *S. cerevisiae*, Rtf1 is considered as a core subunit of PAF1C that recruits the other subunits to target genes via its interaction with DSIF (3, 27, 29, 31, 67). In humans, however, Rtf1 interacts with PAF1C very weakly (chapter 4). It is not known whether PAF1C is recruited to target genes in a manner dependent on Rtf1 in human cells.

In this chapter, the roles of Rtf1 and PAF1C in human HeLa cells were studied by knocking down Rtf1 and PAF1C individually. I examined whether knockdown of these factors has any effect on cell growth, gene expression, and the recruitment of PAF1C to target genes.

5.2 Results

5.2.1 Influence of knockdown of human Rtf1 or the Paf1 complex on cell growth

To examine the effect of knockdown of Rtf1 and PAF1C on cell growth, HeLa cells

were infected by the lentivirus expressing shRNAs targeting Rtf1, Paf1, and Ski8. Scramble sequence under the U6 promoter was used as a control of shRNA targeting Rtf1. The U6 promoter alone was used as a control of shRNAs targeting Paf1 and Ski8. Transduction of these control vectors did not affect cell growth whereas cell growth was retarded to various degrees when Ski8, Rtf1, or Paf1 was knocked down. The most severe growth defect was found in Ski8 knockdown cells. This might be because Ski8 knockdown inhibited the functions of both PAF1C and the SKI complex as Ski8 is a component of these complexes. Unlike the findings in budding yeast, knockdown of Rtf1 showed a more severe defect in cell growth than Paf1 knockdown (Figs. 5-1 and 5-2). The knockdown phenotypes of Ctr9, Cdc73, and Leo1 were also examined. Knockdown of Ctr9 caused a severe growth defect whereas knockdown of Leo1 or Cdc73 did not affect cell growth. These results suggested that Rtf1 might play a more important role in humans than in budding yeast.

To analyze knockdown phenotypes in more detail, the level of H2B monoubiquitination was examined (Fig. 5-2). The level of H2Bub was significantly reduced by the knockdown of Rtf1, Ctr9, Paf1, and Cdc73, but not by the knockdown of Leo1 and Ski8. Coincidentally, yeast Leo1 was also shown to be dispensable for H2Bub (62). These results showed that, in agreement with the findings in yeast, human Rtf1 and PAF1C is both critical for H2B monoubiquitination.

Immunoblot analysis showed that knockdown of Rtf1 or PAF1C subunits resulted in codepletion of nontarget proteins in certain cases (Fig. 5-2). For example, Paf1 knockdown resulted in a reduction of all PAF1C subunits to varying degrees. This may be due to a destabilization of the protein complex upon depletion of a single component.

5.2.2 Identification of genes controlled by Rtf1 and the Paf1 complex in HeLa cells

To investigate the genes regulated by human Rtf1 or PAF1C, RNA-seq was performed after knockdown of Rtf1, Paf1, and Ski8 in triplicate (Figs. 5-3A, B). Paf1 and Ski8 were selected as representative subunits of PAF1C. Since knockdown of any one of Rtf1, Ski8, and Paf1 affected cell growth upon prolonged culture (data not shown), I carefully determined the time course to complete the experiments before the onset of a significant growth retardation. For this reason, total RNA was harvested from Ski8 knockdown cells at day 4 and from Rtf1 or Paf1 knockdown cells at day 7. Using false discovery rate (FDR) of 0.05 and fold-change of 2.0 as cut-off values, 701, 1,039, and 1,556 genes were identified as significantly affected by Paf1, Ski8, and Rtf1 knockdown (Table 5-1). The number of genes downregulated by the knockdown was slightly larger than the number of upregulated genes (Fig. 5-3B). In both cases, there was a significant overlap among the genes affected by the knockdown, suggesting a functional similarity between Rtf1 and PAF1C. In addition, many genes were uniquely affected by Rtf1 knockdown (402 upregulated genes and 495 downregulated genes).

To confirm the results of RNA-seq, quantitative (q) RT-PCR analysis was performed

(Fig. 5-4). For all the 18 genes tested, qRT-PCR reproduced the results of RNA-seq. For example, *LYPD3* was strongly induced only by Rtf1 knockdown, whereas *BTBD3* was selectively repressed by Rtf1 knockdown. By contrast, *NT5E* and *OLFM1* were up- and down-regulated, respectively, by Paf1, Ski8, and Rtf1 knockdown to a similar extent. These results clearly showed that the expression of certain genes is more sensitive to Rtf1 inhibition than to PAF1C inhibition. The above findings also suggest that a significant number of genes are in fact downregulated by Rtf1 and PAF1C singly or in combination.

To gain insight into the biological significance of the differential gene regulation by Rtf1 and PAF1C, Gene Ontology (GO) analysis was performed (Table 5-2). Among the genes commonly affected by Paf1, Ski8, and Rtf1 knockdown, genes involved in nucleosome assembly and cell proliferation were highly enriched. On the other hand, genes involved in mitotic cell cycle were enriched among the genes uniquely affected by Rtf1 knockdown, suggesting that functionally distinct sets of genes are regulated by Rtf1 alone and by the combination of Rtf1 and PAF1C.

The above results prompted us to perform cell cycle analysis of knockdown cells. At the time point when RNA-seq analysis was performed, Rtf1 knockdown increased a cell population in G1 phase, whereas Paf1 and Ski8 knockdown had only a weak, if any, effect on cell cycle (Fig. 5-5). Thus, transcriptional changes caused by Rtf1 knockdown were found to be correlated with the phenotypic changes observed.

shRNA-mediated knockdown of Rtf1, Paf1, and Ski8 all seemed to deplete Rtf1 levels to similar extents (Fig. 5-2), making interpretation of the RNA-seq data complicated. As an attempt to differentiate primary and secondary effects of knockdown, we performed time-course analysis, based on the assumption that secondary effects should be observed with delayed kinetics (Figs. 5-6 and 5-7). Expression of “Rtf1-specific” and “common” genes was negligibly affected on day 5 of Rtf1 knockdown but was activated (*SERPINE1* and *NT5E*) or repressed (*BTBD3* and *ASS1*) on the next day (Fig. 5-7). Concordantly, there was no discernible effect on the protein levels of PAF1C components and Rtf1 on day 5 (Fig. 5-6). On the next day, however, the Rtf1 protein almost disappeared, and the levels of the PAF1C components also decreased to varying degrees. Expression of “common” genes was up- or down-regulated significantly on day 5 of Paf1 knockdown and on day 3 of Ski8 knockdown (Fig. 5-7). On day 5 of Paf1 knockdown, the levels of PAF1C components decreased to varying degrees with the Paf1 level affected most strongly, whereas there was no apparent reduction of the Rtf1 level (Fig. 5-6). On day 3 of Ski8 knockdown, the Ski8 level decreased significantly with only a small effect on the levels of the other PAF1C components, whereas there was no apparent reduction of the Rtf1 level. On the next day, the Ski8 level decreased further, and the levels of the other PAF1C components and Rtf1 also decreased. Collectively, transcriptional defects were observed at the earliest time point when shRNA-mediated knockdown was evident, and the time course of altered gene expression seemed to be correlated well with the time course of knockdown, suggesting that at least

several genes studied here are direct targets of Rtf1, Ski8, and Paf1. Moreover, the above findings suggest that codepletion of Rtf1 is not responsible, at least in part, for the knockdown phenotype observed following Paf1 or Ski8 knockdown.

5.2.3 Human Rtf1-independent recruitment of the Paf1 complex

To investigate the mechanisms underlying the differential gene regulation by Rtf1 and PAF1C, the occupancy of Rtf1 and Paf1 at several gene loci identified in this study was compared. Contrary to my expectation that Rtf1 was more enriched than Paf1 at "Rtf1-specific" genes such as *LYPD3* and *METTL7A*, no clear differences in the Rtf1:Paf1 ratio were observed (Fig. 5-8 and data not shown). Instead, Rtf1 and Paf1 levels were correlated with the level of Pol II over the several genes examined, although Rtf1 and Paf1 were apparently more enriched at the gene body than Pol II. These results suggested that the different outcomes of Rtf1 and PAF1C knockdown arise from the post-recruitment process, i.e., after these factors are recruited to target genes.

The functional differences between human Rtf1 and PAF1C led us to investigate the mechanisms by which these factors find their target genes. In *S. cerevisiae*, Rtf1 binds to Spt5 and serves as a binding platform for PAF1C (27, 29, 67), and Cdc73 is required for the efficient recruitment of Rtf1 (3, 31). Considering the weak physical interaction between Rtf1 and PAF1C in metazoans, however, whether such a reciprocal recruitment of Rtf1 and PAF1C is conserved across species is questionable. To address this issue, I selected three genes that gave strong ChIP signals and examined the distribution of Pol II, Paf1, and Rtf1 in response to Rtf1 knockdown (Fig. 5-8). Compared to the Rtf1 signal, which was significantly reduced, Paf1 occupancy on *DUSP1* and *PLAUR* was negligibly affected by Rtf1 knockdown. *FAM43A* expression was highly sensitive to Rtf1 knockdown (Fig. 5-8). Concordantly, Pol II and Paf1 occupancy on *FAM43A* was strongly affected by Rtf1 knockdown, whereas the Paf1:Pol II ratio was only modestly affected. These results support the idea that human PAF1C is recruited to target genes independently of Rtf1.

5.3 Conclusions

Both of Rtf1 and PAF1C are multifunctional proteins that share several common functions. A different function of human Rtf1 against PAF1C was found *in vitro* in chapters 2 to 4. In chapter 5, the roles of human Rtf1 and PAF1C in transcriptional regulation in living cells were investigated. A possible causal relationship of human Rtf1 and Paf1 in their associations with target genes was also studied.

First, human Rtf1 is important for growth in HeLa cells. Unlike the case in budding yeast, knockdown of Rtf1 caused a severe growth defect in HeLa cells. It is plausible that human Rtf1 plays a more important role in humans than in yeast.

Next, PAF1C is required for H2B monoubiquitination. Consistent with the findings in yeast, depletion of Paf1, Ctr9, or Cdc73 decreased the level of H2Bub whereas depletion

of Leo1 and Ski8 did not.

Third, human Rtf1 and PAF1C regulate partially overlapping sets of genes, respectively, in HeLa cells. More than half of the genes identified are specifically regulated by Rtf1 or PAF1C. Only a small number of genes were found to be specifically affected by the knockdown of Paf1 whereas a larger number of genes were found affected by the knockdown of Rtf1.

Lastly, human PAF1C can be recruited to target genes in a manner independent of Rtf1. This finding is in contrast to the finding in yeast and may be due to the weak physical interaction between Rtf1 and PAF1C in humans. Also, Rtf1 and Paf1 are associated with a few genes examined regardless of whether knockdown of respective factors affects their expression or not.

5.4 Materials and methods

5.4.1 RNA analyses

Total RNAs were prepared from control HeLa cells and Rtf1, Ski8, and Paf1 knockdown cells and subjected to RNA-seq in triplicate. Following the removal of rRNA using the RiboMinus Eukaryote Kit (Life Technologies), libraries were constructed using the Ion Total RNA-Seq Kit and sequenced using the Ion Proton System (Life Technologies). A minimum of 3 million high-quality reads were obtained for each sample. The reads were mapped to the human genome GRch37.p9 using CLC Genomics Workbench, and RPKMs were calculated. The rank product method (RankProd) was used to identify differentially expressed genes using a FDR of 0.05 and fold-change of 2.0 as cut-off values. The average Z scores were used to draw a heat map for the genes differentially expressed in any one of Rtf, Ski8, and Paf1 knockdowns. GO analysis was performed using DAVID Bioinformatics Resources 6.7 (12).

5.4.2 qRT-PCR

Total RNA was prepared using Sepasol-RNA I Super G kit. cDNA was prepared using Reverse transcriptase Superscript III and the purified total RNA. The amount of cDNA was measured by qRT-PCR using the KAPA SYBR FAST qPCR Kit (Takara). Quantification was performed with the ΔΔCt method using *GADPH* as a reference gene.

5.4.3 Fluorescence-activated cell sorting

Control and knockdown cells were prepared in triplicate and fixed with 70% cold ethanol in phosphate buffered saline. After RNase A treatment, fixed cells were stained with 50 μg/ml propidium iodide. The DNA content measurement and cell cycle analysis were performed using a FACSCalibur flow cytometer and the Cell Quest software (Becton Dickinson).

5.4.4 Chromatin immunoprecipitation

ChIP assays were performed as described previously (65), with some modification. Briefly, HeLa cells were crosslinked with 1% formaldehyde for 5 min. Nuclei were isolated and resuspended in micrococcal nuclease reaction buffer (20 mM Tris–HCl, pH 8.0, 5 mM NaCl, 2.5 mM $MgCl_2$, protease inhibitor cocktail (Nacalai Tesque)). Chromatin was sheared with micrococcal nuclease (Takara) for 10 min at 37ºC, and the reaction was stopped by the addition of SDS to a final concentration of 0.2%, followed by dilution with an equal volume of dilution buffer (10 mM Tris-HCl, pH8.0, 195 mM NaCl, 2 mM EDTA, 1 mM EGTA, protease inhibitor cocktail). Chromatin was then solubilized by sonication using a Misonix S-4000 sonicator (Misonix). Soluble chromatin samples were incubated overnight at 4°C with antibodies described below. Immunocomplexes were captured with Dynabeads Protein G (Dynal). Beads were sequentially washed three times with RIPA buffer (10 mM Tris–HCl, pH 8.0, 150 mM NaCl, 1 mM EDTA, 0.5 mM EGTA, 0.1% SDS, 1% Triton X-100, 0.1% sodium deoxycholate), three times with high-salt ChIP wash buffer (10 mM Tris–HCl, pH 8.0, 500 mM NaCl, 1 mM EDTA, 0.5 mM EGTA, 0.1% SDS, 1% Triton X-100, 0.1% sodium deoxycholate), twice with LiCl ChIP wash buffer (10 mM Tris–HCl, pH 8.0, 250 mM LiCl, 1 mM EDTA, 1 mM EGTA, 1% NP-40, 1% sodium deoxycholate), and twice with TE (10 mM Tris–HCl, pH 8.0, 1 mM EDTA). Immunoprecipitates were eluted with elution buffer (50 mM Tris–HCl, pH 8.0, 10 mM EDTA, 1% SDS, 10 mM DTT) and incubated overnight at 65°C. Genomic DNA fragments in inputs and eluates were purified and analyzed using qPCR. The antibodies used for the assays were as follows: 0.5 µl of anti-Rpb4 sera (a gift from Koji Hisatake, University of Tsukuba; rabbit polyclonal), 1 µg of anti-Paf1 (ab20662, Abcam), and 1 µg of anti-Rtf1 (A300-179A, Bethyl).

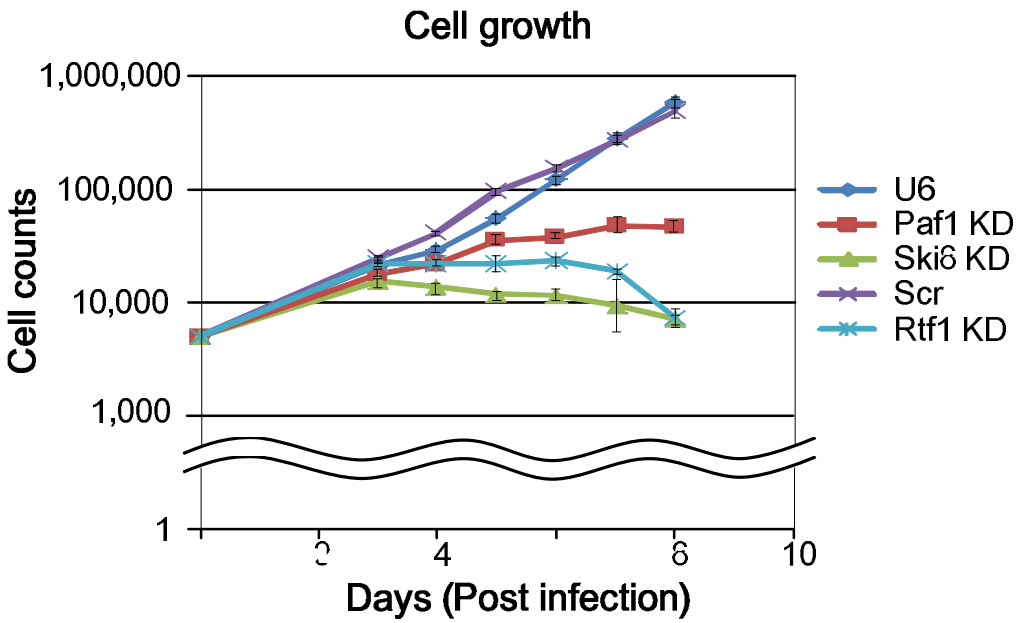

Figure 5-1. Influence of knockdown of human Rtf1 and PAF1C on cell growth.
HeLa cells were infected with lentiviral vectors expressing shRNAs targeting Rtf1, Paf1, and Ski8. U6 and Scr denote control lentiviral vectors. An appropriate antibiotics was added 1 day post-infection. Counting the cell number was started from 3 days post-infection.

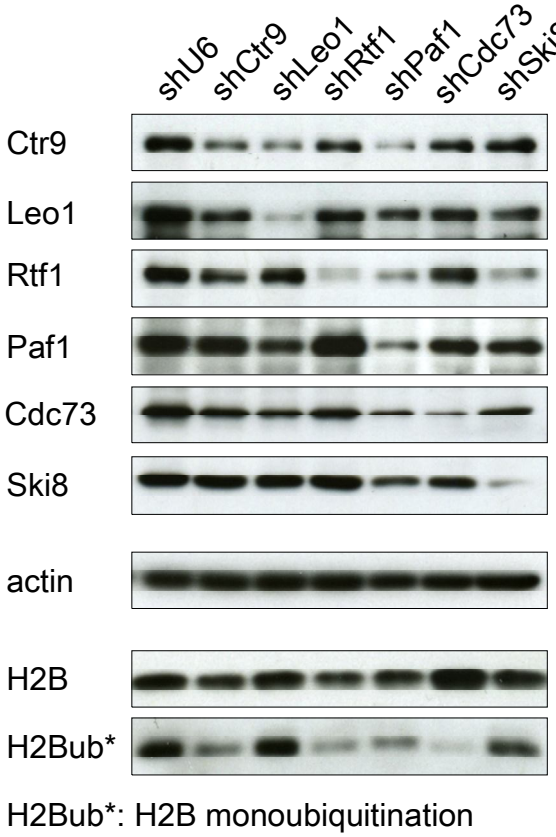

Figure 5-2. Influence of Rtf1 and PAF1C knockdown on H2B monoubiquitination in HeLa cells.

(A) HeLa cells were infected with lentiviral vectors expressing shRNAs targeting the indicated factors. Four or seven days post-infection, the cells were harvested for immunoblot analysis.

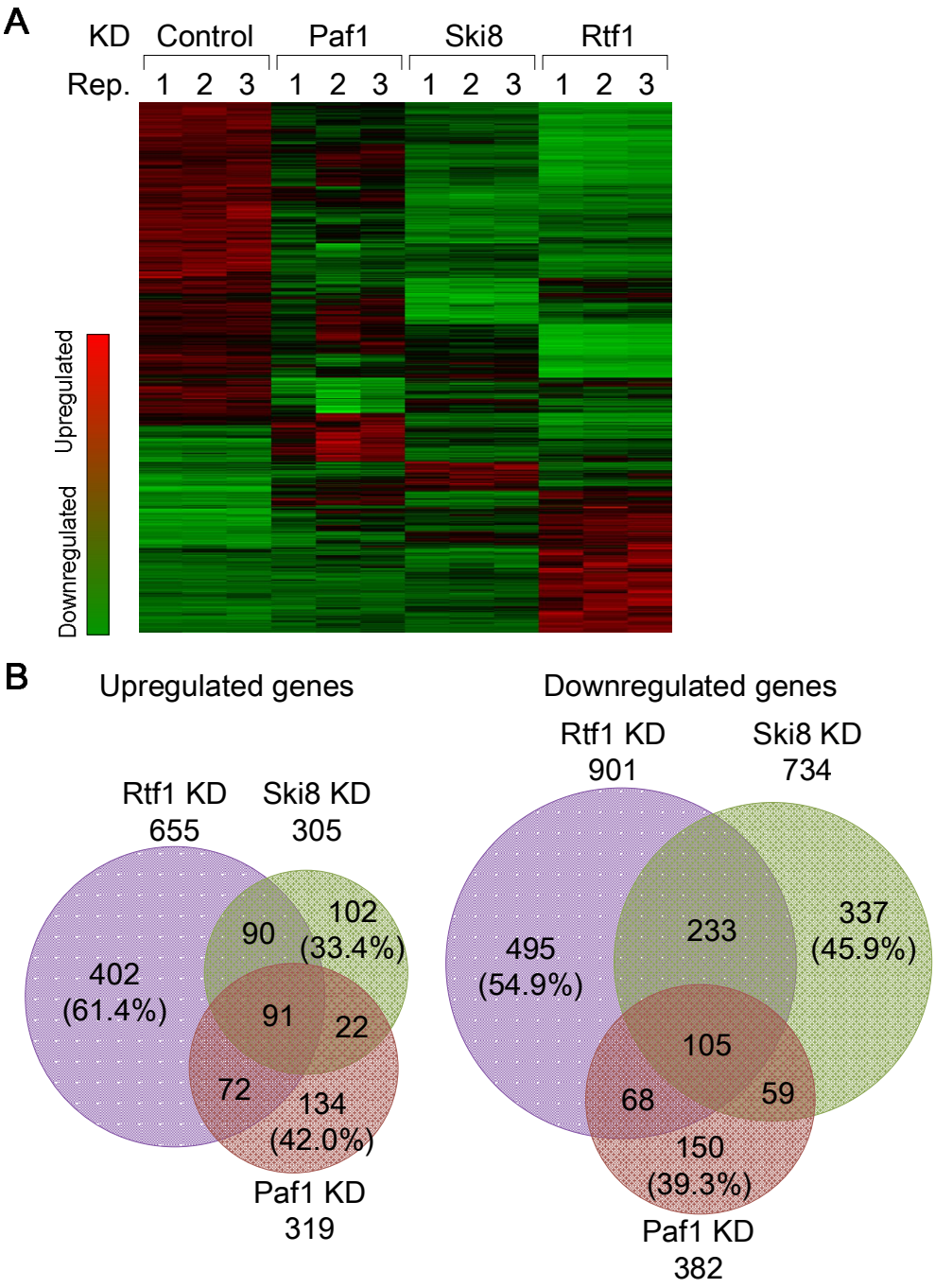

Figure 5-3. Transcript profiles of Rtf1 and PAF1C knockdown cells.

(A) Total RNAs were prepared from Paf1, Ski8, and Rtf1 knockdown cells and from control HeLa cells in triplicate and subjected to RNA-seq. The heat map of 2,349 differentially expressed genes is shown. (B) Venn diagrams comparing the genes affected by Paf1, Ski8, and Rtf1 knockdown. The numbers in parentheses indicate the percentage of genes uniquely regulated by the respective factors.

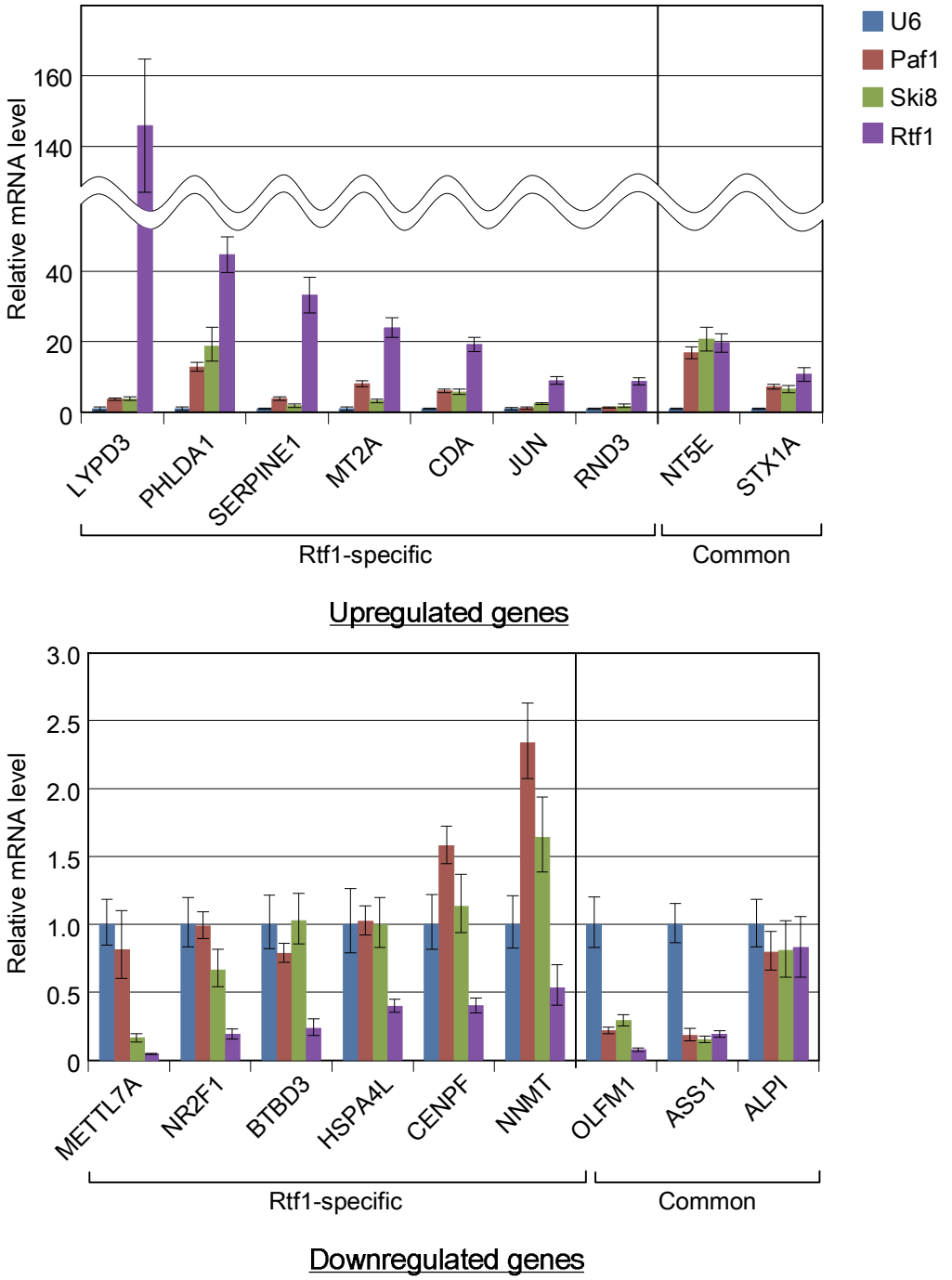

Figure 5-4. Validation of RNA-seq data.

The same set of RNA samples used in RNA-seq was subjected to qRT-PCR analysis. The averaged expression values for each gene were normalized to the values for *GADPH*, and the relative expression levels obtained from control knockdown samples were expressed as 1. Data are presented as the mean ± SEM of n=3 independent experiments.

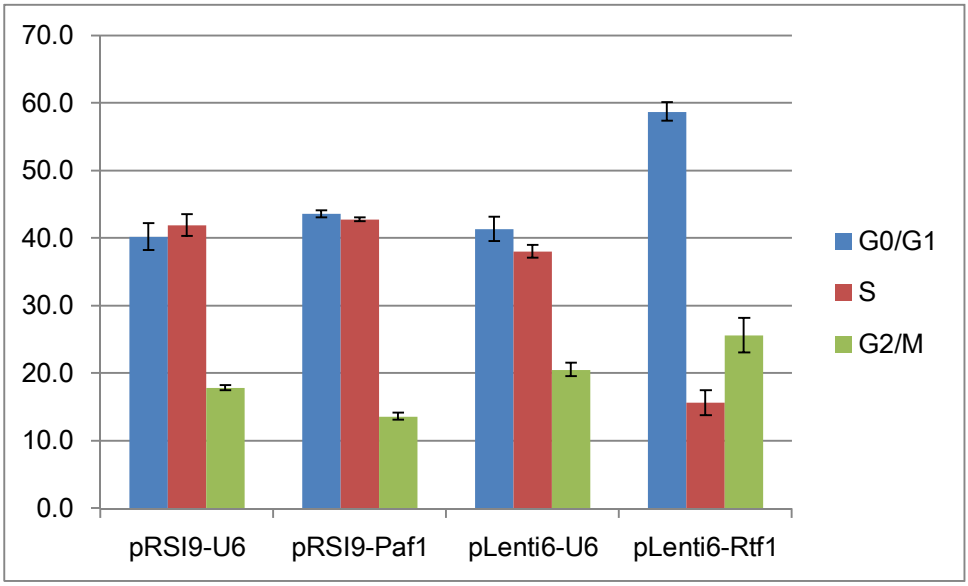

Figure 5-5. Cell cycle analysis of Rtf1 and PAF1C knockdown cells.
HeLa cells were infected with lentiviral vectors as in Figure 5-1. Four or seven days post-infection, infectants were subjected to cell cycle analysis. Data are represented as the mean ± SEM of n = 3 independent experiments.

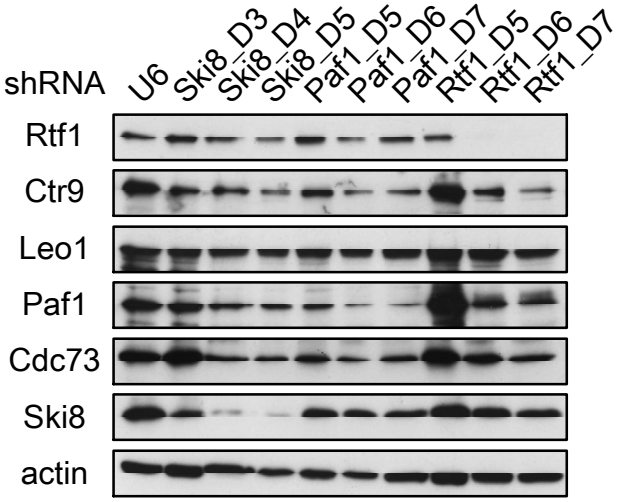

Figure 5-6. The time course analysis of the protein levels of Rtf1 and PAF1C in Ski8, Paf1, and Rtf1 knockdown cells.

HeLa cells were infected with lentiviral vectors containing the shRNAs targeting Ski8, Paf1, Rtf1 or a control vector (U6). Ski8 knockdown cells were harvested at 3, 4, and 5 days post-infection while Paf1 and Rtf1 knockdown cells were harvested at 5, 6, and 7 days post-infection. Whole cell lysates were prepared and subjected to immunoblot analysis.

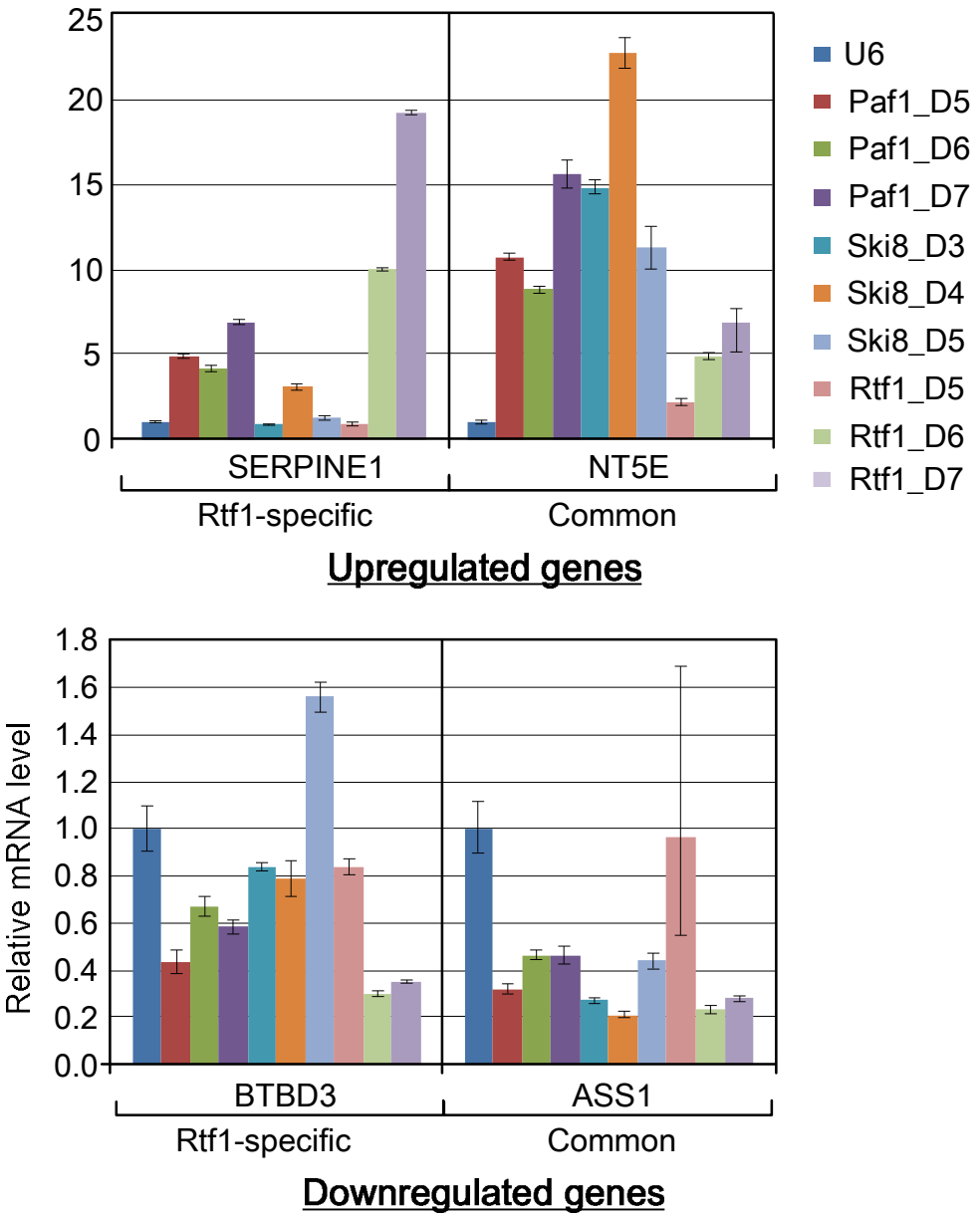

Figure 5-7. The time course analysis of gene expression changes in Ski8, Paf1, and Rtf1 knockdown cells.

HeLa cells were infected with lentiviral vectors containing the shRNAs targeting Ski8, Paf1, Rtf1 or a control vector (U6). Knockdown cells were harvested as in Figure 5-6, and total RNAs were subjected to qRT-PCR analysis. Two upregulated genes (*SERPINE1* and *NT5E*) and two downregulated genes (*BTBD3* and *ASS1*) were selected. The averaged expression values for each gene were normalized to the values for *GADPH*, and the relative expression levels obtained from control knockdown samples were expressed as 1. Data are presented as the mean ± SEM of n=3.

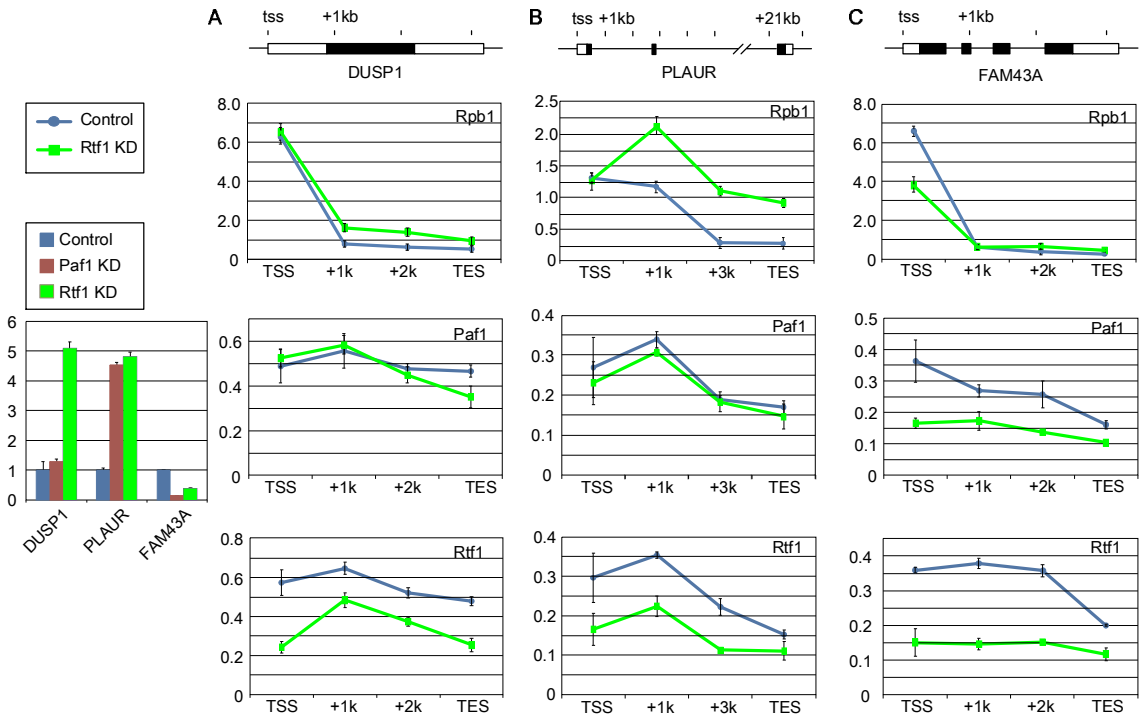

Figure 5-8. Human Rtf1-independent and - dependent recruitment of PAF1C.
HeLa cells were infected with lentiviral vectors as in Figure 5-1. Infectants were subjected to qRT-PCR analysis (A) and ChIP (B). ChIP signals are reported as the percent of input recovered. Data are represented as the mean ± SEM of n = 3 independent experiments.

Table 5-1. Genes were identified after affected by Paf1, Ski8, and Rtf1 knockdown

This table is afforded as electronic data.

Table 5-2. Gene Ontology analysis of Rtf1-, Paf1-, and Ski8-regulated genes

Term	Coun	FDR
The genes uniquely affected by Rtf1 knockdown		
GO:0000278~mitotic cell cycle	42	0.0011
GO:0007067~mitosis	30	0.0016
GO:0000280~nuclear division	30	0.0016
GO:0000087~M phase of mitotic cell cycle	30	0.0023
GO:0022403~cell cycle phase	44	0.0034
GO:0048285~organelle fission	30	0.0036
GO:0000279~M phase	37	0.0072
GO:0022402~cell cycle process	51	0.0458
GO:0001836~release of cytochrome c from mitochondria	8	0.0695
GO:0044271~nitrogen compound biosynthetic process	34	0.0830
The genes affected by Rtf1, Paf1, and Ski8 knockdown in common		
GO:0006334~nucleosome assembly	21	2.14E-08
GO:0031497~chromatin assembly	21	4.28E-08
GO:0065004~protein-DNA complex assembly	21	1.03E-07
GO:0034728~nucleosome organization	21	1.57E-07
GO:0042127~regulation of cell proliferation	67	1.67E-07
GO:0008284~positive regulation of cell proliferation	45	2.06E-07
GO:0006323~DNA packaging	21	1.12E-05
GO:0006333~chromatin assembly or disassembly	21	4.76E-05
GO:0010033~response to organic substance	56	1.90E-04
GO:0009991~response to extracellular stimulus	25	0.0024
GO:0051094~positive regulation of developmental process	28	0.0052
GO:0032101~regulation of response to external stimulus	20	0.0081
GO:0042325~regulation of phosphorylation	38	0.0103
GO:0051174~regulation of phosphorus metabolic process	39	0.0105
GO:0019220~regulation of phosphate metabolic process	39	0.0105
GO:0031667~response to nutrient levels	22	0.0160
GO:0007584~response to nutrient	18	0.0208
GO:0050727~regulation of inflammatory response	13	0.0288
GO:0006979~response to oxidative stress	19	0.0469
GO:0016126~sterol biosynthetic process	9	0.0483

CHAPTER 6: DISSCUSSION

In the present study, we showed for the first time that human Rtf1 directly activates transcription elongation in vitro. Rtf1-mediated transcriptional activation requires coactivator activity, which is most likely unrelated to DSIF or PAF1C (Chapter 2–3). A mutational study showed that the Plus3 domain of human Rtf1, but not its HMD, is required for the coactivator-dependent function (Chapter 4). Consistent with our findings in vitro, we showed that Rtf1 and PAF1C play distinct roles in the expression of a subset of genes in cultured cells (Chapter 5). Moreover, PAF1C was recruited to the genes examined apparently in an Rtf1-independent manner (Chapter 5). Therefore, the present study establishes a role for human Rtf1 as a transcription elongation factor that may function independently of PAF1C.

6.1 PAF1C-dependent and -independent functions of human Rtf1

Our finding that human Rtf1 can act independently of PAF1C is consistent with the in vitro and in vivo results of previous studies. Biochemical studies showed that human PAF1C promotes transcription elongation regardless of the presence of Rtf1 but requires other factors such as DSIF, Tat-SF1, and TFIIS for full transcriptional activation in vitro (5, 18). By contrast, the present study showed that human Rtf1 promotes transcription elongation independently of PAF1C, DSIF, and Tat-SF1. Different requirements for Rtf1- and PAF1C-mediated transcriptional activation indicate distinct mechanisms underlying these processes. In vivo, Mueller and Jaehning (31) showed that many of the phenotypes associated with paf1Δ or ctr9Δ are not enhanced, but are rather suppressed by simultaneous deletion of RTF1 in S. cerevisiae. Another recent paper showed that Rtf1 and PAF1C exert opposing effects on Pol II elongation in S. pombe (30).

Figure 5-8 suggested that PAF1C is recruited to some of its target genes independently of Rtf1. This finding is in contrast to the previous finding in S. cerevisiae that Rtf1 serves as a binding platform for PAF1C (27, 29, 67) and raises the question of how PAF1C is recruited to target genes. As the name suggests, PAF1C physically interacts with Pol II (49, 50), and a previous study showed that a Paf1–Leo1 subcomplex of PAF1C is responsible for its association with Pol II (18). Moreover, a few recent papers revealed that PAF1C directly interacts with histones (7, 19). In Kim et al. (19), for example, PAF1C was identified as an isoform-specific interactor of linker histone H1.2. Thus, there seem to be a few independent mechanisms by which PAF1C is recruited to target genes.

In the present study, we were unable to identify the Rtf1 coactivator. Besides PAF1C subunits, Rtf1 interacts with several factors, including the transcription elongation factor FACT, the chromodomain-containing protein Chd1, and the 19S proteasome (20, 38, 51, 58),

and these factors could be considered as the prime suspects. Our mutational analysis showed that the HMD of human Rtf1 is dispensable for its coactivator-dependent function in vitro (Fig. 4D). The HMD of S. cerevisiae Rtf1 is necessary and sufficient for Rtf1-mediated histone modification, and the Rtf1 HMD from various species complements the defects of HMD deletion in S. cerevisiae to varying degrees (41), suggesting its evolutionarily conserved function. Our mutational study of human Rtf1 was consistent with the previous finding in S. cerevisiae (Fig. 4B). Taken together, these results suggest that the coactivator-dependent function of human Rtf1 is independent of Rtf1-mediated histone modification. On the other hand, the Plus3 domain of human Rtf1 was found to be critical for its coactivator-dependent function (Fig. 4D). Originally implicated in single-stranded DNA binding, the Plus3 domain was recently shown to interact with the phosphorylated form of Spt5 (9, 41, 59). The Plus3 domain may serve as a binding platform for not one but multiple factors including the unidentified Rtf1 coactivator.

6.2 PAF1C-controlled, Rtf1-controlled, and H2Bub-controlled genes

GO analysis showed that genes involved in mitotic cell cycle were enriched among the genes uniquely affected by Rtf1 knockdown (Table 5-1). Concordantly, Rtf1 knockdown only seemed to increase a cell population in G1 phase (Fig. 5E). We were therefore interested in knowing whether transcriptional defects altered cell cycle or, conversely, whether altered cell cycle resulted in transcriptional changes. Of the 51 “Rtf1-specific” genes categorized into GO:0022402~cell cycle process, 44 were downregulated by Rtf1 knockdown, while 7 were upregulated by Rtf1 knockdown. Search for Cyclebase 3.0, a cell cycle-dependent gene expression database (46), indicated that 21 of the 44 downregulated genes are highly expressed in S, G2, and/or M phases. These findings are consistent with the idea that the 21 genes including BUB1, CCNA2, CCNB2, CENPA, and PLK1 were downregulated through an indirect effect of altered cell cycle. There are, however, many other cell cycle-related genes whose altered expression cannot be explained by altered cell cycle.

We also compared our RNA-seq data with microarray data of Shema et al. (48), who identified H2Bub-controlled genes in HeLa cells by knocking down RNF20, a component of the E3 ubiquitin ligase complex that mediates H2B monoubiquitination. Meta-analysis of the two datasets revealed that, of 3,469 genes affected by RNF20 knockdown, 538 genes were also affected by the knockdown of Rtf1, Ski8, or Paf1. It is not surprising to us that only a fraction of genes were identified as coregulated. RNF20 has been reported to target many proteins such as Syntaxin 1, Ebp1, AP-2alpha, and SREBP1c for polyubiquitination and degradation (6, 24, 28, 43). Hence, it is conceivable that RNF20 knockdown affects expression of many genes by mechanisms that are independent of H2B monoubiquitination. Similarly, PAF1C is a multifunctional protein complex that controls transcription in both

chromatin-dependent and -independent manners and also affects Pol II termination and 3′ processing (25, 58). Hence, it is plausible that the knockdown of a PAF1C component affects gene expression in part in an H2Bub-independent manner.

6.3 Functional similarities and differences of Rtf1 proteins among species

In recent papers, Arndt, VanDemark, and colleagues demonstrated that the interaction between the Rtf1 Plus3 domain and phospho-Spt5 is crucial for the recruitment of PAF1C in S. cerevisiae (59). In S. pombe, however, Ctr9 is recruited independently of Rtf1 (30). Similarly, we found that human Paf1 was recruited to actively transcribed genes apparently in an Rtf1-independent manner (Fig. 7), suggesting that another mechanism mediates the recruitment of PAF1C (e.g., via the direct interaction between PAF1C and Pol II). Considering the weak physical interaction between Rtf1 and PAF1C in many species except S. cerevisiae, it is reasonable to assume that an additional mechanism could significantly contribute to PAF1C recruitment.

Despite differences in their binding affinity to PAF1C, S. cerevisiae and human Rtf1 proteins have similar domain structures. Our mutational analysis showed that the highly conserved HMD of human Rtf1 is critical for H2B monoubiquitination in human cells (Fig. 4B). Moreover, the interaction with PAF1C subunits was mediated by the Rtf1 C-terminal region of approximately 100 amino acids. These results are consistent with earlier mutational studies in S. cerevisiae (41, 58) and support the fact that the Rtf1 C-terminal region is not well conserved among species; this divergence may underlie the difference in PAF1C-binding affinity among species.

6.4 Future perspectives

Several issues remain to be answered. The biggest unsolved issue of this work is the identity of the Rtf1 coactivator. Another important point is why some genes are affected only by Rtf1 knockdown despite the recruitment of both Rtf1 and PAF1C to similar levels. A detailed comparison of the genomic binding sites of human Rtf1 and PAF1C subunits would be of interest. Identification of the Rtf1 coactivator will be a key step toward the distinction of PAF1C-dependent and -independent mechanisms of action of Rtf1.

ACKNOWLEDGMENTS

This work was supported by a Grant-in-Aid for Scientific Research on Innovative Areas “Transcription Cycle” from the MEXT, Japan. I thank for the professor Yamaguchi, professor Handa, and members in Yamaguchi lab. Especially, Junichi Yamamoto, Tomoyasu Isobe, Shumpei Tateno, Yuki Murase, Yexi Chen gave me a great help in this study. Also, it was grateful for the Junko Kato for technical assistance.

REFERENCES

1. **Adelman K, Lis JT.** 2006. *Drosophila* Paf1 modulates chromatin structure at actively transcribed genes. Mol. Cell. Biol. **26:**250–260.
2. **Akanuma T, Koshida S, Kawamura A, Kishimoto Y, Takada S.** 2007. Paf1 complex homologues are required for Notch-regulated transcription during somite segmentation. EMBO Rep. **8:**858–863.
3. **Amrich CG, Davis CP, Rogal WP, Shirra MK, Heroux A, Gardner RG, Arndt KM, VanDemark AP.** 2012. Cdc73 subunit of Paf1 complex contains C-terminal Ras-like domain that promotes association of Paf1 complex with chromatin. J. Biol. Chem. **287:**10863–10875
4. **Betz JL, Chang M, Washburn TM, Porter SE, Mueller CL, Jaehning JA.** 2002. Phenotypic analysis of Paf1/RNA polymerase II complex mutations reveals connections to cell cycle regulation, protein synthesis, and lipid and nucleic acid metabolism. Mol. Genet. Genomics. **268**:272–285
5. **Chen Y, Yamaguchi Y, Tsugeno Y, Yamamoto J, Yamada T, Nakamura M, Hisatake K, Handa H.** 2009. DSIF, the Paf1 complex, and Tat-SF1 have nonredundant, cooperative roles in RNA polymerase II elongation. Genes Dev. **23:**2765–2777.
6. **Chin L-S, Vavalle JP, Li L. 2002.** Staring, a novel E3 ubiquitin-protein ligase that targets syntaxin 1 for degradation. J. Biol. Chem. **277:**35071–35079.
7. **Chu X, Qin X, Xu H, Li L, Wang Z, Li F, Xie X, Zhou H, Shen Y, Long J.** 2013. Structural insights into Paf1 complex assembly and histone binding. Nucleic Acids Res. **41:**10619–10629.
8. **Crisucci EM, Arndt KM.** 2011. The roles of the Paf1 complex and associated histone modifications in regulating gene expression. Genet. Res. Int. **2011:**707641.
9. **de Jong RN, Truffault V, Diercks T, Ab E, Daniels MA, Kaptein R, Folkers GE.** 2008. Structure and DNA binding of the human Rtf1 Plus3 domain. Structure **16:**149–159.
10. **Digman JD, Lebovitz RM, Roeder RG.** 1983. Accurate transcription initiation by RNA polymerase II in a soluble extract from isolated mammalian nuclei Nucleic Acids Res. **11:**1475-89
11. **Friedman AA, Tucker G, Singh R, Yan D, Vinayagam A, Hu Y, Binari R, Hong P, Sun X, Porto M, Pacifico S, Murali T, Finley RL Jr, Asara JM, Berger B, Perrimon N.** 2011. Proteomic and functional genomic landscape of receptor tyrosine kinase and ras to extracellular signal-regulated kinase signaling. Sci. Signal. **4:**196 rs10
12. **Huang DW, Sherman BT, Lempicki RA.** 2009. Systematic and integrative analysis of large gene lists using DAVID Bioinformatics Resources. Nat. Protoc. **4:**44–57.

13. **Jaehning JA.** 2010. The Paf1 complex: platform or player in RNA polymerase II transcription? Biochim. Biophys. Acta **1799:**379–388.
14. **James D. Watson, Tania A. Baker, Stephen P. Bell, Alexander Gann, Michael Levine, Richard Losik, Stephen C. Harrison.** 2013. Molecular Biology of the Gene (7th ed.)
15. **Jennifer F. Kugel, James A. Goodrich.** 1998. Promoter escape limits the rate of RNA polymerase II transcription and is enhanced by TFIIE, TFIIH, and ATP on negatively supercoiled DNA Proc. Natl. Acad. Sci. U. S. A. **95:**9232–9237
16. **K. Chaudhary, S. Deb, N. Moniaux, M.P. Ponnusamy, S.K. Batra**. 2007. Human RNA polymerase II-associated factor complex: dysregulation in cancer, Oncogene **26:**7499–7507
17. **Kaplan CD, Holland MJ, Winston F.** 2005. Interaction between transcription elongation factors and mRNA 3'-end formation at the Saccharomyces cerevisiae GAL10-GAL7 locus. J. Biol. Chem. **280:**913–922.
18. **Kim J, Guermah M, Roeder RG.** 2010. The human PAF1 complex acts in chromatin transcription elongation both independently and cooperatively with SII/TFIIS. Cell **140:**491–503.
19. **Kim K, Lee B, Kim J, Choi J, Kim J-M, Xiong Y, Roeder RG, An W.** 2013. Linker Histone H1.2 cooperates with Cul4A and PAF1 to drive H4K31 ubiquitylation-mediated transactivation. Cell Reports **5:**1690–1703.
20. **Krogan NJ, Buratowski S, Greenblatt JF.** 2002. RNA polymerase II elongation factors of Saccharomyces cerevisiae: a targeted proteomics approach. Mol. Cell. Biol. **22:**6979–6992.
21. **Krogan NJ, Dover J, Wood A, Schneider J, Heidt J, Boateng MA, Dean K, Ryan OW, Golshani A, Johnston M, Greenblatt JF, Shilatifard A.** 2003. The Paf1 Complex is required for histone H3 methylation by COMPASS and Dot1p: Linking transcriptional elongation to histone methylation. Mol. Cell **11:**721–729.
22. **Kubota Y, Tsuyama K, Takabayashi Y, Haruta N, Maruyama R, Iida N, Sugimoto A.** 2014. The PAF1 complex is involved in embryonic epidermal morphogenesis in *Caenorhabditis elegans*. Dev. Biol. **391:**43–53.
23. **Langenbacher AD, Nguyen CT, Cavanaugh AM, Huang J, Lu F, Chen JN.** 2011. The PAF1 complex differentially regulates cardiomyocyte specification. Dev. Biol. **353:**19–28.
24. **Lee JH, Lee GY, Jang H, Choe SS, Koo S-H, Kim JB. 2014.** Ring finger protein20 regulates hepatic lipid metabolism through protein kinase A-dependent sterol regulatory element binding protein1c degradation. Hepatology **60:**844–857.
25. **Lee JM, Greenleaf AL.** 1997. Modulation of RNA Polymerase II elongation efficiency by C-terminal heptapeptide repeat domain kinase I. J. Biol. Chem. **272:**10990–10993.
26. **Li B, Carey M, Workman JL.** 2007. The role of chromatin during transcription. Cell.

128:707–719

27. **Liu Y, Warfield L, Zhang C, Luo J, Allen J, Lang WH, Ranish J, Shokat KM, Hahn S.** 2009. Phosphorylation of the transcription elongation factor Spt5 by yeast Bur1 kinase stimulates recruitment of the PAF complex. Mol. Cell. Biol. **29:**4852–4863.
28. **Liu Z, Oh S-M, Okada M, Liu X, Cheng D, Peng J, Brat DJ, Sun S-Y, Zhou W, Gu W, Ye K.** 2009. Human BRE1 is an E3 ubiquitin ligase for Ebp1 tumor suppressor. Mol. Biol. Cell **20:**757–768.
29. **Mayekar MK, Gardner RG, Arndt KM.** 2013. The recruitment of the *Saccharomyces cerevisiae* Paf1 complex to active genes requires a domain of Rtf1 that directly interacts with the Spt4-Spt5 complex. Mol. Cell. Biol. **33:**3259–3273.
30. **Mbogning J, Nagy S, Pagé V, Schwer B, Shuman S, Fisher RP, Tanny JC.** 2013. The PAF complex and Prf1/Rtf1 delineate distinct Cdk9-dependent pathways regulating transcription elongation in fission yeast. PLoS Genet. **9:**e1004029.
31. **Mueller CL, Jaehning JA.** 2002. Ctr9, Rtf1, and Leo1 are components of the Paf1/RNA polymerase II complex. Mol. Cell. Biol. **22:**1971–1980.
32. **Narita T, Yung TM, Yamamoto J, Tsuboi Y, Tanabe H, Tanaka K, Yamaguchi Y, Handa H.** 2007. NELF interacts with CBC and participates in 3' end processing of replication-dependent histone mRNAs. Mol. Cell **26:**349–365.
33. **Ng HH, Dole S, Struhl K.** 2003. The Rtf1 component of the Paf1 transcriptional elongation complex is required for ubiquitination of histone H2B. J. Biol. Chem. **278:**33625–33628.
34. **Ng HH, Robert F, Young RA, Struhl K.** 2003. Targeted recruitment of Set1 histone methylase by elongating Pol II provides a localized mark and memory of recent transcriptional activity. Mol. Cell **11:**709–719.
35. **Nordick K, Hoffman MG, Betz JL, Jaehning JA.** 2008. Direct interactions between the Paf1 complex and a cleavage and polyadenylation factor are revealed by dissociation of Paf1 from RNA polymerase II. Eukaryot. Cell **7:**1158–1167.
36. **P.A. Wade, W. Werel, R.C. Fentzke, N.E. Thompson, J.F. Leykam, R.R. Burgess, J.A. Jaehning, Z.F. Burton.** 1996. A novel collection of accessory factors associated with yeast RNA polymerase II. Protein Expr. Purif. **8:**85-90
37. **P.J. Newey, M.R. Bowl, R.V. Thakker**. 2009. Parafibromin—functional insights J Intern Med. **266:**84-98
38. **Pan YR, Sun M, Wohlschlegel J, Reed SI.** 2013. Cks1 enhances transcription efficiency at the *GAL1* locus by linking the Paf1 complex to the 19S proteasome. Eukaryot. Cell **12:**1192–1201.
39. **Pavri R, Zhu B, Li G, Trojer P, Mandal S, Shilatifard A, Reinberg D.** 2006. Histone H2B monoubiquitination functions cooperatively with FACT to regulate elongation by RNA polymerase II. Cell **125:**703–717.

40. **Penheiter KL, Washburn TM, Porter SE, Hoffman MG, Jaehning JA.** 2005. A posttranscriptional role for the yeast Paf1-RNA polymerase II complex is revealed by identification of primary targets. Mol. Cell **20:**213–223.
41. **Piro AS, Mayekar MK, Warner MH, Davis CP, Arndt KM.** 2012. Small region of Rtf1 protein can substitute for complete Paf1 complex in facilitating global histone H2B ubiquitylation in yeast. Proc. Natl. Acad. Sci. U. S. A. **109:**10837–10842.
42. **Rappaport J, Reinberg D, Zandomeni R, Weinmann R.** 1987. Purification and functional characterization of transcription factor SII from calf thymus. Role in RNA polymerase II elongation J. Biol. Chem. **262:**5227-32.
43. **Ren P, Sheng Z, Wang Y, Yi X, Zhou Q, Zhou J, Xiang S, Hu X, Zhang J.** 2014. RNF20 promotes the polyubiquitination and proteasome-dependent degradation of AP-2α protein. Acta Biochim. Biophys. Sin. **46:**136–140.
44. **Rondon AG, Gallardo M, García-Rubio M, Aguilera A.** 2004. Molecular evidence indicating that the yeast PAF complex is required for transcription elongation. EMBO Rep. **5:**47–53.
45. **Rozenblatt-Rosen O, Nagaike T, Francis JM, Kaneko S, Glatt KA, Hughes CM, LaFramboise T, Manley JL, Meyerson M.** 2009. The tumor suppressor Cdc73 functionally associates with CPSF and CstF 3' mRNA processing factors. Proc. Natl. Acad. Sci. U. S. A. **106:**755–760.
46. **Santos A, Wernersson R, Jensen LJ.** 2015. Cyclebase 3.0: a multi-organism database on cell-cycle regulation and phenotypes. Nucleic Acids Res. **43:**D1140–1144.
47. **Sheldon KE, Mauger DM, Arndt KM.** 2005. A Requirement for the *Saccharomyces cerevisiae* Paf1 complex in snoRNA 3' end formation. Mol. Cell **20:**225–236.
48. **Shema E, Tirosh I, Aylon Y, Huang J, Ye C, Moskovits N, Raver-Shapira N, Minsky N, Pirngruber J, Tarcic G, Hublarova P, Moyal L, Gana-Weisz M, Shiloh Y, Yarden Y, Johnsen SA, Vojtesek B, Berger SL, Oren M.** 2008. The histone H2B-specific ubiquitin ligase RNF20/hBRE1 acts as a putative tumor suppressor through selective regulation of gene expression. Genes Dev. **22:**2664–2676.
49. **Shi X, Chang M, Wolf AJ, Chang CH, Frazer-Abel AA, Wade PA, Burton ZF, Jaehning JA.** 1997. Cdc73p and Paf1p are found in a novel RNA polymerase II-containing complex distinct from the Srbp-containing holoenzyme. Mol. Cell. Biol. **17:**1160–1169.
50. **Shi X, Finkelstein A, Wolf AJ, Wade PA, Burton ZF, Jaehning JA.** 1996. Paf1p, an RNA polymerase II-associated factor in *Saccharomyces cerevisiae*, may have both positive and negative roles in transcription. Mol. Cell. Biol. **16:**669–679.
51. **Simic R, Lindstrom DL, Tran HG, Roinick KL, Costa PJ, Johnson AD, Hartzog GA, Arndt KM.** 2003. Chromatin remodeling protein Chd1 interacts with transcription elongation factors and localizes to transcribed genes. EMBO J. **22:**1846–1856.

52. **Squazzo SL, Costa PJ, Lindstrom DL, Kumer KE, Simic R, Jennings JL, Link AJ, Arndt KM, Hartzog GA.** 2002. The Paf1 complex physically and functionally associates with transcription elongation factors *in vivo*. EMBO J. **21:**1764–1774.
53. **Stolinski LA, Eisenmann DM, Arndt KM.** 1997. Identification of *RTF1*, a novel gene important for TATA site selection by TATA box-binding protein in *Saccharomyces cerevisiae*. Mol. Cell. Biol. **17:**4490–4500.
54. **Tenney K, Shilatifard A.** 2006. *Drosophila* Rtf1 functions in histone methylation, gene expression, and Notch signaling. Proc. Natl. Acad. Sci. U. S. A. **103:**11970–11974.
55. **Tomson BN, Crisucci EM, Heisler LE, Gebbia M, Nislow C, Arndt KM.** 2013. Effects of the Paf1 complex and histone modifications on snoRNA 3'-end formation reveal broad and locus-specific regulation. Mol. Cell. Biol. **33:**170–182.
56. **Tomson BN, Davis CP, Warner MH, Arndt KM.** 2011. Identification of a role for histone H2B ubiquitylation in noncoding RNA 3'-end formation through mutational analysis of Rtf1 in *Saccharomyces cerevisiae*. Genetics **188:**273–289.
57. **Wada T, Takagi T, Yamaguchi Y, Ferdous A, Imai T, Hirose S, Sugimoto S, Yano K, Hartzog GA, Winston F, Buratowski S, Handa H.** 1998. DSIF, a novel transcription elongation factor that regulates RNA polymerase II processivity, is composed of human Spt4 and Spt5 homologs. Genes Dev. **12:**343–356.
58. **Warner MH, Roinick KL, Arndt KM.** 2007. Rtf1 is a multifunctional component of the Paf1 complex that regulates gene expression by directing cotranscriptional histone modification. Mol. Cell. Biol. **27:**6103–6115.
59. **Wier AD, Mayekar MK, Heroux A, Arndt KM, Vandemark AP.** 2013. Structural basis for Spt5-mediated recruitment of the Paf1 complex to chromatin. Proc. Natl. Acad. Sci. U. S. A. **110:**17290–17295.
60. **Wood A, Schneider J, Dover J, Johnston M, Shilatifard A.** 2003. The Paf1 complex is essential for histone monoubiquitination by the Rad6-Bre1 complex, which signals for histone methylation by COMPASS and Dot1p. J. Biol. Chem. **278:**34739–34742.
61. **Wozniak GG, Strahl BD.** 2014. Catalysis-dependent stabilization of Bre1 fine-tunes histone H2B ubiquitylation to regulate gene transcription. Genes Dev. **28:**1647–1652.
62. **Xiao T, Kao CF, Krogan NJ, Sun ZW, Greenblatt JF, Osley MA, Strahl BD.** 2005. Histone H2B ubiquitylation is associated with elongating RNA polymerase II. Mol. Cell. Biol. **25:**637–651.
63. **Yamada T, Yamaguchi Y, Inukai N, Okamoto S, Mura T, Handa H.** 2006. P-TEFb-mediated phosphorylation of hSpt5 C-terminal repeats is critical for processive transcription elongation. Mol. Cell **21:**227–237.
64. **Yamaguchi Y, Takagi T, Wada T, Yano K, Furuya A, Sugimoto S, Hasegawa J, Handa H.** 1999. NELF, a multisubunit complex containing RD, cooperates with DSIF to repress RNA polymerase II elongation. Cell **97:**41–51.

65. **Yamamoto J, Hagiwara Y, Chiba K, Isobe T, Narita T, Handa H, Yamaguchi Y.** 2014. DSIF and NELF interact with Integrator to specify the correct post-transcriptional fate of snRNA genes. Nat. Commun. **5:**4263.
66. **Zhang K, Haversat JM, Mager J.** 2013. CTR9/PAF1c regulates molecular lineage identity, histone H3K36 trimethylation and genomic imprinting during preimplantation development. Dev. Biol. **383:**15–27
67. **Zhou K, Kuo WH, Fillingham J, Greenblatt JF.** 2009. Control of transcriptional elongation and cotranscriptional histone modification by the yeast BUR kinase substrate Spt5. Proc. Natl. Acad. Sci. U. S. A. **106:**6956–6961.
68. **Zhu B, Mandal SS, Pham AD, Zheng Y, Erdjument-Bromage H, Batra SK, Tempst P, Reinberg D.** 2005. The human PAF complex coordinates transcription with events downstream of RNA synthesis. Genes Dev. **19:**1668–1673.

Table S1A. The list of genes that were upregulated by Rtf1, Paf1, or Ski8 knockdown

Gene Symbol	Paf1_KD_pfp	Paf1_KD_FC	Ski8_KD_pfp	Ski8_KD_FC	Rtf1_KD_pfp	Rtf1_KD_FC
ABCA1	0.025	2.289	0.031	2.048	0.034	2.438
ABCA10	0.037	12.313	0.565	3.688	0.223	6.188
ABCC3	0.001	9.329	0.945	1.229	0.007	7.857
ABHD2	0.502	1.254	0.288	1.277	0.044	2.060
ABHD5	1.058	1.051	0.100	1.572	0.004	3.621
ABL2	0.268	1.550	0.001	3.218	0.000	8.518
ABLIM3	0.001	2.869	0.117	1.536	0.001	4.705
ABTB1	0.425	1.474	0.441	1.297	0.011	3.621
ACRC	0.039	3.073	0.143	2.092	0.135	2.330
ACSL5	0.007	5.356	0.012	4.562	0.004	9.192
ACTBL2	1.063	NA	0.914	Inf	0.005	Inf
ACTN1	0.008	2.015	0.399	1.181	0.144	1.425
ADAM19	0.086	2.078	0.011	2.780	0.001	6.180
ADAM32	0.026	2.814	0.041	2.373	0.044	2.845
ADAMTS1	0.653	1.124	0.001	2.355	0.016	2.120
ADAMTS6	0.346	0.691	0.391	0.672	0.043	2.008
ADM	0.316	1.301	0.001	2.442	0.000	5.713
ADORA2B	0.508	1.216	0.439	1.223	0.015	2.211
ADRB1	0.017	0.447	0.000	2.939	0.000	0.061
ADTRP	0.204	2.836	1.078	1.179	0.017	6.388
AGPAT9	0.009	2.320	0.029	1.910	0.001	5.482
AHNAK	0.175	1.342	0.127	1.306	0.004	2.620
AHNAK2	0.717	0.880	0.090	0.504	0.001	3.545
AHR	0.000	3.849	0.001	2.451	0.008	2.602
AIM1	0.008	4.255	0.151	2.078	0.051	3.363
AIM1L	0.623	2.944	1.791	0.500	0.046	12.167
AKAP2	1.371	0.909	0.704	0.473	0.018	4.600
AKAP8L	0.423	1.231	0.004	1.906	0.011	2.166
AKR1B10	0.475	2.271	1.642	0.625	0.031	6.521
AKR1C1	1.557	0.833	1.061	1.556	0.000	58.667
ALDH1A2	0.029	6.000	1.373	0.952	0.987	0.143
ALS2CL	0.118	3.662	0.462	1.706	0.002	11.574
AMIGO2	0.000	6.728	0.001	2.489	0.000	7.297
AMIGO3	0.801	1.133	0.033	2.033	0.045	2.239
AMOTL2	0.108	1.587	0.007	2.093	0.000	7.429
AMPD3	0.740	1.720	0.271	2.500	0.038	6.120
ANGPTL2	0.040	2.894	1.660	0.955	0.418	0.212
ANGPTL4	0.000	9.799	0.000	7.355	0.000	28.568
ANKRD1	0.001	25.947	1.068	0.000	0.010	20.000
ANKRD11	0.011	2.194	0.007	2.194	0.068	1.811
ANKRD50	0.006	2.246	0.877	1.091	0.078	1.652
ANTXR2	0.000	32.953	0.005	5.891	0.001	14.578
ANXA1	0.005	1.974	0.122	1.314	0.001	3.098
ANXA10	0.067	5.270	1.134	1.378	0.000	59.405
ANXA2	0.682	1.103	0.865	1.045	0.010	2.123
ANXA3	0.057	2.004	0.157	0.455	0.000	7.109
ANXA8L2	0.909	Inf	1.092	NA	0.005	Inf
AOC2	0.275	1.651	0.004	3.517	0.007	4.390
AOX1	0.468	2.833	0.092	4.417	0.001	24.917
AP3S2	1.339	0.895	0.486	1.497	0.048	2.865
APOLD1	0.108	2.532	0.992	1.101	0.041	3.569
ARC	0.072	3.179	0.000	9.190	0.000	22.726
AREG	0.105	Inf	0.297	Inf	0.000	Inf
ARHGAP26	0.000	5.301	0.122	1.762	0.052	2.451
ARHGEF18	0.015	1.865	0.003	2.117	0.002	3.132
ARID3A	0.008	3.037	0.005	3.023	0.019	3.198
ARID3B	0.086	1.822	0.043	1.894	0.030	2.443
ARL4C	1.564	0.957	0.967	1.319	0.007	10.532
ARL4D	0.850	1.212	0.331	1.574	0.002	4.176
ARRDC4	0.219	1.645	0.109	1.776	0.040	2.575
ASAH2B	0.086	2.237	0.019	2.096	0.022	2.482
ASAP2	0.037	2.043	0.311	1.337	0.029	2.377
ASB12	1.007	1.086	0.047	2.377	0.045	2.914
ASB2	0.818	4.500	1.723	1.333	0.019	47.333
ASNS	0.005	2.052	0.427	1.128	0.221	0.583
ASPH	0.002	2.205	0.123	1.325	0.612	0.839
ATF3	0.724	0.785	0.085	1.642	0.005	3.298
ATOX1	0.759	1.054	0.467	0.803	0.003	2.912
ATP6AP1L	0.126	3.641	1.296	1.125	0.009	7.813
ATP6V1B2	0.725	0.910	0.819	1.047	0.010	2.214
ATP6V1D	0.258	0.757	0.029	1.595	0.001	3.264
ATXN1	0.037	2.179	0.174	1.523	0.106	1.867
AXL	1.422	0.964	0.723	0.848	0.009	2.292
BACH1	0.624	1.214	0.069	1.591	0.036	2.014
BAIAP2	0.687	1.207	0.105	1.544	0.003	3.696
BCAR3	0.000	4.650	0.000	3.627	0.001	4.633
BCL2L1	0.030	1.702	0.124	1.357	0.004	2.742
BCL6	0.623	1.304	0.019	2.308	0.008	3.507
BDNF	0.029	2.051	0.006	2.416	1.040	1.034
BGLAP	0.174	2.394	1.222	0.929	0.034	4.081
BHLHE40	0.042	1.713	0.080	1.436	0.004	2.733
BHLHE41	1.426	1.031	1.619	0.969	0.045	3.202
BIRC2	0.699	1.236	0.006	2.132	0.024	2.179
BLZF1	0.407	1.214	0.512	1.143	0.004	2.913
BMF	0.320	1.586	0.400	1.356	0.012	3.791
BMP2	1.169	0.897	0.030	1.862	0.009	2.899
BRI3	0.521	1.112	0.330	0.680	0.009	2.375
BRMS1L	0.904	1.155	0.012	2.049	1.700	0.933
BRPF3	0.429	1.357	0.018	2.336	0.077	2.054
BTG1	0.752	1.157	0.770	1.090	0.018	2.078
C10orf10	1.181	1.168	0.143	1.966	0.048	3.252
C11orf68	0.368	1.326	1.620	1.002	0.002	3.387
C11orf91	0.947	0.598	0.021	3.744	0.001	14.085
C11orf94	1.199	0.947	0.048	2.567	0.125	1.973
C14orf176	0.030	17.333	0.913	0.000	1.487	2.500
C16orf45	0.078	1.977	0.009	2.575	0.020	2.887
C16orf79	1.363	0.960	0.037	2.933	1.318	0.664
C17orf103	0.681	1.742	0.852	1.194	0.048	4.694
C19orf33	0.108	0.741	1.568	1.028	0.001	3.469
C19orf38	0.914	0.000	0.185	5.529	0.043	15.529
C19orf66	0.577	0.585	0.040	2.339	1.201	0.821
C1S	0.356	2.057	0.034	3.295	0.034	4.227
C20orf196	0.174	0.319	0.042	2.118	0.531	1.389
C3orf52	0.086	1.832	0.009	2.669	0.028	2.421
C4orf26	0.943	Inf	1.092	NA	0.041	Inf
C5orf46	0.111	5.815	0.333	3.630	0.005	18.926

Table S1A. The list of genes that were upregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

C6orf1	0.980	1.101	0.098	1.544	0.007	2.641
C6orf141	0.000	6.143	0.091	2.112	0.006	5.342
C6orf226	0.268	1.794	1.034	0.895	0.048	3.153
C7orf26	0.470	1.202	0.006	1.999	0.011	2.319
C9orf153	0.046	Inf	0.061	Inf	0.961	Inf
C9orf156	0.470	1.303	0.168	1.361	0.026	2.095
C9orf169	0.000	9.120	0.770	1.236	0.000	27.140
C9orf3	0.002	2.463	0.342	1.237	0.906	0.920
C9orf9	0.763	0.814	0.490	0.322	0.032	3.661
CA9	1.285	1.009	0.000	3.016	0.010	0.200
CAMK2N1	0.443	1.771	1.342	0.848	0.037	3.752
CAPN2	0.005	1.987	0.007	1.815	0.001	3.385
CAPRIN2	0.002	3.307	0.029	2.074	0.008	3.340
CARD16	0.303	1.607	0.026	2.354	0.157	2.016
CARD6	1.604	0.961	0.565	0.650	0.019	2.883
CASP4	0.046	1.735	0.001	2.525	0.001	3.519
CAV1	1.114	1.106	0.335	1.286	0.008	2.709
CBLB	0.011	2.051	0.686	1.131	1.337	1.001
CBX7	0.487	0.916	0.004	2.611	0.267	0.413
CCDC102B	0.001	40.273	0.258	6.545	0.608	2.727
CCDC150	0.331	1.424	0.025	2.083	1.719	0.939
CCDC19	0.007	4.087	0.843	0.626	1.059	1.304
CCDC78	0.047	2.004	0.014	2.266	0.391	0.514
CCL20	1.307	1.412	1.451	1.588	0.039	13.765
CCL26	1.033	1.538	1.566	0.788	0.004	13.000
CCNA1	0.598	2.028	0.406	2.833	0.037	7.694
CCND1	0.000	2.764	0.000	3.511	0.013	2.093
CCNK	0.425	1.347	0.018	1.831	0.024	2.089
CCNYL1	0.649	1.242	0.249	1.303	0.043	2.105
CCRL2	0.903	Inf	0.987	Inf	0.018	Inf
CD177	1.696	1.429	1.704	1.429	0.044	30.286
CD22	0.939	1.800	1.064	0.000	0.041	15.333
CD274	0.872	2.143	0.209	6.000	0.014	24.500
CD36	0.003	2.246	0.241	0.645	0.113	0.438
CD44	0.000	2.885	0.011	1.745	0.013	2.092
CD68	0.399	1.367	0.000	4.468	0.000	7.428
CDA	0.308	1.397	1.452	0.942	0.000	5.898
CDC42EP2	0.000	3.058	0.002	2.361	0.000	5.492
CDC42EP3	0.002	3.853	0.061	2.188	0.008	4.131
CDCP1	0.014	3.400	0.002	4.293	0.004	5.750
CDH13	0.000	4.821	0.001	3.048	0.027	2.362
CDH7	0.034	9.304	1.496	1.174	1.086	0.000
CDKN1A	0.040	1.594	0.561	0.847	0.001	4.154
CDKN2B	0.000	3.387	0.908	0.923	0.001	4.005
CDKN2D	0.478	1.318	0.011	2.464	0.009	3.305
CFB	0.119	0.393	1.066	1.055	0.001	4.821
CGB	1.441	0.521	1.560	0.625	0.049	5.604
CGB5	0.711	0.000	0.590	1.982	0.001	19.364
CGB8	0.906	0.319	0.292	2.681	0.000	28.377
CHAC1	0.001	4.547	0.017	2.490	0.034	2.706
CHIC2	0.860	0.890	0.295	1.215	0.004	2.967
CHPF	0.002	2.416	0.083	1.430	0.703	1.125
CHST2	0.002	3.330	0.042	1.969	0.039	2.333
CITED2	0.669	0.868	0.859	1.071	0.004	2.662
CLCF1	1.134	1.035	0.588	1.204	0.001	4.547
CLDN4	0.477	2.026	0.856	0.372	0.028	5.064
CLEC2B	0.003	2.967	0.000	4.431	0.021	2.771
CLK1	0.708	1.148	0.852	1.105	0.020	2.009
CLK4	0.021	2.100	0.329	1.294	0.017	2.568
CNRIP1	0.156	1.815	0.007	2.896	1.556	1.014
COL4A1	0.000	3.312	0.142	1.310	0.664	0.858
COL5A3	0.616	1.357	0.023	2.336	0.205	1.591
COLEC10	0.016	Inf	0.061	Inf	1.109	NA
CPA4	0.007	1.960	0.313	0.721	0.001	3.493
CPEB4	0.051	2.089	1.149	1.092	0.000	8.148
CPT1B	0.530	2.233	0.021	4.700	0.200	2.600
CREBBP	0.281	1.356	0.012	2.049	1.326	1.059
CRH	0.258	4.560	0.013	10.040	0.259	3.920
CRISPLD2	0.273	2.596	0.030	4.439	0.001	19.632
CRLF1	0.460	0.726	0.009	2.222	0.205	0.384
CRY2	0.894	1.194	0.026	1.767	0.020	2.177
CSF1	0.036	2.760	0.002	4.247	0.042	3.110
CSF2	1.063	NA	0.988	Inf	0.012	Inf
CSGALNACT2	0.249	1.541	0.922	1.103	0.003	4.341
CSRNP1	0.154	1.622	0.053	1.562	0.001	4.003
CSRNP2	0.507	1.259	0.030	1.696	0.004	2.889
CSRP1	0.789	1.139	0.916	0.881	0.009	2.251
CTSL1	1.325	0.992	0.012	1.727	0.010	2.177
CXCL1	0.000	7.548	0.047	1.911	0.070	1.969
CXCL2	0.004	2.524	0.046	1.785	0.001	4.879
CXCL3	0.013	3.678	0.040	3.033	0.001	11.190
CYP1A1	0.009	7.548	0.096	4.357	0.000	39.333
CYP27C1	0.309	0.714	0.000	3.704	0.013	0.158
CYR61	0.406	1.333	1.091	1.108	0.022	2.346
CYSLTR1	1.702	0.789	0.922	0.000	0.013	19.263
CYTH1	0.006	2.116	0.058	1.559	0.105	1.526
DACT1	0.006	2.889	0.026	2.222	0.398	0.464
DAP	0.006	2.029	0.513	0.820	0.862	1.021
DAPK3	0.268	1.262	0.024	1.635	0.005	2.539
DCAF4L1	0.117	4.462	0.043	5.179	0.207	3.256
DCBLD2	0.008	2.185	0.002	2.392	0.005	2.925
DCLK1	0.978	13.000	0.990	6.000	0.018	277.000
DCUN1D3	0.126	1.701	0.501	1.170	0.036	2.120
DDIT3	0.147	1.474	0.001	2.471	0.000	6.543
DDX60L	1.508	0.923	0.352	1.443	0.012	3.701
DGKH	0.037	2.100	0.364	1.299	1.377	0.886
DHRS2	0.017	5.048	0.749	0.222	0.217	2.667
DHRS3	0.623	1.514	0.306	1.653	0.002	6.375
DIO3	0.284	1.716	0.004	3.801	0.000	12.730
DKK1	0.010	4.319	0.002	6.032	0.000	62.819
DLGAP4	1.691	1.005	0.832	0.851	0.019	2.279
DLL4	0.637	2.239	0.200	3.000	0.008	10.652
DMD	0.001	3.212	0.259	1.332	0.355	0.503
DNAJA4	1.404	0.906	0.218	1.352	0.036	2.186
DNAJB2	0.209	1.404	0.128	1.396	0.001	3.755
DNAJB9	0.681	1.349	0.010	2.360	0.065	2.025
DNMBP	0.001	2.914	0.022	1.920	0.126	1.570

Table S1A. The list of genes that were upregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

DOCK4	0.025	2.138	0.007	2.399	0.009	3.055
DOK3	0.682	1.390	0.005	4.362	1.243	1.076
DOK7	0.040	4.796	1.695	0.796	0.131	3.556
DRAP1	0.603	0.862	0.240	1.234	0.013	2.082
DTX3L	1.365	1.043	0.014	2.200	1.512	1.016
DUSP1	0.594	1.107	0.371	1.146	0.001	3.958
DUSP10	0.863	0.838	1.442	0.991	0.005	2.982
DUSP2	1.281	0.972	0.002	2.050	0.521	0.806
DUSP4	0.000	6.281	0.000	4.696	0.000	24.754
DUSP5	0.074	1.627	0.000	4.569	0.000	8.943
DUSP6	0.000	27.847	0.000	8.653	0.000	22.990
ECM1	0.737	1.583	0.162	2.135	0.001	12.594
EDIL3	0.001	3.031	0.002	2.278	0.243	1.301
EDNRA	0.880	1.187	0.037	2.020	0.623	0.656
EFNB1	0.203	1.526	0.008	2.435	0.077	1.893
EFNB2	0.429	1.211	0.117	1.417	0.016	2.297
EGFR	0.001	2.468	0.000	2.938	0.010	2.446
EHD1	0.028	1.690	0.047	1.518	0.002	3.078
EIF5A2	0.810	1.213	0.061	1.970	0.035	2.665
EMP1	0.001	6.216	0.013	3.474	0.000	18.552
ENC1	0.010	2.522	0.012	2.352	0.000	7.628
ENO2	0.665	1.386	0.049	3.543	0.024	5.357
EPAS1	0.040	1.614	0.001	2.347	0.034	1.788
EPC1	0.047	1.699	0.110	1.431	0.011	2.433
EPC2	0.285	1.491	0.189	1.456	0.028	2.202
EPGN	0.000	8.012	0.024	2.228	0.000	7.360
EPHA2	0.000	8.265	0.000	8.399	0.000	18.378
EPHB2	0.000	3.157	0.003	2.054	0.032	1.839
EPPK1	0.009	2.015	0.306	0.688	0.187	1.353
EPS8	0.006	2.462	0.042	1.784	0.079	1.762
ERAP1	0.182	1.562	0.014	2.148	0.898	1.146
ERCC6	0.026	2.230	0.082	1.684	0.005	3.571
ERN1	0.062	2.752	0.014	3.513	0.017	4.427
ERRFI1	0.000	4.055	0.000	2.817	0.000	7.877
ETS1	1.498	0.895	1.708	0.961	0.011	3.406
ETV1	0.002	4.648	0.011	3.228	0.118	2.186
ETV3	0.130	1.554	0.037	1.742	0.031	2.110
ETV4	0.000	6.249	0.000	5.415	0.001	4.942
ETV5	0.000	13.560	0.000	10.143	0.001	9.240
EXT1	0.003	2.183	0.005	2.024	0.011	2.312
F2R	0.010	2.642	0.757	0.770	0.280	1.706
F2RL1	0.001	2.816	0.026	1.687	0.002	3.167
F3	0.982	1.137	0.462	1.258	0.008	3.266
FAM103A1	0.399	1.298	0.079	1.570	0.009	2.888
FAM105A	0.001	2.786	1.315	0.918	0.057	0.307
FAM107B	0.492	1.251	0.220	0.630	0.015	2.247
FAM109A	0.039	2.225	0.213	1.574	0.033	2.643
FAM126A	0.037	1.890	0.056	1.652	0.025	2.180
FAM129A	0.005	2.611	0.941	1.069	0.192	0.371
FAM196B	0.008	5.500	0.442	1.654	0.010	6.551
FAM214B	1.395	1.047	0.208	0.487	0.002	4.288
FAM222A	0.338	1.875	0.038	3.216	0.099	2.920
FAM25A	1.063	NA	1.092	NA	0.000	Inf
FAM25B	1.063	NA	1.092	NA	0.005	Inf
FAM25C	1.063	NA	1.092	NA	0.001	Inf
FAM25G	1.063	NA	1.092	NA	0.012	Inf
FAM40B	1.256	0.967	0.082	1.582	0.013	2.670
FAM50A	0.295	1.261	0.334	0.743	0.011	2.290
FAM70B	1.063	NA	0.185	Inf	0.003	Inf
FAM83A	0.190	0.560	0.055	0.337	0.001	5.041
FAM83H	0.006	2.172	0.061	1.529	0.189	1.378
FAM89B	0.034	1.673	0.045	1.538	0.009	2.283
FAM9B	0.006	Inf	0.500	Inf	0.062	Inf
FCRLB	1.360	0.889	0.359	1.914	0.036	4.506
FEM1C	1.372	1.023	1.653	0.975	0.035	2.066
FHL2	0.001	2.460	0.006	1.882	0.020	1.957
FICD	0.772	0.678	0.940	1.057	0.037	2.626
FLCN	0.420	1.240	0.283	1.279	0.006	2.774
FLRT3	0.000	3.547	0.639	0.719	0.224	0.431
FMN1	0.002	6.200	1.131	1.165	0.412	1.800
FN1	0.001	3.317	0.157	1.382	0.035	2.061
FNDC3B	0.001	2.838	0.024	1.830	0.199	1.407
FNDC4	0.419	1.772	0.021	3.171	0.517	0.350
FNIP1	0.854	1.141	0.113	1.455	0.023	2.201
FOLR1	0.707	1.312	1.300	0.955	0.001	6.410
FOLR3	0.915	0.000	0.920	0.000	0.047	13.000
FOSB	0.141	2.621	0.000	11.356	0.001	14.586
FOSL1	0.000	4.645	0.000	8.549	0.000	14.837
FOSL2	0.001	2.788	0.001	2.446	0.002	3.493
FOXD1	0.002	2.836	0.000	3.543	0.004	3.331
FOXL1	0.384	1.257	0.203	1.308	0.006	2.765
FOXQ1	1.197	0.929	0.007	2.554	0.000	11.294
FRG2B	0.958	Inf	0.427	Inf	0.005	Inf
FRMD6	0.424	1.521	1.753	1.002	0.018	2.604
FST	0.303	1.515	0.004	2.541	0.021	2.523
FSTL1	0.183	1.381	0.002	2.126	0.325	0.676
FTH1	0.096	1.429	0.460	1.135	0.003	2.660
FZD10	0.046	2.360	0.866	0.680	0.127	1.934
FZD8	0.029	1.719	0.000	2.741	0.046	1.747
GABARAPL2	0.360	0.777	0.268	0.683	0.020	2.035
GADD45A	0.014	1.996	0.088	1.560	0.001	4.505
GADD45B	0.212	1.403	0.000	3.382	0.000	7.397
GALNT9	0.556	0.631	1.113	0.819	0.004	4.138
GAST	1.063	NA	0.746	Inf	0.001	Inf
GBP3	0.450	1.969	0.079	3.594	0.042	5.016
GBX1	0.822	1.741	1.076	0.000	0.037	9.630
GCH1	1.236	1.051	0.946	0.801	0.048	2.036
GDF15	0.000	8.694	0.939	1.045	0.015	2.768
GEM	1.717	0.889	0.238	2.533	0.013	9.244
GEMIN8	1.540	0.927	0.873	0.808	0.048	2.098
GFPT2	0.001	3.468	0.000	4.129	0.000	10.493
GLI4	0.400	1.248	1.093	0.955	0.014	2.427
GLIS1	0.156	3.200	0.023	4.873	0.189	2.818
GLRX	0.000	3.383	0.008	1.900	0.005	2.648
GLS	0.037	1.694	0.001	2.416	0.008	2.467
GNAT3	0.003	2.949	1.134	0.843	0.246	0.344
GOLGA8A	0.074	1.673	0.006	2.136	1.473	0.953

Table S1A. The list of genes that were upregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

GPATCH3	1.271	1.066	0.082	1.474	0.018	2.192
GPCPD1	0.035	2.196	0.257	1.371	0.207	1.532
GPR157	1.146	0.841	0.017	2.370	0.835	1.239
GPR3	0.308	2.033	0.015	4.000	0.043	3.880
GPR39	0.006	2.740	1.137	0.900	0.050	2.193
GPR97	0.776	Inf	0.380	Inf	0.048	Inf
GPRC5A	0.000	3.247	0.000	2.796	0.000	10.270
GPRC5C	0.009	2.311	0.009	2.077	0.035	2.061
GPX1	0.700	1.063	0.164	1.268	0.009	2.180
GRAMD1B	0.000	3.734	0.000	4.647	0.047	1.693
GRAP	0.173	15.286	0.176	12.143	0.020	56.429
H1F0	0.078	1.764	0.029	1.877	0.042	2.057
HARBI1	0.857	0.910	0.884	1.018	0.027	2.469
HAS2	0.002	12.000	1.199	1.156	0.055	7.219
HBA1	0.038	4.148	0.004	7.333	0.001	14.457
HBA2	0.573	1.496	0.001	5.807	0.002	7.837
HBEGF	0.198	2.157	0.002	5.139	0.001	13.407
HBP1	0.553	1.287	0.498	1.154	0.013	2.332
HCFC2	0.844	1.290	0.534	1.286	0.026	2.725
HCN2	0.021	2.244	0.081	1.694	0.560	0.579
HDAC9	1.660	0.870	1.782	1.000	0.043	10.217
HES1	0.000	3.830	0.000	3.844	0.001	3.681
HES2	0.249	2.237	0.462	1.645	0.022	5.237
HHAT	0.010	8.189	1.081	1.649	0.025	8.459
HIC1	1.440	0.967	0.716	0.667	0.023	6.167
HIPK2	0.040	2.011	0.002	2.539	0.184	1.489
HIPK2	0.257	1.649	0.041	2.282	0.945	1.080
HIST1H2AA	0.134	0.276	0.042	0.000	0.048	2.194
HIST1H3C	0.844	Inf	1.092	NA	0.034	Inf
HIST1H4L	1.063	NA	0.049	Inf	0.793	Inf
HKDC1	0.437	13.250	0.921	6.500	0.006	107.500
HLA-B	0.650	1.130	0.002	2.016	0.214	1.323
HMGA2	0.000	5.524	0.002	2.564	0.117	1.637
HMOX1	0.025	0.502	0.152	1.315	0.014	2.212
HOMER3	0.121	1.536	0.336	1.307	0.004	3.114
HRH1	0.079	2.489	0.026	2.895	0.002	7.759
HS1BP3	0.084	0.497	0.059	0.353	0.038	2.014
HS3ST1	0.222	2.278	0.126	2.324	0.032	4.000
HSPA1A	0.955	0.930	0.001	2.224	0.320	0.697
HSPA6	0.198	1.603	0.000	6.992	0.113	1.746
HTR7	0.000	13.303	0.000	11.576	0.004	9.121
IBA57	1.334	1.022	0.131	1.575	0.022	2.686
ICAM1	0.001	4.656	0.000	6.534	0.101	2.301
ID1	0.131	1.572	0.000	4.065	0.045	1.683
ID2	0.002	3.947	0.000	7.155	0.000	16.535
ID3	0.042	1.581	0.000	2.472	0.253	1.227
IER2	0.895	1.032	0.012	1.732	0.005	2.430
IER3	0.005	2.149	0.017	1.714	0.001	4.049
IER5	0.034	1.709	0.012	1.836	0.001	4.079
IFI44L	0.002	2.693	1.654	0.963	0.094	0.337
IGFBP3	0.041	1.975	0.015	2.198	0.001	6.148
IGFBP4	0.006	2.103	0.063	1.498	0.574	0.797
IGFBP5	0.002	3.113	0.277	0.514	0.124	0.189
IGFBP6	0.826	0.872	0.184	0.642	0.002	3.120
IL11	1.159	0.953	0.000	2.971	0.000	6.839
IL18BP	0.001	3.657	0.026	2.251	0.135	1.789
IL1A	0.910	0.000	1.562	1.700	0.036	23.100
IL1B	1.063	NA	1.092	NA	0.001	Inf
IL1RN	1.734	1.429	0.380	6.714	0.033	33.286
IL20RB	0.017	2.919	0.517	1.349	0.698	1.360
IL24	0.960	Inf	0.286	Inf	0.000	Inf
IL33	0.978	3.571	0.021	28.714	0.015	44.714
IL4R	0.000	3.233	0.000	4.470	0.004	3.090
IL6	0.001	5.743	0.005	3.711	0.042	3.303
IL6R	0.238	0.554	0.843	0.827	0.012	2.824
IL6ST	0.002	2.343	0.000	3.042	0.002	3.219
IL7R	0.000	5.690	0.000	11.275	0.000	11.812
IL8	0.000	60.451	0.000	26.676	0.000	142.863
INO80C	0.472	1.397	0.035	1.719	0.014	2.398
INPP4B	0.001	3.126	0.397	1.317	0.151	1.555
IRAK2	0.235	1.363	0.088	1.477	0.014	2.312
IRF1	0.866	1.184	0.018	2.055	0.026	2.285
IRF2BPL	0.002	2.360	0.017	1.779	0.325	1.205
IRF7	0.470	1.447	0.460	1.346	0.011	3.557
IRS1	0.271	1.543	1.597	1.039	0.017	3.165
IRS2	0.454	0.721	0.017	2.112	0.005	3.339
ITGA2	0.000	7.509	0.000	4.831	0.000	6.845
ITGA3	0.050	1.573	0.289	0.710	0.018	2.019
ITGA5	0.000	3.174	0.003	1.967	0.001	3.609
ITGA6	0.007	2.105	0.971	1.053	0.001	3.812
ITGB4	0.008	6.382	0.968	1.291	0.163	3.127
ITGB5	0.000	4.261	0.002	2.149	0.005	2.554
ITGB6	0.013	Inf	0.690	Inf	0.948	Inf
ITGBL1	0.001	3.987	0.367	0.548	0.585	1.289
ITIH4	1.327	0.895	1.340	0.855	0.026	3.766
ITPKC	0.336	1.353	0.097	1.473	0.016	2.358
JAG1	0.000	5.499	0.000	4.052	0.002	3.626
JUN	0.510	0.865	0.014	1.793	0.000	6.633
JUP	0.000	2.887	0.778	1.079	0.188	1.335
KCNJ12	0.001	2.789	0.107	1.456	0.003	3.261
KCNJ15	0.001	4.431	0.058	1.918	1.434	1.007
KCTD16	0.026	11.000	0.618	2.286	0.550	2.619
KCTD5	0.394	1.194	0.025	1.646	0.004	2.715
KHDC1L	0.952	3.174	0.234	5.261	0.039	10.652
KHNYN	0.568	1.298	0.192	1.304	0.013	2.324
KIAA1217	0.006	2.748	0.754	0.822	0.108	1.801
KIAA1609	0.268	1.514	0.856	1.099	0.003	3.703
KIFC3	0.002	2.188	0.000	2.409	0.014	2.058
KLF10	0.063	1.615	0.043	1.565	0.011	2.281
KLF4	1.311	1.027	0.041	1.712	0.017	2.355
KLF6	0.423	1.408	0.746	0.876	0.001	4.304
KLHL15	1.312	1.113	0.004	2.200	0.044	1.899
KLHL21	1.058	1.046	0.002	2.046	0.001	3.633
KLHL28	0.602	1.503	0.042	2.341	0.048	2.705
KRT15	0.287	2.810	0.922	0.429	0.001	19.635
KRT16	1.063	NA	1.092	NA	0.020	Inf
KRT17	0.000	0.090	0.001	0.304	0.002	2.840

Table S1A. The list of genes that were upregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

KRT34	0.947	Inf	1.092	NA	0.002	Inf
KRT6A	0.938	Inf	0.991	Inf	0.004	Inf
KRT81	0.846	1.036	1.273	1.059	0.000	21.361
KRT86	1.258	0.833	1.373	0.845	0.014	5.536
LAG3	0.811	0.579	0.046	3.982	0.891	1.719
LAMA4	0.005	2.661	0.898	0.855	0.226	1.439
LAMB1	0.001	2.281	0.006	1.847	0.428	0.756
LAMB3	0.007	2.201	0.852	1.084	0.000	14.588
LAMC2	0.573	3.917	1.224	2.667	0.007	34.250
LANCL2	0.422	1.315	0.008	2.156	0.102	1.635
LAPTM5	0.078	Inf	0.517	Inf	0.003	Inf
LBH	1.325	0.793	1.427	1.034	0.030	4.517
LCAT	0.449	1.952	0.028	3.639	0.023	5.301
LETM2	0.006	5.034	0.002	5.828	0.001	13.678
LGALS9B	0.211	3.683	0.009	7.659	0.019	9.122
LGALS9C	0.405	Inf	0.047	Inf	0.008	Inf
LIF	0.001	2.843	0.292	1.234	0.006	2.823
LIME1	0.835	0.896	0.012	2.331	0.455	1.429
LIN28B	0.662	1.377	0.019	2.446	0.356	0.368
LIN37	0.628	1.234	0.026	1.795	0.030	2.156
LIPG	0.000	4.014	0.000	3.251	0.002	3.913
LIPH	0.217	2.521	1.504	0.766	0.018	5.223
LOC100287063	0.085	0.626	0.236	1.266	0.045	2.415
LOC100652741	0.028	2.333	0.163	1.546	0.156	1.905
LOC100652758	1.239	0.861	0.625	0.721	0.045	2.132
LOC100652766	0.007	2.143	0.383	1.178	0.348	1.264
LOC100996295	0.059	2.477	0.011	3.148	0.637	0.564
LOC100996314	0.081	1.664	0.007	2.171	0.035	2.224
LOC100996556	0.011	Inf	0.207	Inf	0.440	Inf
LOC100996607	1.061	0.900	0.022	2.481	0.172	0.000
LOC441124	0.244	1.819	0.612	0.568	0.012	4.200
LOC441698	0.347	0.573	0.179	0.300	0.020	3.438
LOC642799	0.740	1.405	0.025	4.473	0.395	2.216
LOC646670	0.546	2.654	0.031	7.769	0.023	11.500
LONRF3	0.016	1.913	0.001	2.517	0.005	2.819
LPXN	0.557	1.486	1.032	0.771	0.005	6.600
LRRC17	0.000	5.402	0.112	0.400	0.861	1.132
LTBP2	0.029	4.963	0.502	1.759	0.047	5.148
LURAP1L	0.006	2.534	0.291	0.561	0.007	3.052
LY6K	1.349	1.005	0.004	1.896	0.010	2.177
LYPD1	1.450	0.940	0.030	3.985	0.441	2.134
LYPD3	0.002	4.101	0.002	3.989	0.000	143.539
MAFB	0.127	2.529	0.000	6.314	0.000	19.451
MAFF	0.328	2.578	0.009	6.889	0.000	27.756
MAFG	0.020	2.276	0.014	2.209	0.020	2.572
MAL2	0.000	6.926	0.394	0.527	0.410	0.518
MALL	0.000	6.046	0.000	10.169	0.020	4.177
MALT1	0.302	1.405	0.076	1.709	0.036	2.266
MANEAL	0.630	0.798	0.008	2.081	1.504	0.936
MAP1LC3B	0.370	1.423	0.873	1.059	0.001	4.311
MAP1LC3B2	0.027	6.805	0.858	1.439	0.025	8.220
MAP2	0.013	2.593	0.133	0.213	0.206	0.187
MAP2K3	0.006	2.347	0.135	1.447	0.001	4.765
MAP3K12	0.005	2.315	1.308	0.925	0.440	0.667
MAPK13	0.545	1.835	0.185	2.165	0.050	3.949
MARCH6	0.499	1.211	0.179	1.280	0.020	2.002
MAST4	0.230	3.262	0.547	1.810	0.015	9.119
MB21D2	0.528	1.260	1.223	0.915	0.014	2.496
MBP	0.003	2.821	0.030	1.941	0.099	1.747
MCL1	0.077	1.461	0.005	1.876	0.004	2.507
MED29	0.812	0.905	0.302	1.184	0.019	2.019
MEGF6	0.149	0.229	0.350	0.429	0.009	3.898
MEIS3	0.209	1.765	0.290	1.722	0.020	3.364
MET	0.002	2.230	0.856	1.066	0.028	1.876
METTL12	0.545	1.280	0.011	2.001	0.019	2.350
MFGE8	0.003	2.658	0.845	1.094	0.010	2.689
MFHAS1	0.038	1.663	0.002	2.162	0.044	1.756
MFSD2A	0.059	2.104	0.000	4.677	0.038	2.496
MGLL	0.853	2.094	0.290	2.750	0.024	9.656
MICAL1	0.072	1.753	1.284	1.007	0.037	2.193
MICAL2	0.001	3.192	0.001	2.578	0.002	3.479
MLXIP	0.027	4.685	0.289	2.110	0.004	8.877
MMP1	0.943	Inf	0.237	Inf	0.001	Inf
MMP13	0.006	6.554	0.733	0.661	1.581	1.000
MMP2	0.001	2.390	0.027	0.447	1.120	1.032
MMP3	0.863	Inf	0.989	Inf	0.002	Inf
MON1A	0.018	1.879	0.004	2.146	0.190	1.359
MORC3	0.255	1.377	0.122	1.463	0.033	2.111
MOSPD1	1.289	1.061	0.853	0.817	0.018	2.452
MPP3	0.312	1.392	0.004	2.420	1.252	1.044
MPP4	1.561	0.781	1.728	0.625	0.003	18.281
MPRIP	0.005	2.017	0.116	1.354	0.098	1.487
MPZ	0.070	2.803	0.027	2.737	0.029	3.453
MSANTD3	1.371	1.000	0.394	0.757	0.013	2.267
MSX2	0.018	2.667	0.156	1.756	0.021	3.582
MT1E	0.002	2.412	0.992	0.947	0.009	2.429
MT1X	0.507	1.174	0.286	1.204	0.001	5.063
MT2A	0.000	4.940	0.000	3.214	0.000	20.127
MTRNR2L6	0.000	3.741	0.027	1.612	0.044	1.708
MVP	0.252	1.429	0.166	1.298	0.003	2.802
MXD1	0.068	2.787	0.006	4.074	0.005	6.231
MYEOV	0.000	43.957	0.000	9.271	0.000	50.471
MYH15	0.243	5.722	0.896	2.500	0.037	13.389
MYO10	0.004	2.133	0.010	1.835	0.203	1.302
MYO1G	0.952	3.889	0.670	2.556	0.026	28.667
MYPN	0.210	0.683	0.122	1.411	0.002	3.500
NABP1	1.344	1.015	0.088	1.455	0.002	3.245
NAGK	0.101	1.472	0.068	1.475	0.013	2.252
NAT8	0.275	0.494	0.012	2.513	0.106	0.000
NAV3	0.090	2.652	0.017	3.438	0.004	6.438
NBPF4	0.034	2.098	0.320	1.397	0.494	0.606
NBPF6	0.008	3.104	0.072	1.813	1.227	0.879
NBR1	0.191	1.508	0.149	1.314	0.017	2.119
NCEH1	0.017	1.981	0.001	2.733	0.024	2.190
NDRG1	0.229	1.319	0.003	2.108	0.000	8.382
NDST2	0.945	1.132	0.891	1.179	0.034	2.191
NDUFA4L2	1.027	1.250	0.002	4.904	0.429	1.837

Table S1A. The list of genes that were upregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

NEDD4	0.038	2.004	1.324	0.915	0.014	2.383
NEDD4L	0.467	0.778	0.451	1.147	0.013	2.442
NEK10	0.025	3.629	0.194	2.062	0.099	2.804
NEU1	0.029	1.645	0.621	0.861	0.013	2.104
NFAT5	0.016	2.344	0.065	1.769	0.167	1.561
NFKB2	0.531	1.191	0.005	1.966	0.010	2.231
NFKBID	0.029	2.250	0.027	2.068	0.757	0.765
NFKBIZ	0.370	1.585	0.004	2.855	0.009	3.498
NID2	0.471	1.263	0.152	1.348	0.020	2.121
NKX3-1	1.244	0.907	0.002	2.881	0.003	3.788
NOV	0.002	2.334	0.165	1.338	0.375	0.660
NOX5	0.006	18.813	0.416	4.000	0.217	6.125
NPAS2	0.000	3.892	0.004	2.269	0.412	1.185
NR1D1	0.269	1.396	0.002	2.826	0.000	6.820
NR1H2	0.204	1.426	0.002	2.076	0.061	1.619
NRP1	0.001	2.962	0.113	1.424	0.002	3.633
NT5E	0.000	12.758	0.000	15.815	0.000	12.989
NTN4	0.003	10.857	0.103	4.200	0.001	22.486
NTSR1	0.050	19.444	0.332	6.222	0.017	34.000
NUAK1	0.003	2.533	0.372	1.254	0.181	1.422
NYAP2	0.017	2.199	1.248	1.014	0.955	1.098
OPTN	1.597	0.972	1.325	0.917	0.005	3.515
OR2B6	0.000	Inf	0.259	Inf	0.438	Inf
OSGIN1	0.062	1.866	0.011	2.234	0.000	11.098
OSMR	0.660	1.163	0.000	3.996	0.017	2.160
OTUD1	0.216	1.960	0.000	6.121	0.002	7.250
OVCA2	0.134	0.456	0.051	1.668	0.035	2.621
PADI3	0.007	7.884	0.627	0.000	0.020	7.953
PAX8	0.913	0.552	0.731	1.724	0.034	9.310
PAX9	0.872	1.129	0.041	2.109	1.669	0.964
PBX3	0.029	2.101	0.161	1.465	0.728	0.905
PCDH7	0.000	4.736	1.369	1.054	0.026	2.363
PCNXL2	0.013	2.088	0.183	1.359	0.776	1.152
PDGFA	0.651	0.583	0.130	2.151	0.004	5.978
PDK4	0.037	2.411	0.834	0.680	0.684	1.566
PDP1	0.012	2.451	0.320	1.300	0.001	5.134
PEA15	0.012	1.965	0.046	1.674	0.013	2.333
PER2	0.520	1.650	0.044	2.388	0.346	1.463
PFKFB4	0.153	1.682	0.052	1.822	0.002	4.444
PGBD5	0.757	5.500	0.043	26.167	0.996	4.333
PHC2	0.009	1.931	0.001	2.422	0.015	2.089
PHLDA1	0.000	8.911	0.000	12.381	0.000	22.877
PHLDA2	0.017	1.907	0.005	2.098	0.001	4.666
PHLDB2	0.002	2.720	0.895	0.874	0.020	2.282
PHLDB3	1.010	0.975	1.133	1.068	0.032	2.547
PI3	0.028	Inf	0.005	Inf	0.000	Inf
PID1	0.909	0.869	0.003	2.716	0.158	1.568
PINK1	0.681	0.886	0.131	1.349	0.001	3.628
PITPNC1	0.196	1.566	0.019	2.116	1.570	0.942
PITRM1	0.148	1.405	0.024	1.655	0.006	2.500
PKIA	0.012	3.064	0.097	1.948	0.318	1.779
PLA2G4C	0.871	0.700	0.512	1.917	0.007	8.367
PLA2R1	0.007	7.100	0.643	1.520	0.471	1.980
PLAC8L1	1.529	1.192	0.029	8.269	0.634	2.615
PLAT	0.007	4.223	0.263	1.955	1.834	0.902
PLAU	0.000	11.514	0.000	5.815	0.000	11.752
PLAUR	0.000	3.826	0.001	2.404	0.001	3.658
PLCXD2	0.009	3.635	0.479	0.497	0.016	3.719
PLEK2	0.002	2.195	0.664	0.815	0.007	2.418
PLEKHG5	0.197	1.773	0.208	1.557	0.032	2.948
PLK2	0.002	2.383	0.031	1.681	0.000	5.277
PLK3	0.001	3.163	0.000	4.054	0.001	6.177
PLXDC2	0.049	2.641	1.239	0.818	0.294	0.129
PMAIP1	0.871	1.080	0.572	1.103	0.001	3.380
POLD4	0.829	1.162	0.112	0.551	0.003	2.819
POLR2A	0.405	1.195	0.017	1.712	0.011	2.214
POLR3C	1.076	1.017	0.002	2.173	0.077	1.544
POMZP3	0.559	0.768	1.139	0.979	0.018	2.594
PORCN	1.023	1.059	0.503	1.182	0.003	3.311
PPAP2B	1.410	1.000	0.000	4.149	0.033	2.273
PPAPDC1A	0.333	2.385	0.620	0.557	0.001	9.746
PPP1R13L	0.400	1.299	1.253	0.974	0.041	2.073
PPP1R15A	0.029	2.148	0.000	4.170	0.000	19.848
PPP1R21	0.046	2.006	0.747	0.760	0.116	1.796
PPP2R5B	0.132	1.592	1.465	1.040	0.015	2.569
PQLC2	0.198	0.597	0.532	1.196	0.024	2.176
PRH1	0.659	1.094	0.021	3.223	0.324	1.662
PRIC285	0.117	1.552	0.062	1.604	0.004	3.100
PRKAB1	0.204	1.364	0.020	1.780	0.017	2.221
PRKCDBP	0.121	0.643	0.179	0.640	0.011	2.160
PRKCE	0.015	2.114	0.020	1.935	1.681	0.923
PRKCH	0.099	Inf	0.136	Inf	0.004	Inf
PRKRIP1	0.335	1.316	0.023	1.750	0.014	2.297
PRNP	0.003	2.150	0.027	1.630	0.004	2.610
PRPF3	0.372	1.275	0.002	2.129	0.127	1.421
PRR22	0.800	1.139	0.040	2.214	1.572	0.846
PRRX2	0.346	1.465	0.000	4.721	0.228	0.407
PSCA	0.628	1.631	0.333	2.277	0.010	7.646
PSMB9	0.240	0.482	0.034	2.269	0.218	0.176
PSTK	0.062	2.147	0.008	2.786	0.067	2.286
PTGER4	0.012	2.132	0.154	0.442	1.128	0.907
PTGES	1.385	1.014	0.223	1.251	0.001	4.430
PTGS2	0.000	4.935	0.000	3.402	0.000	10.757
PTHLH	0.591	0.337	0.482	0.236	0.008	6.292
PTK2B	0.543	1.326	0.144	0.262	0.026	2.604
PTK6	0.311	1.534	1.041	0.829	0.013	3.355
PTPRG	0.045	2.031	0.045	1.913	1.757	0.984
PTRF	0.054	1.533	0.222	1.222	0.002	2.923
PXDC1	0.758	1.213	0.030	1.766	0.008	2.681
PXK	0.005	2.912	0.039	2.263	0.099	1.965
PXN	0.001	2.455	0.129	1.324	0.020	1.969
QSOX1	0.044	1.595	0.028	1.613	0.003	2.904
RAB27B	0.040	2.983	0.024	3.164	0.003	6.724
RABL2A	0.244	1.602	0.434	1.417	0.038	2.759
RAD54L2	0.155	1.511	0.003	2.322	0.535	1.312
RAET1E	0.003	4.928	0.009	3.381	0.005	5.489
RAET1G	0.029	3.735	0.820	1.333	0.013	5.314

Table S1A. The list of genes that were upregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

RASD1	0.617	1.744	0.030	3.577	0.004	8.346
RASGRF2	0.016	2.307	0.193	1.500	0.263	1.376
RASSF9	0.004	3.241	1.178	1.044	0.706	0.645
RBAK	0.239	2.228	0.039	3.494	0.511	2.152
RELB	0.780	0.800	0.002	2.518	0.016	2.485
RERE	0.012	2.649	0.047	2.020	0.703	0.641
RERG	0.000	Inf	0.703	Inf	1.109	NA
REXO1	0.140	1.498	0.007	2.082	0.239	1.324
RFESD	0.026	2.145	0.665	1.158	0.317	1.324
RFTN1	0.005	6.672	0.002	6.902	1.707	1.148
RGP1	0.193	1.425	0.637	1.099	0.020	2.128
RGS16	0.001	2.837	0.142	1.345	0.229	0.550
RHBDF1	0.009	2.476	1.199	0.848	0.002	4.362
RHEBL1	0.406	1.641	0.002	3.286	0.371	1.825
RHOC	1.490	1.009	0.881	0.904	0.014	2.023
RHOF	0.527	1.126	0.944	0.899	0.015	2.031
RIOK3	0.126	1.488	0.148	1.348	0.026	2.019
RIPK4	0.037	3.380	0.104	2.360	0.051	3.420
RND1	1.546	1.500	0.446	4.167	0.041	14.278
RND3	1.376	1.024	0.455	1.171	0.000	5.263
RNF19B	0.120	1.536	0.005	2.115	0.001	4.219
RNF224	0.379	3.409	0.302	2.614	0.005	11.636
RNF25	0.666	1.190	1.299	1.029	0.009	2.323
RNF38	0.062	2.060	0.015	2.460	0.627	1.306
RPS6KA2	0.001	3.345	0.369	1.199	0.287	0.549
RRAD	1.033	4.000	0.617	4.900	0.001	79.900
RRAGC	0.501	0.524	1.137	0.938	0.020	3.409
RRNAD1	0.435	0.778	1.495	0.959	0.020	2.072
RSPO3	0.948	2.375	0.001	49.125	0.002	79.250
RUNX1	0.009	3.262	0.003	3.659	0.004	5.360
RUNX2	0.042	4.712	0.357	1.942	0.214	2.712
RUSC2	0.179	1.462	0.347	1.250	0.037	2.010
RYR2	0.001	4.249	1.389	1.015	0.919	0.802
S100A10	0.390	1.195	0.162	1.277	0.001	3.708
S100A14	0.806	0.000	0.882	0.000	0.002	14.522
S100A16	0.001	2.374	0.044	1.505	0.000	7.426
S100A6	0.944	0.905	0.302	0.740	0.001	3.163
S100A9	0.356	Inf	1.092	NA	0.000	Inf
S100P	0.001	Inf	0.040	Inf	0.003	Inf
SAMD4A	0.508	1.958	0.235	1.823	0.045	3.698
SAT1	0.000	3.625	0.000	3.070	0.000	6.101
SCG2	0.029	2.510	0.088	1.924	0.131	1.914
SDC4	0.856	1.166	0.515	1.134	0.001	4.452
SDCBP	0.016	1.766	0.111	1.351	0.008	2.275
SDE2	1.524	1.063	0.027	1.725	0.020	2.154
SDR16C5	0.046	2.547	1.616	0.876	0.324	1.609
SDSL	1.152	0.944	0.998	1.030	0.023	2.251
SELPLG	1.668	0.837	0.854	0.755	0.017	7.980
SEMA3B	0.270	0.722	0.025	0.350	0.020	2.115
SEMA4B	0.033	1.677	0.003	2.037	0.111	1.456
SEMA7A	0.071	2.486	0.013	3.183	0.000	12.923
SERINC2	0.058	1.714	0.362	0.670	0.029	2.134
SERPINB2	0.922	Inf	0.990	Inf	0.001	Inf
SERPINB3	1.063	NA	0.000	Inf	0.067	Inf
SERPINB5	0.026	4.697	0.989	0.606	0.135	2.970
SERPINB7	0.840	0.613	0.458	2.774	0.047	7.710
SERPINB8	0.066	2.517	0.005	3.731	0.004	5.634
SERPINE1	0.000	4.380	0.129	1.471	0.000	32.187
SERPINE2	0.000	6.751	0.000	2.902	0.024	1.921
SERTAD1	0.269	1.463	0.000	3.150	0.000	7.620
SGK223	0.002	2.732	0.681	1.179	0.015	2.594
SH2B3	0.000	4.769	0.037	1.905	0.058	2.015
SH3BGRL3	0.093	1.433	1.321	0.950	0.003	2.766
SH3BP4	0.062	1.820	0.582	1.128	0.002	3.636
SH3RF2	0.100	1.886	1.371	1.023	0.014	2.721
SHB	0.009	2.200	0.002	2.471	0.003	3.430
SHC4	0.003	6.132	0.035	3.579	0.021	5.276
SHISA2	0.135	Inf	0.425	Inf	0.030	Inf
SKI	1.289	1.025	0.001	2.427	0.080	1.609
SLC10A5	0.799	1.625	0.043	5.400	1.298	1.225
SLC12A4	0.259	1.361	1.236	1.029	0.026	2.205
SLC12A8	0.047	6.656	0.027	7.063	0.017	10.938
SLC16A14	0.020	2.288	0.022	2.147	1.516	1.021
SLC16A6	0.176	2.385	0.006	4.593	0.003	8.154
SLC25A45	0.017	2.749	0.017	2.485	0.002	5.208
SLC29A3	0.203	1.522	0.200	1.512	0.040	2.147
SLC30A1	0.145	1.415	0.026	1.659	0.013	2.193
SLC35G2	1.367	0.997	0.008	2.135	0.003	3.443
SLC3A2	1.425	1.000	0.003	1.946	0.009	2.155
SLC45A4	0.127	1.797	0.034	2.003	0.700	0.696
SLC4A4	0.036	2.028	0.074	1.676	0.446	0.598
SLC6A15	0.477	1.370	0.011	2.035	0.266	0.519
SLC9A7	0.028	6.122	0.862	0.439	0.433	2.000
SLCO1B3	0.814	1.506	0.585	1.627	0.004	8.060
SLFN5	0.000	5.734	0.057	1.933	0.000	8.298
SLIT2	0.008	2.636	0.120	1.664	0.309	1.435
SLITRK6	0.002	46.889	0.924	1.333	0.866	1.111
SLPI	0.002	2.092	0.023	1.600	0.004	2.507
SMAD3	0.130	1.600	0.386	1.199	0.043	2.033
SMAD6	0.010	2.430	0.000	3.853	0.489	1.276
SMAD7	0.001	3.852	0.000	5.568	0.002	4.631
SMAD9	1.447	1.329	0.034	3.592	0.691	0.395
SMAGP	0.015	2.123	0.010	2.267	0.583	1.211
SMOX	0.037	2.023	0.002	2.713	0.028	2.434
SMPD1	0.577	1.190	1.334	0.966	0.014	2.292
SMTN	0.012	2.040	0.026	1.785	0.001	4.802
SNAI1	1.181	1.089	0.006	1.991	0.004	2.863
SNAI2	0.204	1.545	0.062	1.679	0.001	4.721
SNAPC1	0.810	1.137	0.045	1.645	0.015	2.289
SOCS1	0.185	5.792	0.000	22.625	0.004	20.917
SOCS6	0.021	2.055	0.871	1.065	0.184	1.465
SOX13	0.008	2.111	1.596	0.956	0.039	1.897
SOX9	0.026	2.764	0.903	1.176	0.048	2.806
SPANXB1	1.063	NA	0.820	Inf	0.000	Inf
SPHK1	0.030	1.719	0.000	2.795	0.000	6.478
SPINK1	1.063	NA	0.711	Inf	0.009	Inf
SPINK4	0.907	1.035	0.000	0.111	0.005	2.663

Table S1A. The list of genes that were upregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

SPOCD1	0.040	4.474	0.035	4.228	0.004	10.211
SPOCK1	0.000	3.160	0.090	1.446	0.437	1.210
SPP1	0.027	6.897	1.381	0.769	0.229	3.103
SPRED2	0.002	3.538	0.047	2.024	0.037	2.553
SPRY1	0.023	2.588	0.278	1.464	0.176	0.104
SPRY2	0.000	9.387	0.000	9.207	0.000	18.667
SPRY4	0.007	14.619	0.002	16.238	0.013	17.190
SPRYD3	0.141	1.491	0.227	0.621	0.013	2.336
SPSB1	0.040	1.702	0.026	1.729	0.003	3.203
SQRDL	0.160	0.519	0.379	0.635	0.041	2.095
SQSTM1	0.379	1.248	0.032	1.539	0.002	3.001
SRGAP1	0.040	1.929	0.017	2.082	0.068	1.886
SRGN	0.256	1.284	0.166	1.268	0.001	3.319
SRPX2	0.102	2.527	0.544	0.427	0.050	3.291
SRXN1	0.816	1.057	0.051	1.501	0.015	2.076
ST8SIA2	0.002	4.515	0.085	1.911	0.628	1.259
STAMBPL1	0.016	2.027	0.062	1.627	0.033	2.085
STARD13	0.374	1.447	0.615	0.667	0.040	2.347
STC1	0.072	1.799	0.000	5.000	0.000	6.989
STC2	0.185	1.408	0.003	2.168	0.001	5.053
STK17A	0.086	1.522	0.223	1.252	0.002	3.184
STMN3	1.405	1.429	0.013	12.429	0.496	3.286
STOML1	0.493	1.286	0.355	1.263	0.045	2.246
STX11	0.904	1.537	0.212	2.805	0.020	8.390
STX1A	0.000	5.794	0.000	5.203	0.001	5.203
STX3	0.485	1.455	0.105	1.748	0.009	3.626
SYNC	0.146	0.557	0.044	0.285	0.020	2.377
SYTL3	0.795	1.603	1.273	1.222	0.046	4.667
TAF13	0.329	0.775	0.552	1.138	0.009	2.350
TAF9B	0.737	1.184	1.369	0.992	0.003	2.862
TAGLN	0.511	2.012	0.840	0.381	0.007	7.250
TAPT1	0.428	1.242	0.165	1.402	0.006	3.074
TBC1D10A	0.733	1.291	1.522	1.002	0.042	2.084
TBC1D19	0.007	2.984	0.060	2.020	0.012	3.344
TBC1D2	0.037	1.892	0.504	1.226	0.001	6.053
TBC1D23	0.565	1.212	0.218	1.271	0.024	2.055
TBC1D8	0.001	3.097	0.071	1.547	0.306	1.248
TBX2	0.002	5.314	0.000	8.951	0.011	5.412
TCF7	0.821	0.868	0.568	1.226	0.026	2.889
TCF7L2	0.012	2.151	0.435	1.306	0.316	1.265
TCTA	1.391	1.023	0.757	0.860	0.016	2.158
TEX14	1.230	3.100	0.175	8.900	0.048	20.800
TFAP2C	0.216	5.889	0.058	9.333	0.001	38.500
TFF1	1.063	NA	1.092	NA	0.002	Inf
TFPI2	0.000	3.026	0.053	1.465	0.009	2.193
TGFA	0.000	12.547	0.486	1.289	0.004	3.683
TGFB1	0.003	2.217	0.128	0.547	0.470	1.200
TGFBR2	0.000	3.515	0.005	2.084	0.004	2.983
TGM2	0.000	4.129	0.058	1.505	0.001	3.509
THBS1	0.000	5.419	0.061	1.586	0.002	3.806
THSD4	0.000	7.323	0.048	2.085	0.012	3.387
TIMM23B	0.968	0.738	0.031	2.460	0.234	1.642
TIMP1	0.028	1.831	0.000	2.442	0.001	3.441
TIMP4	0.146	1.587	0.443	0.673	0.019	2.513
TINAGL1	0.000	7.394	0.040	1.775	0.000	6.398
TIPARP	0.006	2.480	0.368	1.276	0.001	5.599
TJP2	0.059	2.099	0.017	2.438	0.012	3.388
TK2	0.153	1.596	0.139	1.484	0.013	2.798
TLR4	0.002	3.754	0.046	2.081	0.009	3.720
TLX3	0.906	0.830	0.019	2.087	0.208	1.537
TM4SF1	0.720	1.163	0.018	1.666	0.010	2.205
TM4SF19	1.063	NA	0.500	Inf	0.001	Inf
TMEM158	0.963	1.094	0.000	4.605	0.001	5.362
TMEM2	0.005	2.485	0.489	1.239	0.005	3.127
TMEM37	0.037	2.339	0.076	0.348	0.124	0.300
TMEM79	0.820	1.117	0.120	1.552	0.013	2.737
TMEM87A	0.021	1.855	0.036	1.611	0.016	2.123
TNC	0.013	1.923	0.002	2.192	0.051	1.732
TNFAIP2	0.002	2.211	0.448	0.786	0.218	1.271
TNFAIP3	0.735	1.291	0.006	2.639	0.001	6.024
TNFRSF10D	0.016	1.897	0.004	2.048	0.001	4.342
TNFRSF11B	0.915	0.870	0.000	4.943	0.016	0.172
TNFRSF12A	0.809	1.171	0.000	2.650	0.001	3.709
TNFSF9	0.126	1.633	0.057	1.890	0.026	2.315
TNS4	0.003	7.150	0.520	1.900	0.002	12.933
TOLLIP	1.057	0.950	0.193	1.343	0.026	2.221
TOM1	0.204	1.468	1.290	1.105	0.012	2.383
TOP1	0.002	2.133	0.040	1.528	0.159	1.344
TOR4A	0.024	1.790	0.037	1.584	0.004	2.766
TPBG	0.009	2.022	0.008	2.015	0.004	2.934
TPRA1	0.801	0.862	0.112	1.355	0.013	2.143
TRAF1	0.793	1.931	0.196	3.621	0.014	12.552
TRAPPC6B	0.907	1.047	0.472	0.742	0.017	2.353
TRIB1	0.527	1.285	0.562	1.207	0.010	2.743
TRIM23	1.573	1.029	0.127	1.426	0.015	2.390
TRIM8	0.057	1.634	0.011	1.871	0.014	2.232
TRPV2	0.024	3.021	1.301	0.864	0.023	3.843
TSC22D2	0.470	1.207	0.105	1.429	0.005	2.792
TSKS	1.041	0.852	0.041	2.475	0.324	0.142
TSPAN1	0.010	3.669	0.890	1.228	0.019	4.047
TSPAN14	0.040	2.144	1.569	0.946	0.135	1.744
TSPAN19	0.029	Inf	0.002	Inf	0.996	Inf
TSPAN8	0.008	2.584	0.021	0.000	0.146	0.205
TSPAN9	0.017	2.520	0.196	1.487	0.439	0.513
TSSK6	0.801	0.914	0.909	0.796	0.030	2.574
TTC32	0.908	1.603	0.680	1.368	0.016	3.500
TTLL11	1.602	0.940	0.014	2.583	1.263	0.821
TUBB3	0.120	1.423	0.001	2.310	0.000	5.944
TUFT1	0.038	1.757	0.062	1.563	0.001	4.515
TXNIP	0.001	2.428	0.004	1.946	0.003	2.733
UBASH3B	0.003	5.540	0.000	7.115	0.005	7.057
UCN2	0.090	1.757	0.111	1.517	0.001	5.845
ULBP2	0.002	3.215	0.084	1.757	0.001	6.895
UNC13A	0.525	2.575	0.014	6.675	0.996	1.400
UPP1	0.751	1.069	0.237	1.222	0.001	4.316
UQCRHL	0.244	2.412	0.021	4.691	0.271	3.221
URGCP-MRPS24	0.018	2.054	0.209	1.244	0.004	2.840

Table S1A. The list of genes that were upregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

USP43	1.180	1.141	0.786	1.494	0.016	5.388
VDR	0.007	2.120	0.072	1.507	0.205	1.346
VEGFC	0.000	7.675	0.000	5.412	0.020	2.859
VGF	1.059	1.102	0.179	2.046	0.012	5.148
VIPR2	0.072	0.169	0.116	0.172	0.045	2.607
VLDLR	0.233	1.915	0.037	2.814	1.509	0.788
VMO1	0.000	4.971	0.137	0.132	0.138	0.000
WDFY2	0.034	2.013	0.200	1.385	0.091	1.763
WDR20	1.289	1.067	0.047	1.755	0.012	2.679
WDR47	1.301	1.087	1.235	1.051	0.037	2.135
WDR5B	0.656	1.366	0.378	0.450	0.045	2.594
WDR69	1.063	NA	1.092	NA	0.006	Inf
WFDC3	0.308	0.000	0.533	2.277	0.000	21.831
WIZ	0.302	1.345	0.025	1.844	0.032	2.142
WNT5A	0.191	1.623	0.036	2.047	0.270	1.556
WNT7B	1.491	0.907	0.177	3.000	0.013	9.488
WNT9A	1.144	0.899	1.069	0.916	0.005	3.787
WTIP	0.074	2.914	0.005	4.452	1.040	1.108
WWC3	0.000	3.393	0.014	1.840	0.486	1.177
YAE1D1	1.164	1.117	0.937	1.033	0.011	2.434
YJEFN3	0.103	2.085	0.043	1.959	0.045	2.633
YOD1	0.834	1.205	0.004	2.316	0.141	1.514
YPEL5	0.333	1.410	0.721	0.717	0.015	2.674
YTHDF1	0.232	1.344	0.232	1.273	0.019	2.073
ZBTB38	0.062	1.719	0.104	1.482	0.009	2.703
ZBTB43	0.814	1.130	0.148	1.406	0.031	2.166
ZC3H12C	0.303	1.679	0.091	1.876	0.026	2.947
ZCCHC6	0.165	1.532	0.160	1.380	0.034	2.036
ZDHHC11	0.038	1.871	0.002	2.521	0.858	1.202
ZDHHC18	0.981	1.212	0.004	2.319	0.037	2.028
ZFAND2A	0.722	0.900	0.001	2.562	0.038	1.796
ZFP36	0.089	1.826	0.002	2.990	0.002	4.413
ZFP36L1	0.375	1.246	0.769	0.848	0.024	2.067
ZFP37	1.128	1.025	0.017	2.609	0.103	2.035
ZFYVE1	0.170	1.573	1.546	0.928	0.005	3.200
ZIC2	0.288	1.507	0.002	2.589	0.005	3.206
ZMYM5	0.718	1.276	0.423	1.335	0.037	2.733
ZNF185	0.767	1.344	0.841	1.374	0.000	18.092
ZNF187	0.040	1.903	0.057	1.737	0.017	2.531
ZNF296	1.202	0.887	0.087	2.307	0.020	3.713
ZNF311	0.017	2.574	0.361	0.492	0.839	1.225
ZNF319	0.835	1.224	0.237	1.409	0.028	2.584
ZNF333	1.254	1.156	0.570	1.467	0.049	3.622
ZNF335	0.070	1.582	0.092	1.444	0.013	2.324
ZNF385A	0.872	0.852	0.183	0.454	0.033	2.259
ZNF425	1.683	1.571	0.735	0.500	0.026	19.000
ZNF473	0.682	1.183	0.005	2.300	0.346	1.240
ZNF503	1.131	1.021	0.857	1.175	0.041	2.307
ZNF622	0.925	1.059	0.037	1.526	0.004	2.607
ZNF654	0.077	2.619	0.032	2.873	0.144	2.195
ZNF673	0.324	1.372	0.022	2.022	0.026	2.328
ZNF697	0.469	1.369	0.078	1.727	0.017	2.716
ZNF777	0.204	1.445	0.006	2.046	0.045	1.846
ZNF79	0.021	2.032	0.192	1.369	0.009	2.726
ZSCAN21	0.795	0.872	0.353	0.705	0.022	2.134
ZSWIM4	0.019	2.324	0.045	2.114	0.018	2.790
ZSWIM6	0.142	1.490	0.040	1.629	0.015	2.268
ZYX	0.013	1.900	0.018	1.765	0.003	3.007

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown

Gene Symbol	Paf1_KD_pfp	Paf1_KD_FC	Ski8_KD_pfp	Ski8_KD_FC	Rtf1_KD_pfp	Rtf1_KD_FC
AAED1	0.041	0.481	0.811	1.044	0.828	1.119
AARSD1	0.066	0.593	0.075	0.544	0.045	0.431
ABCA2	0.085	0.544	0.053	0.430	0.003	0.116
ABCB6	0.111	0.632	0.005	0.340	0.033	0.379
ABHD14A	0.191	0.687	0.016	0.366	0.126	0.476
ABHD14B	0.656	0.841	0.005	0.329	0.173	0.567
ABRACL	0.085	0.642	0.000	0.234	0.000	0.164
ACACA	0.231	0.746	0.022	0.435	0.001	0.191
ACAD11	0.166	0.609	0.006	0.182	0.335	0.606
ACAT1	0.042	0.572	0.023	0.438	0.015	0.335
ACAT2	0.029	0.529	0.002	0.314	0.010	0.327
ACLY	0.053	0.608	0.035	0.494	0.016	0.373
ACOT13	0.011	0.376	0.083	0.517	0.069	0.396
ADA	0.014	0.368	0.074	0.487	0.012	0.241
ADAM15	0.662	0.892	0.129	0.616	0.043	0.429
ADAMTS3	1.331	1.060	0.030	0.175	0.079	0.183
ADAP2	0.023	0.190	0.039	0.205	0.031	0.050
ADARB2	0.364	0.742	0.403	0.727	0.035	0.264
ADCK4	0.657	0.876	0.028	0.404	0.398	0.716
ADCY3	0.714	0.865	0.089	0.531	0.003	0.192
ADD3	0.730	0.912	0.019	0.415	0.003	0.228
ADRB1	0.017	0.447	0.000	2.939	0.000	0.061
ADSSL1	0.016	0.391	0.002	0.227	0.007	0.211
AES	0.601	0.874	0.024	0.468	0.004	0.289
AFAP1	0.085	0.614	0.001	0.246	0.001	0.171
AGAP3	1.059	0.921	0.549	0.834	0.042	0.388
AGBL5	0.148	0.691	0.005	0.319	0.101	0.477
AGL	0.123	0.628	0.120	0.529	0.034	0.299
AGMAT	0.216	0.691	0.215	0.637	0.009	0.226
AGRN	0.730	1.140	0.022	0.379	0.364	0.679
AHCY	0.129	0.678	0.114	0.605	0.016	0.375
AIF1L	0.068	0.611	0.000	0.131	0.000	0.140
AIG1	0.050	0.549	0.005	0.325	0.000	0.110
AK4	0.111	0.600	0.191	1.291	0.015	0.229
AKAP12	0.034	0.517	0.002	0.254	0.486	0.769
AKR7A2	0.037	0.503	0.032	0.461	0.435	0.763
AKT1	0.051	0.571	0.002	0.334	0.063	0.467
ALDH18A1	0.720	0.849	0.133	0.618	0.033	0.406
ALDH1B1	0.189	0.721	0.092	0.565	0.007	0.297
ALDH1L2	1.523	0.933	0.052	0.348	0.010	0.088
ALDH3B1	0.051	0.517	0.007	0.321	0.732	0.830
ALG8	0.086	0.649	0.027	0.465	0.033	0.403
ALKBH7	0.146	0.651	0.157	0.615	0.014	0.302
ALPI	0.000	0.026	0.000	0.065	0.000	0.009
ALPL	0.000	0.218	0.075	0.533	0.000	0.111
ALX1	0.358	0.741	0.202	0.669	0.035	0.307
ALX4	0.138	0.642	0.267	0.653	0.033	0.274
AMDHD2	0.566	0.805	0.026	0.353	0.178	0.504
AMTN	1.232	0.944	0.020	0.423	0.718	1.079
ANAPC15	0.155	0.695	0.021	0.458	0.048	0.438
ANKLE1	0.860	1.051	0.701	0.822	0.019	0.167
ANKRD13B	0.269	0.724	0.179	0.611	0.015	0.266
ANKRD27	0.156	0.722	0.000	0.205	0.010	0.288
ANKRD29	0.032	0.457	0.099	0.541	0.005	0.171
ANKRD32	0.084	0.647	0.002	0.290	0.003	0.246
ANP32A	0.440	0.802	0.153	0.638	0.013	0.349
ANXA6	0.493	0.836	0.005	0.369	0.006	0.308
ANXA9	0.333	0.710	0.008	0.083	0.136	0.299
APLP1	1.138	1.053	0.109	0.498	0.038	0.278
APOA1BP	0.102	0.637	0.031	0.487	0.016	0.374
APOBEC3C	0.132	1.524	0.003	0.213	0.009	0.200
APOC1	0.024	0.437	0.015	0.381	0.019	0.303
APOO	0.048	0.544	0.193	0.658	0.033	0.397
APP	0.814	1.159	0.669	0.882	0.016	0.361
AQP3	0.000	0.188	0.000	0.049	0.203	0.588
ARF3	1.197	1.042	0.002	0.337	0.100	0.514
ARG2	0.004	0.279	0.001	0.145	0.155	0.483
ARHGAP11A	0.130	0.670	0.040	0.462	0.092	0.464
ARHGEF16	0.481	0.830	0.013	0.388	0.040	0.389
ARHGEF25	0.167	0.588	0.100	0.412	0.026	0.140
ARHGEF40	0.042	0.496	0.002	0.242	0.075	0.418
ARL16	0.044	0.497	0.566	0.804	0.555	0.761
ARL2	0.107	0.649	0.030	0.480	0.110	0.526
ARL3	0.218	0.697	0.044	0.401	0.070	0.364
ARL5A	0.419	0.804	0.727	0.885	0.039	0.356
ARL6IP4	0.132	0.678	0.003	0.340	0.131	0.547
ARL6IP5	0.763	0.862	0.027	0.458	0.133	0.532
ARMC1	0.225	0.753	0.040	0.461	1.016	0.892
ARPC1B	1.141	0.930	0.037	0.494	0.058	0.458
ARRB2	1.379	0.962	0.138	0.592	0.029	0.336
ARRDC1	0.114	0.611	0.002	0.254	0.013	0.288
ARV1	0.305	0.745	0.035	0.429	0.205	0.567
ASAP3	0.701	0.844	0.009	0.293	0.002	0.087
ASB13	0.069	0.571	0.380	0.771	0.025	0.368
ASB3	0.148	0.567	0.008	0.110	1.388	1.047
ASB9	0.223	0.663	0.000	0.154	0.002	0.154
ASF1B	0.029	0.493	0.298	0.720	0.288	0.655
ASGR1	0.048	0.279	0.312	0.538	0.068	2.285
ASIC1	0.071	0.505	0.009	0.262	0.003	0.085
ASL	0.812	0.909	0.018	0.398	0.082	1.538
ASMTL	0.219	0.691	0.188	0.628	0.035	0.339
ASPM	0.811	0.893	0.088	0.552	0.002	0.226
ASS1	0.000	0.151	0.000	0.083	0.000	0.134
ATAD3B	0.017	0.473	0.383	0.780	0.037	0.413
ATG9A	1.345	1.008	0.043	0.481	0.510	0.795
ATP11C	1.334	0.915	0.239	0.664	0.008	0.228
ATP1A1	0.892	1.120	0.410	0.793	0.049	0.444
ATP5G1	0.042	0.549	0.064	0.550	0.038	0.428
ATP5H	0.074	0.636	0.024	0.473	0.074	0.485
ATP5I	0.008	0.459	0.109	0.603	0.334	0.710
ATP5O	0.025	0.496	0.227	0.691	0.133	0.551
ATP8B2	1.333	0.949	0.012	0.347	0.044	0.359
ATPIF1	0.054	0.557	0.025	0.472	0.122	0.534
AURKB	0.667	0.872	0.712	0.895	0.035	0.414

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

B3GNT1	0.029	0.440	0.458	0.782	0.025	0.296
B3GNT9	0.818	1.046	0.028	0.373	1.574	1.013
B3GNTL1	0.600	1.215	0.030	0.346	0.135	0.441
B4GALNT4	0.682	1.178	1.273	0.995	0.013	0.141
B4GALT1	0.000	0.274	0.929	0.879	0.049	0.409
B4GALT2	0.104	0.628	0.361	0.764	0.038	0.413
B9D1	0.066	0.545	0.000	0.143	0.017	0.291
BACE2	0.128	0.666	0.051	0.484	0.003	0.220
BAG1	0.046	0.608	0.110	0.593	0.008	0.315
BARX1	0.472	0.801	0.520	0.783	0.000	0.074
BASP1	0.001	0.326	0.017	0.447	0.060	0.464
BATF3	0.021	0.434	0.143	0.583	0.002	0.166
BCKDHB	0.254	0.678	0.006	0.216	0.029	0.236
BCL7C	0.024	0.498	0.066	0.536	0.143	0.550
BDH1	0.025	0.377	0.028	0.334	0.006	0.104
BDKRB2	0.457	1.326	0.009	0.083	0.364	0.593
BID	0.330	1.314	0.724	0.884	0.019	0.291
BIRC7	0.744	0.842	0.049	0.483	0.357	0.698
BLM	0.950	0.918	0.201	0.617	0.037	0.323
BLOC1S1	0.395	0.754	0.026	0.469	0.130	0.544
BLVRA	0.146	0.685	0.021	0.415	0.054	0.409
BLVRB	0.005	0.386	0.005	0.361	0.285	0.670
BMP5	0.133	1.421	0.219	0.670	0.008	0.292
BMP6	0.012	0.360	0.310	0.674	0.408	0.664
BMPR1B	1.493	0.995	0.108	0.520	0.030	0.297
BNIP3	0.019	0.450	0.879	0.933	1.228	1.019
BOLA1	0.127	0.615	0.022	0.393	0.549	0.784
BOLA3	0.023	0.485	0.126	0.613	0.378	0.730
BRP44L	0.022	0.484	0.022	0.445	0.079	0.483
BST2	0.808	0.899	0.375	0.776	0.011	0.346
BTBD2	0.170	0.687	0.018	0.437	0.003	0.266
BTBD3	0.064	0.638	0.225	0.682	0.000	0.139
BTN3A1	0.085	0.459	0.091	0.393	0.017	0.092
BTN3A2	0.043	0.517	0.182	0.596	0.007	0.186
BTN3A3	0.141	0.623	0.096	0.482	0.013	0.178
BUB1	0.708	0.853	0.173	0.641	0.036	0.400
BUB1B	0.689	0.849	0.445	0.798	0.021	0.362
BZW2	0.603	0.912	0.092	0.576	0.008	0.317
C11orf10	0.021	0.478	0.123	0.617	0.455	0.779
C11orf71	0.003	0.174	0.067	0.410	0.144	0.499
C11orf74	0.265	0.759	0.081	0.456	0.007	0.174
C12orf45	0.016	0.471	0.267	0.714	0.063	0.455
C12orf75	0.056	0.541	0.051	0.485	0.014	0.312
C15orf23	0.086	0.611	0.196	0.661	0.046	0.423
C16orf13	0.004	0.392	0.003	0.360	0.020	0.382
C16orf59	0.106	0.625	0.114	0.563	0.004	0.212
C16orf74	0.034	0.444	0.002	0.120	0.222	0.507
C17orf58	0.026	0.364	0.037	0.358	0.136	0.428
C17orf61	0.004	0.401	0.007	0.401	0.243	0.659
C17orf96	0.902	0.901	0.643	1.104	0.019	0.261
C19orf60	0.119	0.641	0.002	0.272	0.250	0.647
C1orf233	1.046	1.057	1.102	0.913	0.014	0.242
C1orf53	0.022	0.329	0.097	0.455	0.188	0.472
C1orf85	0.146	0.668	0.007	0.388	0.328	0.700
C1QBP	0.057	0.583	0.394	0.786	0.033	0.420
C1QL1	0.050	0.364	1.369	1.047	0.150	0.391
C1RL	0.905	0.858	0.017	0.209	0.242	0.490
C20orf112	1.280	1.017	0.331	0.631	0.050	0.183
C20orf27	1.606	0.959	0.218	0.683	0.006	0.300
C21orf33	0.074	0.636	0.027	0.452	0.104	0.499
C2orf28	0.013	0.453	0.077	0.551	0.289	0.668
C3orf26	1.072	0.932	0.161	0.644	0.039	0.424
C4orf33	1.183	1.004	0.029	0.369	0.063	0.353
C4orf34	0.146	0.496	0.034	0.260	0.090	1.680
C6orf108	0.175	0.704	0.000	0.261	0.011	0.339
C6orf57	0.449	1.316	0.044	0.139	0.411	0.523
C8orf22	0.001	0.310	0.891	0.893	0.000	0.063
C8orf73	0.165	0.598	0.000	0.027	0.090	0.342
C9orf116	0.037	0.470	0.003	0.266	0.007	0.250
C9orf41	0.895	1.114	0.036	0.423	0.105	0.454
CA11	0.878	1.107	0.005	0.175	0.025	0.201
CA9	1.285	1.009	0.000	3.016	0.010	0.200
CACNA2D1	0.753	1.216	0.679	0.873	0.032	0.334
CADPS2	1.422	0.963	0.021	0.239	0.010	0.066
CALD1	0.172	0.742	0.008	0.393	0.000	0.186
CAMK1	0.136	0.554	0.162	0.518	0.037	0.199
CAMK4	1.399	1.098	0.264	0.614	0.033	0.144
CAMKMT	0.192	0.659	0.231	0.645	0.031	0.322
CARD10	0.025	0.426	0.006	0.266	0.310	0.628
CARHSP1	0.036	0.483	0.086	0.536	0.273	0.638
CARM1	0.658	0.824	0.453	0.806	0.011	0.321
CASP9	0.005	0.335	0.067	0.484	0.121	0.477
CAT	0.060	0.595	0.003	0.320	0.071	0.456
CBS	0.101	0.629	0.000	0.221	0.000	0.069
CBX2	0.487	0.804	0.766	0.865	0.012	0.281
CBX5	0.344	0.799	0.186	0.652	0.010	0.307
CBY1	0.964	0.891	0.047	0.382	0.255	0.564
CCBL1	0.265	0.726	0.005	0.286	0.019	0.295
CCDC106	0.829	0.869	0.024	0.387	0.105	0.443
CCDC109B	0.103	0.628	0.027	0.278	0.319	0.557
CCDC167	0.007	0.381	0.491	0.795	0.447	0.723
CCDC28B	0.106	0.622	0.036	0.452	0.008	0.278
CCDC3	1.402	1.024	0.080	0.340	0.037	0.118
CCDC34	0.138	0.606	0.123	0.526	0.033	0.286
CCDC56	0.001	0.327	0.264	0.717	0.294	0.687
CCDC77	1.183	0.973	0.562	0.835	0.037	0.377
CCDC85C	0.493	0.838	1.527	0.965	0.011	0.315
CCDC88C	0.779	0.830	1.589	0.941	0.037	0.284
CCNA2	0.112	0.674	0.921	0.912	0.020	0.373
CCNB1	0.227	0.760	0.019	0.454	0.010	0.333
CCNB2	0.443	0.833	0.051	0.520	0.023	0.382
CCNG1	1.187	1.021	0.037	0.484	0.007	0.306
CD248	0.119	0.468	0.020	0.108	0.037	0.053
CD276	0.615	1.254	0.015	0.414	0.010	0.305
CD320	0.159	0.699	0.445	0.809	0.017	0.376

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

CD82	0.243	0.709	0.048	0.443	0.056	0.369
CD9	0.035	0.508	0.005	0.351	1.481	0.992
CD99L2	0.650	1.254	0.126	0.527	0.022	0.242
CDC26	0.136	0.654	0.000	0.097	0.427	0.709
CDCA3	1.220	0.934	0.069	0.547	0.042	0.420
CDCA7	0.234	0.729	0.038	0.463	0.025	0.341
CDCA7L	0.085	0.641	0.037	0.477	0.036	0.391
CDK5	0.028	0.484	0.015	0.421	0.092	0.487
CDKL1	0.094	0.529	0.722	0.821	0.037	0.277
CDKN2AIPNL	0.170	0.695	0.025	0.436	0.013	0.314
CDKN2C	0.836	0.840	0.212	0.677	0.005	0.279
CDKN3	0.042	0.612	0.002	0.317	0.004	0.269
CENPA	0.071	0.618	0.071	0.496	0.025	0.302
CENPF	1.504	0.962	0.346	0.755	0.001	0.188
CENPH	0.024	0.480	0.006	0.337	0.110	0.504
CENPM	1.260	0.955	0.174	0.650	0.038	0.396
CENPW	0.068	0.606	0.200	0.678	0.025	0.388
CEP44	0.104	0.609	0.047	0.422	0.020	0.259
CERCAM	0.375	0.800	0.003	0.334	0.057	0.440
CETN3	0.777	0.856	0.007	0.330	0.043	0.343
CFD	0.004	0.345	0.001	0.257	0.001	0.198
CFH	0.562	0.808	0.014	0.382	0.000	0.134
CFL2	0.146	0.707	0.007	0.351	0.681	0.860
CGA	0.004	0.405	0.028	0.481	0.253	0.663
CHAC2	0.084	0.619	0.037	0.473	0.064	0.446
CHCHD10	0.399	0.789	0.143	0.594	0.005	0.266
CHCHD6	0.521	0.796	0.021	0.453	0.061	0.447
CHCHD7	0.097	0.607	0.007	0.357	0.215	0.596
CHD3	0.721	0.846	0.377	0.761	0.023	0.352
CHPT1	0.155	0.668	0.078	0.555	0.024	0.361
CHRFAM7A	0.004	0.215	0.001	0.088	0.005	0.090
CHRNA7	0.034	0.182	0.057	0.197	0.172	0.288
CIRBP	0.018	0.454	0.598	0.824	0.320	1.187
CISD3	0.077	0.554	0.040	0.454	0.130	0.501
CKAP4	0.822	1.124	0.590	0.853	0.006	0.287
CKB	0.497	0.817	0.003	0.361	0.000	0.190
CKS1B	0.272	0.769	0.640	0.876	0.031	0.413
CLNS1A	0.000	0.272	0.162	0.646	0.055	0.450
CLU	0.000	0.209	0.000	0.282	0.011	0.348
CMBL	0.012	0.467	0.001	0.275	0.000	0.103
CMC1	0.174	0.676	0.036	0.462	0.245	0.634
CMTM7	0.292	1.385	0.227	1.521	0.036	0.122
CNDP2	0.198	0.706	0.072	0.495	0.044	0.360
CNN3	0.065	0.604	0.052	0.499	0.031	0.380
CNPY2	0.027	0.506	0.004	0.375	0.034	0.416
COA5	0.207	0.679	0.024	0.359	0.326	0.634
COBL	0.001	0.236	0.000	0.070	0.000	0.043
COL11A1	0.034	0.483	0.002	0.180	0.000	0.034
COL12A1	1.444	0.950	0.039	0.471	0.003	0.230
COL16A1	1.681	1.009	0.008	0.388	0.007	0.298
COL1A1	0.328	0.746	0.079	0.483	0.003	0.141
COL1A2	0.823	1.081	0.113	0.588	0.000	0.040
COL25A1	0.551	1.230	0.080	0.538	0.001	0.190
COL3A1	0.017	1.783	0.018	0.449	0.000	0.079
COL5A2	0.032	1.718	0.508	0.814	0.025	0.329
COL8A1	0.032	0.548	0.001	0.259	0.000	0.068
COL9A1	0.780	0.763	0.047	0.128	0.080	0.075
COL9A3	0.001	0.216	0.000	0.140	0.002	0.113
COMMD4	0.373	0.774	0.033	0.482	0.558	0.820
COMMD6	0.085	0.566	0.001	0.208	0.394	0.698
COMTD1	0.032	0.472	0.002	0.253	0.005	0.227
COPG2	0.692	0.848	0.072	0.490	0.050	0.382
CORO1B	0.104	0.613	0.015	0.409	0.165	0.572
CORO2A	1.549	0.918	0.045	0.307	0.090	0.300
COX17	0.013	0.455	0.048	0.515	0.097	0.520
COX20	0.025	0.443	0.146	0.580	1.128	1.088
COX6B2	0.025	0.356	0.021	0.315	0.893	0.811
COX7B	0.006	0.414	0.063	0.549	0.077	0.493
CPE	1.441	0.963	0.218	0.665	0.015	0.326
CPM	0.000	0.254	0.410	0.774	0.024	0.342
CPNE7	0.300	0.740	0.145	0.592	0.041	0.358
CPS1	0.003	0.384	0.007	0.403	0.000	0.085
CPVL	0.286	0.757	0.020	0.411	0.050	0.389
CPZ	0.102	0.472	0.015	0.195	0.029	0.147
CRABP2	0.085	0.591	0.016	0.425	0.031	0.387
CREB3L2	0.108	0.659	0.035	0.464	0.010	0.297
CRIP1	0.198	1.325	0.002	0.306	0.875	1.035
CRIP2	0.689	0.861	0.003	0.305	0.003	0.215
CRTAP	0.146	0.693	0.056	0.513	0.013	0.323
CRTC3	0.035	0.503	0.127	0.565	0.016	0.287
CSDC2	0.005	0.186	0.006	0.182	0.006	0.068
CSK	0.906	0.914	0.027	0.426	0.283	0.637
CSRP2	0.005	0.409	0.048	0.511	0.185	0.598
CTNNAL1	0.128	0.672	0.043	0.500	0.178	1.315
CTNNB1	0.844	1.093	0.007	0.394	0.441	1.120
CTSC	0.958	1.057	0.231	0.689	0.038	0.416
CTXN1	0.117	0.447	0.012	0.080	0.045	0.105
CUTA	0.114	0.649	0.019	0.448	0.301	0.687
CXCR7	0.004	0.192	0.017	0.271	0.019	0.186
CXorf61	0.041	0.538	0.006	0.366	0.488	0.787
CXXC4	1.203	0.857	0.047	0.181	0.089	0.155
CXXC5	0.038	0.508	0.000	0.205	0.001	0.181
CYB5A	0.082	0.627	0.020	0.435	0.011	0.312
CYFIP2	0.024	0.331	0.038	0.313	0.036	0.208
CYP27C1	0.309	0.714	0.000	3.704	0.013	0.158
CYP51A1	0.023	0.517	0.031	0.475	0.023	0.378
CYSTM1	0.105	0.638	0.006	0.359	0.176	0.563
DAB1	0.000	0.046	0.000	0.000	0.000	0.006
DAB2	0.000	0.248	0.001	0.281	0.002	0.251
DAK	0.558	0.854	0.002	0.259	0.063	0.411
DARS2	0.231	0.748	0.005	0.368	0.237	0.629
DBI	0.028	0.557	0.003	0.363	0.003	0.280
DBN1	0.907	1.103	0.059	0.526	0.015	0.351
DBNDD1	0.026	0.347	0.005	0.127	0.028	0.161
DBNDD2	0.443	0.784	0.025	0.413	0.084	0.438

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

DBP	0.003	0.238	0.002	0.201	0.015	0.241
DCP2	0.090	0.538	0.409	0.724	0.026	0.208
DCPS	0.112	0.603	0.143	0.597	0.015	0.305
DCTN1	1.313	0.960	0.044	0.498	0.388	0.733
DCTPP1	0.114	0.643	0.475	0.816	0.010	0.334
DCXR	0.009	0.423	0.054	0.530	0.070	0.471
DDAH2	1.266	0.975	0.008	0.347	0.362	0.691
DDB2	0.826	1.074	0.008	0.389	0.312	0.675
DDIT4	0.179	0.706	0.033	0.490	0.003	0.281
DDN	0.045	0.541	0.092	0.543	0.000	0.138
DDR1	0.568	1.182	0.003	0.272	0.228	0.575
DDR2	0.936	1.080	0.030	0.370	0.003	0.130
DDX19B	0.399	0.784	0.261	0.696	0.047	0.396
DECR1	1.545	0.976	0.007	0.342	0.883	0.906
DEPDC1B	1.300	0.978	0.316	0.676	0.032	0.315
DERA	0.184	0.696	0.017	0.406	1.162	1.019
DHCR24	0.018	0.521	0.012	0.420	0.000	0.194
DHCR7	0.077	0.605	0.002	0.325	0.000	0.104
DHFR	1.167	0.938	0.000	0.226	0.019	0.350
DHODH	0.241	0.720	0.092	0.524	0.019	0.276
DHRS1	0.690	0.823	0.006	0.251	0.129	0.429
DHRS11	0.028	0.418	0.020	0.242	0.179	0.433
DHRS13	0.004	0.321	0.116	0.550	0.017	0.287
DHRS4	0.057	0.510	0.008	0.270	0.207	0.569
DHRS7	0.263	0.718	0.020	0.358	0.468	1.181
DHTKD1	0.091	0.573	0.028	0.312	0.202	0.482
DIXDC1	0.906	0.848	0.598	0.815	0.041	0.238
DLG3	1.171	0.860	0.027	0.300	0.041	0.233
DLGAP5	0.528	0.818	0.057	0.511	0.005	0.265
DMKN	0.242	0.720	0.002	0.284	0.104	0.488
DNAJC12	0.004	0.374	0.019	0.425	0.000	0.158
DNAJC19	0.010	0.409	0.018	0.394	0.038	0.375
DNASE2	1.320	0.952	0.043	0.459	0.026	0.332
DNM1	1.325	0.949	0.101	0.561	0.024	0.345
DNMT3A	0.548	0.825	0.050	0.420	0.010	0.185
DPM1	0.003	0.367	0.083	0.567	1.452	1.012
DPM3	0.054	0.546	0.006	0.379	0.068	0.463
DPP7	0.014	0.437	0.083	0.558	0.007	0.303
DPYSL2	1.377	0.961	0.015	0.395	0.005	0.240
DRAM2	0.040	0.478	0.289	0.697	0.243	0.587
DSCC1	0.585	0.835	0.121	0.580	0.015	0.315
DSEL	0.730	0.900	0.005	0.332	0.274	0.642
DSN1	0.698	0.829	0.012	0.377	1.729	1.005
DTD1	0.344	0.806	0.067	0.524	0.022	0.349
DUSP9	0.068	0.482	0.378	0.684	0.013	0.156
DUT	0.025	0.479	0.035	0.477	0.052	0.432
DYNLT1	0.707	0.826	0.000	0.244	0.738	1.096
E2F1	1.373	1.028	0.012	0.381	0.988	0.913
E2F2	0.155	0.566	0.045	0.342	0.030	0.193
E2F5	1.252	1.101	1.482	0.997	0.018	0.276
EBPL	0.038	0.512	0.139	0.620	0.001	0.216
ECEL1	0.000	0.027	0.000	0.186	0.000	0.069
ECH1	0.064	0.596	0.024	0.460	0.013	0.341
ECI1	0.101	0.611	0.004	0.352	0.142	0.548
ECI2	0.105	0.679	0.017	0.432	0.010	0.323
EEF1A2	1.004	1.061	0.064	0.544	0.006	0.318
EEF1E1	0.067	0.605	0.123	0.605	0.033	0.408
EEF2K	0.130	0.619	0.179	0.603	0.025	0.295
EFEMP1	0.101	1.423	0.077	0.555	0.000	0.199
EFNA1	0.003	0.312	0.058	0.491	0.320	0.679
EGFL7	0.032	0.410	0.643	1.141	0.264	0.567
EGFL8	0.030	0.242	0.128	0.440	0.224	0.402
EHBP1L1	0.146	0.662	0.008	0.364	0.178	0.559
EHMT2	0.177	0.720	0.087	0.563	0.015	0.353
EIF3B	1.438	0.941	0.019	0.454	0.478	0.788
EIF3C	0.613	0.852	0.113	0.604	0.010	0.344
EIF3K	0.113	0.692	0.169	0.655	0.018	0.380
EIF4A1	0.781	0.872	0.001	0.314	0.588	0.840
EIF4EBP1	0.155	0.684	0.030	0.488	0.005	0.309
ELF3	0.005	0.179	0.008	0.153	0.126	0.348
ELP2	0.092	0.657	0.039	0.485	0.068	0.456
EMC9	0.020	0.421	0.024	0.435	0.006	0.253
EMP2	0.023	0.487	0.006	0.304	0.011	0.242
ENG	0.629	1.176	1.699	0.970	0.005	0.227
ENO3	0.029	0.512	0.000	0.157	0.014	0.346
EPB41	0.012	0.405	0.042	0.436	0.003	0.172
EPB41L2	0.079	0.673	0.020	0.422	0.085	0.459
EPHA3	0.821	1.085	0.052	0.437	0.017	0.254
EPHB4	1.439	1.001	0.901	0.908	0.033	0.400
EPS8L2	1.416	0.953	0.024	0.308	0.166	1.413
ERCC5	0.106	0.575	0.028	0.386	0.280	0.614
ERGIC1	0.648	0.848	0.000	0.226	0.056	0.452
ERH	0.142	0.671	0.008	0.413	0.545	0.819
ERI3	0.212	0.724	0.084	0.563	0.038	0.414
ESPL1	0.809	1.058	0.867	1.043	0.029	0.362
ESRP2	0.261	0.657	0.008	0.166	0.020	0.134
ESYT1	0.814	1.063	0.188	0.657	0.025	0.378
ETAA1	0.111	0.567	0.011	0.294	0.139	0.466
ETFB	0.051	0.548	0.005	0.371	0.166	0.573
ETS2	0.003	0.349	0.006	0.362	0.055	0.429
EVL	0.407	0.801	0.007	0.339	0.001	0.150
EXOC4	1.189	1.013	0.075	0.531	0.002	0.216
EXOSC7	0.449	0.832	0.379	0.771	0.048	0.426
EXOSC8	0.048	0.550	0.017	0.418	0.162	0.548
EYA1	1.175	1.095	0.380	0.729	0.047	0.311
FABP5	0.157	0.685	0.265	0.721	0.049	0.443
FADS1	0.038	0.591	0.080	0.562	0.043	0.430
FADS2	1.536	1.009	0.054	0.460	0.034	0.351
FADS3	0.379	0.807	0.039	0.484	0.192	0.585
FAH	0.820	0.876	0.013	0.375	0.035	0.368
FAHD2A	0.147	0.668	0.006	0.369	0.123	0.522
FAHD2B	0.331	0.760	0.046	0.435	1.056	0.863
FAM101B	0.026	0.525	0.002	0.283	0.000	0.101
FAM102B	0.051	0.569	0.082	0.534	0.001	0.163
FAM123B	0.374	0.749	1.691	0.973	0.019	0.186

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

FAM13C	0.027	0.398	0.008	0.232	0.005	0.085
FAM162A	0.035	0.520	0.037	0.491	0.007	0.310
FAM165B	0.006	0.356	0.138	0.591	0.228	0.606
FAM175A	0.078	0.567	0.040	0.393	0.153	0.487
FAM195A	0.027	0.483	0.146	0.629	0.035	0.401
FAM213A	0.091	0.620	0.019	0.359	0.005	0.154
FAM43A	0.000	0.099	0.001	0.250	0.003	0.230
FAM46A	0.174	0.711	0.012	0.395	0.051	0.417
FAM64A	0.910	1.096	0.126	0.586	0.013	0.308
FAM69A	0.238	0.631	0.038	0.257	0.091	0.265
FAM81A	0.133	0.676	0.006	0.311	0.001	0.131
FAM86C1	0.437	0.804	0.044	0.432	0.372	0.683
FANCD2	0.791	1.167	0.450	0.783	0.045	0.365
FANCI	1.264	1.003	0.215	0.667	0.032	0.381
FANCL	0.845	0.874	0.028	0.440	0.511	0.784
FARP1	0.695	0.827	0.190	0.556	0.031	0.208
FASN	0.003	0.379	0.003	0.345	0.000	0.184
FAT1	0.303	0.780	0.287	0.718	0.013	0.328
FBL	0.161	0.703	0.142	0.630	0.027	0.397
FBN2	1.455	1.046	0.227	0.633	0.023	0.280
FBXL19	0.840	0.891	0.453	0.797	0.015	0.323
FBXO2	0.080	1.923	0.043	0.168	0.411	0.558
FBXO4	0.042	0.511	0.019	0.353	0.106	0.425
FBXW9	0.521	0.790	0.035	0.396	0.357	0.663
FCF1	0.012	0.435	1.656	0.958	0.917	0.914
FDFT1	0.075	0.656	0.000	0.214	0.003	0.263
FDPS	0.042	0.559	0.017	0.448	0.027	0.401
FGFR2	0.023	0.068	0.092	0.217	0.069	0.021
FGFR4	0.063	0.528	0.022	0.409	0.000	0.094
FIBIN	0.030	0.394	0.002	0.097	0.015	0.157
FIBP	0.031	0.513	0.002	0.333	0.102	0.516
FIGLA	0.020	0.392	0.598	1.103	0.343	0.654
FKBP11	0.028	0.491	0.057	0.523	0.016	0.336
FKBP3	0.027	0.521	0.012	0.414	0.168	0.576
FKBP4	0.804	0.847	0.888	1.039	0.047	0.430
FKBP9	1.078	1.053	0.012	0.393	0.059	0.430
FLYWCH2	0.192	0.721	0.012	0.419	0.014	0.356
FMNL1	0.602	1.181	0.027	0.347	0.548	1.181
FNDC3A	0.094	0.634	1.027	0.959	0.030	0.358
FOS	0.000	0.166	0.198	0.649	0.459	0.754
FOXM1	1.511	0.988	1.447	0.970	0.026	0.380
FOXO6	0.012	0.189	0.012	0.143	0.024	0.091
FOXP2	0.146	0.614	0.067	0.356	0.014	0.081
FOXRED2	0.553	0.845	0.302	0.712	0.047	0.373
FPGT	0.029	0.169	0.061	0.193	0.271	0.402
FSD1	0.862	0.881	0.037	0.470	0.010	0.288
FSTL3	0.023	0.482	0.092	0.567	0.956	0.892
FUOM	0.170	0.694	0.012	0.403	0.020	0.366
FUT10	0.130	0.627	0.584	0.782	0.043	0.334
FYN	0.022	0.441	0.003	0.273	0.010	0.253
G6PD	1.250	0.951	0.000	0.276	0.010	0.335
GABARAP	1.597	0.973	0.025	0.465	0.298	0.680
GALK1	0.051	0.536	0.002	0.294	0.030	0.386
GALNT7	0.764	0.848	0.034	0.454	0.011	0.296
GAMT	0.209	0.698	0.000	0.114	0.000	0.116
GART	0.183	0.731	0.173	0.642	0.034	0.397
GAS1	0.003	0.312	0.005	0.332	0.000	0.110
GAS6	0.242	0.657	0.034	0.311	0.514	0.744
GATSL2	0.003	0.188	0.093	0.459	0.033	0.261
GBAS	0.072	0.627	0.026	0.450	0.499	0.786
GCAT	0.026	0.414	0.247	0.656	0.064	0.373
GCDH	0.536	0.848	0.052	0.507	0.045	0.412
GCHFR	1.286	0.969	0.000	0.131	0.001	0.200
GCSH	0.251	0.746	0.129	0.610	0.001	0.207
GDF11	0.668	1.167	0.150	0.611	0.014	0.310
GDPD3	0.712	0.805	0.033	0.294	0.106	1.636
GEMIN5	0.953	0.893	0.346	0.750	0.033	0.384
GGCT	0.022	0.477	0.111	0.576	0.019	0.346
GINS2	0.105	0.624	0.045	0.509	0.009	0.323
GJA1	0.353	1.347	0.702	1.077	0.009	0.258
GKAP1	0.009	0.061	0.610	0.661	0.244	0.392
GLCE	0.092	0.610	0.019	0.359	0.056	0.362
GLP2R	0.027	0.431	0.052	0.418	0.039	0.308
GLT8D2	0.065	0.536	0.056	0.481	0.033	0.351
GMDS	0.816	1.024	0.484	0.796	0.022	0.255
GMPR2	0.967	1.105	0.005	0.346	0.203	0.592
GNAL	0.103	0.461	0.277	0.591	0.042	0.133
GNB4	0.165	0.733	0.138	0.597	0.046	0.394
GNG10	0.061	0.547	0.001	0.268	0.084	0.486
GOLM1	0.085	0.637	0.229	0.686	0.012	0.336
GOT1	0.531	0.853	0.350	0.754	0.049	0.421
GPC5	0.844	0.904	0.417	0.748	0.031	0.369
GPD1L	0.175	0.690	0.155	0.608	0.001	0.150
GPD2	1.199	0.903	0.142	0.596	0.029	0.351
GPI	0.276	0.778	0.253	0.709	0.024	0.394
GPR124	0.225	0.621	0.098	0.417	0.016	0.112
GPR126	0.839	1.123	0.027	0.356	0.025	0.258
GPR133	0.068	0.395	0.037	0.232	0.554	0.673
GPR37	0.149	0.634	0.000	0.158	0.018	0.276
GPRIN3	1.485	0.973	0.017	0.350	0.000	0.019
GPSM1	0.027	0.420	0.488	0.783	0.418	0.703
GPT	0.007	0.230	0.001	0.052	0.005	0.046
GPX4	0.217	0.729	0.036	0.498	0.082	0.496
GPX8	0.585	0.813	0.074	0.492	0.003	0.179
GRAMD1A	0.414	0.774	0.059	0.489	0.022	0.327
GRHPR	0.052	0.555	0.037	0.491	0.060	0.458
GRK6	0.197	0.700	0.020	0.432	0.027	0.371
GRTP1	0.103	0.511	0.024	0.287	0.027	0.216
GSN	0.010	1.942	0.039	0.413	0.144	1.405
GSTA4	0.018	0.205	0.036	0.233	0.029	0.090
GSTK1	0.508	0.836	0.010	0.406	0.137	0.546
GSTM4	0.757	0.873	0.230	0.653	0.025	0.281
GSTT1	0.119	0.570	0.003	0.233	0.389	0.707
GSTT2	0.956	0.945	0.027	0.404	0.713	0.902
GTPBP6	0.281	0.759	0.601	0.829	0.038	0.420

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

GXYLT2	0.594	0.768	0.056	0.341	0.043	0.206
GYLTL1B	0.024	0.333	0.009	0.217	0.003	0.048
HADH	0.084	0.637	0.161	0.633	0.000	0.173
HADHB	0.009	0.432	0.332	0.724	1.528	0.968
HAGHL	0.000	0.241	0.002	0.264	0.003	0.218
HAUS1	0.220	0.743	0.018	0.410	0.205	0.588
HAUS4	1.038	0.921	0.005	0.338	0.058	0.442
HAUS7	0.189	0.709	0.243	0.696	0.046	0.419
HCFC1R1	0.055	0.551	0.006	0.377	0.201	0.599
HDDC3	0.024	0.483	0.046	0.508	0.894	0.930
HDGFRP2	0.853	0.874	0.230	0.684	0.042	0.411
HEATR1	1.307	0.944	0.988	1.036	0.040	0.406
HEATR2	0.080	0.610	0.283	0.716	0.029	0.378
HES4	0.009	0.222	0.093	0.427	0.556	0.686
HEY1	0.006	0.205	0.028	0.286	0.061	0.263
HFE	0.058	0.497	0.071	0.460	0.019	0.228
HINT1	0.013	0.493	0.299	0.738	0.296	0.685
HINT2	0.057	0.543	0.004	0.338	0.159	0.581
HIST1H1B	0.017	0.468	0.397	0.789	0.001	0.247
HIST1H2AA	0.134	0.276	0.042	0.000	0.048	2.194
HIST1H2AB	0.115	0.646	0.124	0.622	0.002	0.258
HIST1H2AC	0.005	0.410	0.035	0.500	0.005	0.313
HIST1H2AD	0.018	0.488	0.469	1.101	0.201	0.650
HIST1H2AE	0.003	0.347	0.253	0.712	0.005	0.306
HIST1H2AG	0.000	0.249	0.164	0.656	0.005	0.309
HIST1H2AH	0.001	0.333	0.711	0.847	0.043	0.445
HIST1H2AI	0.041	0.585	0.411	0.765	0.029	0.407
HIST1H2AJ	0.004	0.416	0.805	0.871	0.016	0.367
HIST1H2AK	0.000	0.258	0.007	0.409	0.001	0.229
HIST1H2AL	0.000	0.282	0.158	0.647	0.001	0.221
HIST1H2AM	0.000	0.265	0.269	0.736	0.023	0.400
HIST1H2BA	0.039	0.262	0.172	0.600	0.902	0.878
HIST1H2BD	0.000	0.310	0.261	0.718	0.014	0.367
HIST1H2BE	0.003	0.286	0.099	0.536	0.033	0.351
HIST1H2BF	0.011	0.451	0.112	0.605	0.003	0.287
HIST1H2BG	0.269	0.770	0.404	0.790	0.033	0.418
HIST1H2BH	0.000	0.233	0.013	0.438	0.003	0.283
HIST1H2BI	0.003	0.365	0.074	0.567	0.000	0.213
HIST1H2BJ	0.001	0.338	0.192	0.673	0.004	0.300
HIST1H2BK	0.111	0.665	0.454	0.811	0.035	0.424
HIST1H2BL	0.002	0.359	0.054	0.535	0.001	0.228
HIST1H2BM	0.000	0.264	0.063	0.523	0.003	0.243
HIST1H2BN	0.001	0.332	0.819	0.898	0.016	0.375
HIST1H2BO	0.000	0.254	0.502	0.831	0.019	0.387
HIST1H3A	0.002	0.351	0.057	0.537	0.000	0.197
HIST1H3B	0.016	0.471	0.158	0.645	0.003	0.280
HIST1H3D	0.000	0.269	0.162	0.653	0.010	0.342
HIST1H3G	0.000	0.266	0.014	0.443	0.003	0.283
HIST1H3H	0.012	0.446	0.171	0.655	0.006	0.319
HIST1H3J	0.003	0.183	0.071	0.427	1.026	0.800
HIST1H4A	0.054	0.602	0.105	0.593	0.006	0.315
HIST1H4B	0.029	0.513	0.050	0.524	0.007	0.329
HIST1H4D	0.000	0.130	0.124	0.618	0.021	0.392
HIST1H4E	0.018	0.476	1.369	1.021	0.111	0.531
HIST1H4H	0.024	0.492	1.416	0.962	0.243	0.644
HIST1H4I	0.000	0.078	0.023	0.488	0.003	0.295
HIST1H4K	0.000	0.293	0.105	0.626	0.044	0.485
HIST2H2AB	0.020	0.477	0.350	0.767	0.009	0.338
HIST2H2AC	0.014	0.463	0.292	0.736	0.026	0.405
HIST2H2BE	0.068	0.601	0.523	0.836	0.001	0.236
HIST2H2BF	0.003	0.344	0.415	0.799	0.009	0.324
HIST2H3A	0.044	0.537	1.419	0.895	0.011	0.337
HIST2H3C	0.048	0.549	1.426	0.903	0.011	0.343
HIST2H3D	0.013	0.457	0.588	0.857	0.006	0.322
HIST2H4A	0.000	0.273	1.456	0.935	0.020	0.394
HIST2H4B	0.000	0.276	1.168	0.914	0.019	0.390
HIST3H2A	0.000	0.296	0.236	0.717	0.120	0.532
HIST3H2BB	0.000	0.310	0.042	0.506	0.032	0.411
HMBS	0.024	0.491	0.130	0.601	0.200	0.597
HMG20B	0.006	0.405	0.002	0.330	0.452	0.772
HMGB1	0.119	0.694	0.207	0.673	0.020	0.373
HMGB2	0.279	0.769	0.113	0.598	0.011	0.335
HMGCS1	0.005	0.426	0.013	0.405	0.128	0.527
HMGN1	0.068	0.613	0.004	0.366	0.003	0.287
HMGN3	0.008	0.440	0.002	0.322	0.000	0.171
HMGN5	0.240	0.713	0.000	0.042	0.003	0.150
HMMR	0.442	0.816	0.096	0.557	0.033	0.376
HORMAD1	1.090	1.026	0.186	0.582	0.040	0.321
HOXC11	0.310	0.766	1.047	0.911	0.014	0.302
HOXC4	0.290	0.601	0.042	0.162	0.087	0.145
HPD	0.000	0.187	0.000	0.033	0.000	0.033
HPDL	0.134	0.656	0.075	0.555	0.002	0.248
HPRT1	0.084	0.651	0.132	0.624	0.025	0.379
HR	0.156	0.683	0.000	0.221	0.000	0.106
HRSP12	0.052	0.539	0.155	0.622	0.006	0.279
HS6ST1	0.006	0.390	0.001	0.258	0.045	0.391
HS6ST2	1.348	1.041	0.769	0.870	0.013	0.303
HSCB	0.041	0.440	0.462	0.748	0.763	0.841
HSD17B4	0.467	0.835	0.071	0.531	0.019	0.346
HSD17B7	0.083	0.596	0.002	0.234	0.006	0.221
HSD17B8	0.012	0.395	0.001	0.224	0.440	1.208
HSPA4L	0.969	0.929	0.445	0.793	0.002	0.183
HSPB1	0.055	0.573	1.682	0.972	0.031	0.415
HSPD1	0.112	0.691	0.704	0.895	0.027	0.406
HSPE1	0.121	0.655	0.801	1.071	0.032	0.410
HSPG2	0.065	0.474	0.097	0.468	0.013	0.166
HTRA1	0.482	0.807	1.111	0.989	0.024	0.336
HTRA3	0.762	1.063	0.247	0.656	0.009	0.183
HYAL1	0.053	0.564	0.010	0.358	0.168	0.531
IARS2	0.051	0.631	0.022	0.455	0.070	0.469
IDE	0.170	0.686	0.202	0.626	0.019	0.287
IDH1	0.104	0.665	0.008	0.403	0.006	0.309
IDH2	0.025	0.492	0.045	0.490	0.096	0.473
IDI1	0.006	0.411	0.003	0.331	0.446	0.759
IFI27L1	0.111	0.645	0.002	0.302	0.324	0.695

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

IFI30	1.135	1.006	0.002	0.335	0.089	0.504
IFI44	0.461	0.729	0.016	0.200	0.016	0.102
IFIT1	1.277	0.883	0.012	0.157	0.010	0.023
IFITM1	0.015	0.466	0.094	0.573	0.000	0.186
IFITM3	0.303	0.738	1.132	1.020	0.003	0.279
IFT81	0.114	0.551	0.046	0.324	0.177	0.428
IGFBP2	0.214	1.299	0.007	0.372	0.002	0.220
IL17RC	0.026	0.341	0.300	0.646	0.270	0.549
IL18	1.017	1.087	0.002	0.197	0.362	0.688
IL27RA	0.149	0.627	0.012	0.320	0.020	0.271
IL32	0.493	0.833	0.059	0.549	0.032	0.403
ILVBL	0.389	0.806	0.090	0.566	0.007	0.301
IMPA2	0.003	0.364	0.001	0.278	0.001	0.224
IMPDH1	0.878	0.958	1.340	1.006	0.034	0.407
INCENP	0.836	0.899	0.057	0.517	0.033	0.394
INSIG1	0.004	0.367	0.007	0.347	0.281	0.658
INSL4	0.000	0.085	0.000	0.164	0.000	0.177
INTS3	0.114	0.676	0.088	0.558	0.027	0.378
IQGAP2	1.003	1.107	0.000	0.161	0.000	0.090
IRAK1BP1	0.184	0.589	0.109	0.456	0.022	0.137
IRX4	0.195	0.608	0.125	0.445	0.012	0.061
ISCA1	0.023	0.476	0.527	0.820	0.231	1.259
ISCA2	0.025	0.483	0.018	0.402	0.309	0.663
ISG15	0.024	0.474	0.001	0.290	0.052	0.445
ISOC2	0.187	0.701	0.173	0.654	0.001	0.220
ITGB3BP	0.226	0.722	0.026	0.435	0.019	0.337
ITPR1	1.254	1.029	0.437	0.787	0.027	0.348
JAG2	0.213	0.679	1.292	1.004	0.010	0.227
JMJD7	0.277	0.665	0.005	0.148	1.398	0.872
JPH1	0.028	0.493	0.010	0.336	0.000	0.092
JTB	0.034	0.522	0.005	0.378	0.014	0.361
JUND	0.004	0.372	0.002	0.307	0.107	0.519
KANK2	0.477	1.318	0.007	0.350	0.371	0.702
KANK4	0.000	0.108	0.000	0.043	0.000	0.020
KATNB1	0.275	0.755	0.034	0.463	0.005	0.274
KAZALD1	0.248	1.366	0.181	0.624	0.015	0.287
KBTBD7	0.101	0.572	0.028	0.407	0.099	0.432
KCNH2	0.075	0.510	0.003	0.178	0.002	0.036
KCNN2	0.025	0.338	0.055	0.354	0.014	0.122
KCNU1	0.017	0.109	0.017	0.049	0.056	0.071
KDELC2	1.314	0.952	0.006	0.240	0.316	0.625
KDELR3	0.064	0.521	0.007	0.304	0.040	0.340
KHDRBS3	0.000	0.216	0.001	0.219	0.000	0.128
KIAA0101	0.067	0.577	0.012	0.413	0.010	0.334
KIAA0146	0.741	0.872	0.338	0.718	0.048	0.367
KIAA0182	1.169	0.916	0.047	0.428	0.144	0.479
KIAA0391	0.325	0.754	0.031	0.423	0.228	0.583
KIAA0825	0.801	1.232	0.020	0.336	0.006	0.155
KIAA0922	1.089	1.072	0.377	0.736	0.025	0.272
KIAA1161	0.721	1.092	0.035	0.321	0.153	0.425
KIAA1211	0.085	0.548	0.037	0.375	0.006	0.147
KIAA1522	0.179	0.684	0.007	0.328	0.140	0.508
KIAA1598	1.370	0.989	0.045	0.414	0.210	0.538
KIAA1804	0.237	0.704	0.196	0.609	0.024	0.280
KIF11	0.218	0.721	0.300	0.708	0.049	0.390
KIF15	0.372	0.783	0.243	0.654	0.035	0.322
KIF1C	0.387	0.814	0.127	0.604	0.032	0.394
KIF20A	0.187	0.733	0.087	0.569	0.006	0.303
KIF26A	0.015	0.443	0.000	0.108	0.000	0.075
KIF26B	0.029	0.323	0.117	0.418	0.015	0.056
KLHDC3	0.415	0.786	0.139	0.626	0.014	0.353
KLHDC8B	0.268	0.700	0.275	0.666	0.033	0.285
KLHL13	0.021	0.495	0.006	0.329	0.000	0.099
KLHL5	0.821	1.053	0.025	0.316	0.465	0.717
KLRC4-KLRK1	0.038	0.483	0.000	0.063	0.002	0.132
KRT17	0.000	0.090	0.001	0.304	0.002	2.840
KRT8	0.726	0.939	0.002	0.328	1.218	1.072
L1CAM	0.414	0.810	0.037	0.465	0.485	0.778
LACTB	0.057	0.487	0.035	0.364	1.070	1.141
LAGE3	0.018	0.433	0.359	0.753	0.225	0.599
LAMA3	0.182	0.609	0.035	0.266	0.130	0.343
LAMA5	0.103	0.570	0.022	0.377	0.381	0.700
LAMB2	0.918	0.912	0.045	0.447	0.251	0.605
LAMC3	0.445	0.809	0.490	0.807	0.001	0.135
LANCL1	0.059	0.608	0.028	0.454	0.168	0.557
LAPTM4B	0.957	0.927	0.277	0.724	0.024	0.393
LBR	0.113	0.676	0.076	0.545	0.011	0.319
LCN10	0.003	0.258	0.014	0.325	0.000	0.056
LDHA	0.681	0.865	0.137	0.631	0.048	0.448
LDHB	0.107	0.690	0.006	0.399	0.002	0.273
LEPRE1	0.290	1.243	0.383	0.776	0.015	0.347
LEPREL2	0.647	1.171	0.293	0.724	0.002	0.206
LGALS3BP	1.312	1.019	0.134	0.622	0.000	0.202
LGR4	0.226	0.749	0.012	0.396	0.004	0.256
LHPP	0.108	0.519	0.020	0.260	0.060	0.274
LIG1	0.818	0.855	0.007	0.373	0.003	0.257
LITAF	0.084	0.616	0.044	0.498	0.171	0.575
LMAN1	0.352	0.759	0.105	0.585	0.046	0.428
LMF2	0.316	0.765	0.046	0.469	1.039	0.919
LMNB1	0.583	0.818	0.320	0.727	0.033	0.361
LMO1	0.026	0.452	0.000	0.120	0.003	0.160
LOC100127983	0.212	0.700	0.062	0.502	0.009	0.260
LOC100130921	0.266	0.483	0.040	0.196	0.025	0.046
LOC100287177	1.662	0.986	0.047	0.420	0.167	0.487
LOC100507035	0.003	0.089	0.172	0.627	0.083	0.337
LOC100996473	0.015	0.289	0.024	0.244	1.267	0.893
LOC643699	0.000	0.136	0.000	0.079	0.000	0.074
LOC728047	0.002	0.178	0.000	0.093	0.003	0.094
LONP1	0.496	0.813	0.173	0.649	0.049	0.439
LPCAT1	0.242	0.743	0.293	0.732	0.016	0.366
LPCAT3	0.689	0.872	0.003	0.293	0.080	0.444
LPCAT4	0.630	0.870	0.016	0.400	0.041	0.389
LPHN1	0.339	0.769	0.199	0.638	0.000	0.088
LPIN1	0.233	0.697	0.037	0.382	0.517	0.757
LPL	0.068	0.632	0.042	0.500	0.002	0.227

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

LRIG3	0.911	0.922	0.229	0.634	0.019	0.261
LRP5	0.237	1.271	0.410	0.787	0.022	0.367
LRRC16A	0.276	0.742	0.067	0.477	0.005	0.196
LRRC20	0.117	0.557	0.005	0.174	0.018	0.178
LRRC45	0.148	0.640	0.024	0.402	0.033	0.342
LRRC58	0.004	0.341	0.746	0.893	0.703	0.864
LSM10	0.007	0.407	0.435	0.800	0.327	0.698
LSM4	0.025	0.498	0.027	0.478	0.083	0.496
LSS	0.045	0.524	0.005	0.328	0.017	0.319
LTA4H	1.511	0.967	0.021	0.449	0.222	0.618
LUM	0.132	1.458	0.005	0.246	0.038	0.312
LUZP2	0.003	0.317	0.002	0.244	0.000	0.088
LY6E	1.316	0.949	0.015	0.440	0.015	0.363
LZTS2	1.494	0.958	0.704	0.845	0.047	0.381
MACROD1	0.066	0.574	0.005	0.368	0.001	0.205
MAD2L1	0.292	0.775	0.010	0.405	0.015	0.355
MAFK	0.004	0.368	0.279	0.714	0.888	1.048
MAGED2	1.432	0.981	0.047	0.499	0.227	0.613
MAN2B1	0.625	0.835	0.046	0.446	0.229	0.577
MAP2K6	0.022	0.440	0.000	0.112	0.000	0.027
MAP4	1.524	0.934	0.021	0.429	0.090	0.474
MAP4K2	1.446	1.035	0.167	0.607	0.036	0.340
MAP7	0.002	0.323	0.010	0.377	0.001	0.186
MAPK12	0.109	0.627	0.008	0.381	0.010	0.304
MAPK15	0.032	0.282	0.057	0.276	0.044	0.122
MAPK3	0.205	0.701	0.063	0.502	0.034	0.374
MARCH1	0.001	0.329	0.000	0.176	0.000	0.056
MARCKS	0.130	0.659	0.000	0.148	0.031	0.394
MATK	0.010	0.373	0.039	0.437	0.002	0.141
MBOAT1	0.160	0.603	0.111	0.514	0.037	0.267
MCAM	0.828	1.081	0.001	0.294	0.407	0.748
MCCC1	0.260	0.735	0.219	0.631	0.013	0.205
MCCC2	0.221	0.756	0.195	0.658	0.017	0.349
MCEE	0.008	0.343	0.137	0.577	0.469	0.754
MCF2L	0.275	0.749	0.055	0.480	0.009	0.255
MCM4	0.890	0.875	0.013	0.427	0.214	0.619
MCM6	0.448	0.803	0.040	0.485	0.243	0.631
MDH2	0.165	0.696	0.357	0.768	0.002	0.259
MED11	0.004	0.339	0.056	0.503	0.139	0.531
MEGF9	0.091	0.572	0.008	0.290	0.012	0.236
MEIS1	0.229	0.693	0.050	0.435	0.049	0.342
MEST	0.990	0.914	0.050	0.520	0.003	0.264
METRN	0.110	0.572	0.021	0.373	0.025	0.279
METRNL	0.003	0.340	0.039	0.490	0.296	0.675
METTL7A	0.077	0.619	0.000	0.135	0.000	0.050
METTL7B	0.608	1.154	0.024	0.205	0.063	0.205
MEX3A	1.370	1.029	0.967	1.077	0.014	0.160
MFAP2	0.005	0.408	0.000	0.173	0.000	0.033
MFAP4	0.012	0.369	0.000	0.139	0.000	0.031
MFSD12	0.018	0.469	0.015	0.434	0.919	0.885
MFSD3	0.056	0.555	0.002	0.291	0.008	0.305
MGST2	0.403	0.767	0.355	0.746	0.011	0.267
MID1IP1	0.060	0.550	0.237	0.687	0.049	0.423
MIEN1	0.071	0.580	0.007	0.390	0.089	0.496
MITD1	0.015	0.438	0.085	0.541	0.171	0.566
MKI67	1.391	1.024	0.186	0.627	0.046	0.366
MKX	1.727	0.996	0.039	1.614	0.019	0.253
MLF1IP	0.140	0.708	0.061	0.484	0.038	0.341
MLH1	0.840	0.884	0.026	0.437	0.825	0.910
MLLT1	0.910	0.947	0.522	0.829	0.021	0.364
MLLT6	0.839	1.120	0.192	0.636	0.029	0.348
MLST8	0.108	0.634	0.149	0.627	0.034	0.401
MMAB	0.026	0.426	0.021	0.355	0.013	0.231
MMACHC	0.092	0.623	0.122	0.590	0.010	0.273
MMP17	0.190	0.630	0.051	0.425	0.005	0.167
MMP2	0.001	2.390	0.027	0.447	1.120	1.032
MND1	0.793	0.858	0.014	0.377	0.136	0.472
MOCOS	0.068	0.610	0.100	0.585	0.009	0.325
MORN2	0.025	0.340	0.331	0.676	0.364	0.692
MOV10	0.208	0.730	0.017	0.412	0.002	0.214
MPG	0.129	0.651	0.003	0.342	0.545	0.813
MPI	0.421	0.830	0.164	0.631	0.041	0.407
MPLKIP	0.065	0.614	0.012	0.392	0.970	0.905
MPP2	0.495	0.795	0.092	0.509	0.011	0.231
MPST	0.012	0.413	0.002	0.305	0.005	0.262
MRC2	0.886	0.882	0.009	0.369	0.001	0.168
MRPL23	0.009	0.448	0.037	0.487	0.055	0.439
MRPL36	0.017	0.465	0.111	0.602	0.341	0.717
MRPL4	0.022	0.475	0.298	0.739	0.037	0.414
MRPS24	0.023	0.488	0.205	0.678	0.115	0.551
MRPS27	0.133	0.725	0.013	0.423	0.176	0.585
MRPS34	0.022	0.487	0.154	0.642	0.078	0.491
MRPS36	0.040	0.498	0.083	0.540	0.079	0.448
MRPS9	0.346	0.782	0.000	0.217	0.147	0.545
MSI2	0.238	0.717	0.138	0.593	0.009	0.269
MSMO1	0.017	0.468	0.053	0.482	0.094	0.464
MSRB2	0.049	0.485	0.012	0.298	0.147	0.465
MST4	0.067	0.614	0.094	0.566	0.027	0.380
MTA1	0.207	0.715	0.223	0.679	0.031	0.391
MTFMT	0.184	0.641	0.009	0.260	0.313	0.606
MTG1	0.242	0.727	0.040	0.479	0.473	0.773
MTHFD1	0.173	0.731	0.086	0.568	0.015	0.360
MTMR4	0.079	0.619	0.074	0.515	0.010	0.273
MTUS1	0.051	0.518	0.009	0.303	0.115	0.444
MUC13	0.746	1.200	0.019	1.878	0.043	0.224
MVD	0.032	0.463	0.036	0.429	0.332	0.647
MXD3	0.563	0.777	0.042	0.152	0.200	0.339
MXI1	0.581	0.849	1.315	0.940	0.013	0.240
MXRA7	0.657	0.837	0.035	0.454	0.160	0.544
MYB	1.312	0.968	0.065	0.260	0.046	0.082
MYBL2	0.233	0.762	0.105	0.591	0.005	0.301
MYC	0.201	0.727	0.758	0.910	0.003	0.282
MYO18A	1.463	0.936	0.031	0.412	0.528	0.784
MZT2B	0.007	0.405	0.043	0.501	0.039	0.417
NADSYN1	0.482	0.813	0.005	0.334	0.097	0.476

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

NAE1	0.014	0.467	0.066	0.537	0.206	0.602
NAGA	0.116	0.618	0.170	0.601	0.010	0.222
NAGLU	0.421	0.775	0.024	0.369	0.246	0.580
NAPRT1	0.052	0.485	0.007	0.324	0.074	0.409
NAT14	0.342	0.769	0.002	0.251	0.045	0.373
NBEAL2	0.222	0.701	0.056	0.461	0.004	0.184
NCAPD2	1.019	1.068	0.194	0.664	0.031	0.401
NCAPD3	1.150	0.923	0.390	0.772	0.048	0.413
NDRG2	0.166	0.581	0.484	0.749	0.026	0.135
NDST4	0.772	0.779	0.040	0.148	0.054	0.061
NDUFA1	0.022	0.481	0.027	0.486	0.573	0.834
NDUFA11	0.035	0.478	0.493	0.785	0.136	0.515
NDUFA12	0.431	0.811	0.001	0.303	0.329	0.703
NDUFA13	0.007	0.476	0.169	0.663	0.583	0.838
NDUFA7	0.007	0.464	0.119	0.605	0.388	0.722
NDUFAF2	0.110	0.671	0.025	0.476	0.080	0.487
NDUFB1	0.024	0.490	0.007	0.411	0.085	0.511
NDUFB5	0.416	0.818	0.006	0.380	0.417	0.749
NDUFS2	0.017	0.478	0.181	0.655	0.524	0.804
NDUFV2	0.004	0.396	0.243	0.703	0.223	0.625
NEIL2	0.576	0.802	1.429	0.938	0.015	0.251
NELF	0.231	0.729	0.007	0.389	0.000	0.139
NELL2	0.185	0.619	0.060	0.352	0.006	0.016
NFATC4	0.292	0.679	0.177	0.523	0.016	0.123
NFE2	0.032	0.121	0.049	0.112	0.130	0.153
NFIA	1.611	1.006	0.647	0.803	0.007	0.156
NFIB	0.265	0.730	0.084	0.493	0.006	0.192
NFIX	0.550	0.835	1.522	0.998	0.047	0.311
NFKBIB	0.035	0.485	0.276	1.200	1.220	0.931
NGFRAP1	0.372	0.834	0.001	0.311	0.001	0.230
NHP2	0.043	0.539	0.053	0.530	0.016	0.377
NHS	0.732	0.937	0.040	0.368	0.193	0.488
NINJ1	1.520	0.996	0.005	0.355	0.019	0.355
NIPSNAP1	0.146	0.698	0.000	0.208	0.011	0.309
NIPSNAP3A	0.023	0.453	0.003	0.256	0.056	0.370
NKX3-2	0.190	0.551	0.153	0.443	0.019	0.033
NLN	0.934	0.895	0.543	0.817	0.045	0.336
NME1	0.007	0.438	0.570	0.856	0.073	0.486
NME2	0.043	0.541	0.437	0.808	0.011	0.353
NME3	0.027	0.495	0.000	0.217	0.010	0.323
NME4	0.007	0.414	0.075	0.556	0.000	0.154
NNMT	0.017	1.839	1.742	0.999	0.004	0.272
NNT	0.173	0.731	0.017	0.435	0.004	0.277
NOLC1	0.115	0.696	0.586	0.859	0.041	0.432
NOTCH3	0.349	0.751	0.013	0.308	0.002	0.084
NPDC1	0.014	0.397	0.109	0.548	0.136	0.512
NPM1	0.047	0.601	0.006	0.400	0.047	0.448
NPM3	0.004	0.397	0.086	0.572	0.007	0.318
NPR3	0.021	0.513	0.064	0.522	0.000	0.156
NQO2	0.018	0.465	0.183	0.660	0.016	0.365
NR2F1	0.195	0.710	0.064	0.539	0.000	0.114
NR2F6	0.345	0.787	0.160	0.638	0.025	0.386
NR4A1	0.003	0.331	0.927	0.904	0.086	0.480
NRAS	0.589	0.852	0.028	0.458	0.571	0.823
NREP	0.085	0.599	0.000	0.185	0.033	0.358
NRG4	0.216	0.620	0.006	0.165	0.015	0.129
NRGN	0.282	0.653	0.009	0.183	0.027	0.164
NRM	0.529	0.784	0.049	0.487	0.022	0.337
NRSN2	1.637	0.984	0.014	0.378	0.332	0.649
NSMCE4A	0.049	0.543	0.023	0.442	0.091	0.485
NT5DC1	0.114	0.651	0.039	0.443	0.104	0.463
NT5DC2	0.714	0.875	0.008	0.404	0.015	0.366
NTAN1	0.026	0.506	0.024	0.446	0.081	0.460
NTHL1	0.023	0.473	0.048	0.501	0.066	0.453
NTPCR	0.358	0.786	0.000	0.239	0.075	0.467
NUCB1	1.365	0.954	0.516	0.833	0.049	0.433
NUCKS1	0.721	0.859	0.234	0.693	0.002	0.245
NUDCD2	0.042	0.545	0.017	0.432	0.050	0.431
NUDCD3	1.340	0.968	0.168	0.635	0.007	0.259
NUDT14	0.353	0.756	0.003	0.247	0.030	0.327
NUDT18	0.211	0.611	0.012	0.238	0.031	0.222
NUDT19	0.003	0.277	0.998	1.035	0.066	0.407
NUDT5	0.144	0.704	0.075	0.559	0.024	0.391
NUDT8	0.025	0.460	0.000	0.167	0.011	0.265
NUP210	0.255	0.761	0.000	0.243	0.000	0.115
NXN	0.407	1.250	1.296	0.936	0.031	0.355
NXPH4	0.776	1.143	0.725	1.114	0.043	0.217
OASL	0.178	0.624	0.002	0.161	0.333	0.626
OLFM1	0.000	0.189	0.000	0.251	0.000	0.064
OLFM2	0.044	0.406	0.037	0.308	0.022	0.165
OLFML2A	0.032	0.360	0.006	0.156	0.017	0.144
OSBPL10	0.737	1.160	0.083	0.538	0.019	0.344
OSBPL1A	0.135	0.674	0.060	0.456	0.014	0.247
OSTC	0.023	0.483	0.150	0.631	0.266	0.657
OXLD1	0.215	0.716	0.005	0.358	0.255	0.643
P2RX5	0.658	0.884	0.155	0.626	0.027	0.375
PAAF1	0.128	0.633	0.022	0.370	0.439	0.728
PABPN1	0.532	0.819	0.043	0.488	0.228	0.618
PACSIN3	0.065	0.598	0.122	0.604	0.013	0.343
PAF1	0.004	0.385	0.275	0.710	0.512	0.788
PAFAH1B3	0.111	0.631	0.063	0.533	0.005	0.286
PAICS	0.285	0.781	0.071	0.554	0.014	0.363
PALM	0.047	0.170	0.380	0.529	0.145	0.193
PARP1	1.110	0.929	0.265	0.716	0.034	0.416
PARVB	0.542	0.819	0.040	0.444	0.016	0.294
PBK	0.054	0.584	0.392	0.779	0.010	0.327
PBX2	0.286	0.777	0.908	0.906	0.012	0.309
PC	0.008	0.432	0.001	0.284	0.003	0.262
PCBD1	0.717	0.855	0.107	0.564	0.014	0.314
PCCB	0.079	0.642	0.306	0.737	0.028	0.382
PCDH17	0.781	1.263	0.160	0.496	0.049	0.211
PCK2	0.308	0.722	0.036	0.365	0.078	0.348
PCMTD2	0.827	0.899	0.027	0.363	0.202	0.528
PCNA	0.170	0.693	0.035	0.491	0.201	0.608
PCOLCE	0.018	0.485	0.008	0.406	0.000	0.052

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

PCOLCE2	1.212	0.943	0.592	0.784	0.021	0.329
PCSK9	0.012	0.449	0.006	0.369	0.000	0.118
PCYT2	0.157	0.636	0.006	0.295	0.844	0.850
PDCD4	0.146	0.657	0.080	0.507	0.023	0.313
PDE1A	0.009	0.403	0.008	0.322	0.000	0.052
PDE3A	1.226	0.890	0.309	0.736	0.011	0.332
PDE3B	0.196	0.694	0.037	0.437	0.002	0.157
PDE4B	1.422	0.946	0.528	0.692	0.027	0.073
PDE7B	0.808	0.879	0.127	0.524	0.014	0.198
PDE8B	0.132	0.605	0.095	0.453	0.009	0.125
PDF	0.025	0.358	0.167	0.538	0.116	0.364
PDGFRB	0.052	0.338	0.035	0.180	0.076	0.169
PDGFRL	0.634	0.825	0.000	0.176	0.001	0.123
PDLIM1	1.499	0.982	0.304	0.719	0.048	0.406
PDSS1	0.070	0.538	1.385	0.909	0.032	0.329
PECR	0.123	0.546	0.063	0.399	0.040	0.235
PEG10	0.019	0.520	0.000	0.222	0.000	0.158
PEMT	0.308	0.747	0.091	0.538	0.040	0.374
PET100	0.065	0.582	0.037	0.499	0.395	0.742
PEX11A	0.039	0.229	0.613	0.686	0.612	0.550
PFAS	0.281	0.779	0.540	0.839	0.010	0.323
PFKL	0.153	0.679	0.001	0.271	0.019	0.361
PFKM	0.883	0.877	0.313	0.741	0.006	0.314
PGAM4	0.004	0.198	1.574	0.989	0.483	0.727
PGM1	0.289	0.782	0.017	0.412	0.004	0.257
PHB2	0.009	0.442	0.355	0.767	0.030	0.411
PHF15	0.482	0.836	0.273	0.712	0.033	0.390
PHGDH	0.231	0.737	0.006	0.389	0.000	0.122
PHKB	0.222	0.719	0.015	0.389	0.086	0.455
PHPT1	0.017	0.498	0.077	0.556	0.079	0.486
PHTF1	0.373	0.759	0.014	0.293	0.094	0.381
PHYHD1	0.357	0.705	0.007	0.118	0.072	0.241
PI4K2B	0.727	0.892	0.029	0.374	0.526	0.724
PIF1	0.790	0.836	0.259	0.659	0.006	0.168
PIGB	0.043	0.479	0.823	0.827	0.585	0.719
PIGF	0.666	0.823	0.018	0.289	0.886	0.888
PIGP	0.511	0.704	0.025	0.245	0.388	1.178
PIGQ	0.049	0.483	0.095	0.542	0.419	0.732
PIGS	0.254	0.737	0.039	0.475	0.074	0.450
PIGY	0.822	0.847	0.043	0.491	0.368	1.158
PIH1D1	0.446	0.826	0.088	0.571	0.013	0.347
PIK3CD	0.276	0.701	0.047	0.390	0.900	0.863
PIK3R2	0.165	0.680	0.005	0.349	0.002	0.220
PIM1	0.023	0.391	0.018	0.300	0.225	0.525
PIN1	0.027	0.497	0.035	0.482	0.381	0.731
PIP4K2C	0.822	1.138	0.014	0.386	0.315	1.198
PIR	0.049	0.577	0.010	0.403	0.003	0.274
PITX1	0.025	0.481	0.192	0.659	0.176	0.573
PJA2	0.065	0.611	0.008	0.375	0.335	0.695
PKMYT1	0.764	0.854	0.105	0.518	0.047	0.332
PKN1	0.004	0.364	0.092	0.548	0.066	0.430
PKN3	0.132	0.649	0.007	0.359	0.000	0.135
PKP4	0.184	0.711	0.096	0.549	0.006	0.256
PLA2G12A	0.464	0.799	0.002	0.200	0.033	0.309
PLA2G3	0.036	0.270	0.009	0.092	0.013	0.000
PLA2G4A	0.097	0.665	0.222	0.683	0.003	0.277
PLAC8	0.591	1.167	0.263	0.662	0.039	0.306
PLCB3	0.916	0.947	0.003	0.316	0.033	0.391
PLEKHA2	0.780	1.140	0.355	0.759	0.003	0.247
PLEKHH3	0.652	1.142	0.043	0.473	0.005	0.246
PLEKHJ1	0.044	0.532	0.008	0.380	0.057	0.441
PLEKHO1	0.025	0.373	0.156	0.545	0.117	0.407
PLK1	0.639	0.866	0.879	0.907	0.014	0.359
PLOD3	0.476	0.835	0.299	0.734	0.024	0.384
PLXNB2	0.950	0.898	0.006	0.372	0.043	0.409
PLXND1	0.221	0.707	0.091	0.538	0.000	0.136
PMP22	0.106	0.647	0.115	0.598	0.005	0.303
PNRC2	0.004	0.389	0.695	0.888	0.313	0.685
PODXL2	0.760	0.882	0.114	0.599	0.005	0.290
POLA2	1.393	0.918	0.022	0.441	0.153	0.552
POLD1	0.079	0.596	0.007	0.383	0.014	0.346
POLE2	0.410	0.770	0.099	0.525	0.027	0.309
POLR2I	0.010	0.424	0.050	0.517	0.102	0.511
POLR3G	0.005	0.356	0.060	0.483	0.014	0.281
PPA2	0.234	0.751	0.083	0.561	0.030	0.393
PPIH	0.093	0.623	0.159	0.642	0.030	0.397
PPL	0.727	0.900	0.023	0.212	0.546	1.350
PPOX	0.092	0.601	0.001	0.207	0.198	0.549
PPP1R16A	0.021	0.431	0.005	0.320	0.013	0.300
PPP1R35	0.913	0.872	0.002	0.297	0.048	0.432
PPP2R5E	0.430	0.808	0.038	0.431	0.729	0.907
PPT2	0.265	0.768	0.438	0.796	0.022	0.360
PRADC1	0.115	0.657	0.048	0.498	0.172	0.562
PRC1	1.092	0.961	0.462	0.810	0.045	0.436
PRCP	0.610	0.847	0.355	0.754	0.014	0.333
PRDX5	0.070	0.590	0.014	0.440	0.443	1.115
PRDX6	0.300	0.803	0.047	0.520	0.043	0.438
PRIM1	0.008	0.423	0.037	0.472	0.029	0.380
PRKCSH	1.180	1.046	0.582	0.858	0.050	0.446
PRKCZ	0.442	0.807	0.005	0.285	0.159	0.528
PRKX	0.146	0.650	0.105	0.524	0.030	0.301
PRMT1	0.291	0.775	0.090	0.579	0.007	0.323
PRMT3	0.025	0.498	0.057	0.513	0.078	0.457
PRPS2	0.092	0.661	0.120	0.604	0.025	0.383
PRR11	0.189	0.700	0.095	0.525	0.015	0.274
PRR24	0.833	0.774	0.036	0.086	0.639	0.705
PRSS21	0.161	0.689	0.121	0.612	0.024	0.385
PRSS23	0.004	0.399	0.000	0.233	0.126	0.536
PRSS56	0.004	0.024	0.007	0.000	0.031	0.024
PRTFDC1	0.352	0.787	0.007	0.361	0.017	0.337
PSAT1	0.715	1.150	0.314	0.742	0.000	0.177
PSIP1	0.279	0.763	0.024	0.437	0.036	0.384
PSMA3	0.017	0.488	0.203	0.678	0.049	0.443
PSMB10	1.282	1.043	0.138	0.596	0.037	0.374
PSME1	0.037	0.531	0.348	0.764	0.032	0.414

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

PSMG1	0.048	0.580	0.234	0.688	0.011	0.331
PSMG4	0.042	0.522	0.092	0.554	0.034	0.374
PSPH	0.724	0.881	0.079	0.537	0.002	0.232
PSRC1	0.685	0.845	0.067	0.526	0.001	0.183
PTCH2	0.022	0.491	1.415	0.976	0.313	0.698
PTGFRN	0.854	1.151	0.146	1.319	0.027	0.325
PTMS	0.504	1.279	0.020	0.462	0.056	0.464
PTN	0.132	0.658	0.005	0.311	0.006	0.239
PTOV1	0.189	0.742	0.024	0.456	0.042	0.417
PTPMT1	0.022	0.440	0.207	0.662	0.176	0.556
PTPN13	0.034	1.770	0.143	0.534	0.026	0.232
PTPRS	1.343	0.925	0.124	0.595	0.036	0.393
PTTG1	0.025	0.497	0.220	0.689	0.033	0.413
PVRL2	0.632	1.204	0.679	0.797	0.045	0.341
PWP1	0.005	0.407	0.292	0.736	0.578	0.829
PXDN	1.540	1.011	0.486	0.820	0.011	0.325
PYCR1	0.101	0.636	0.052	0.520	0.002	0.229
PYGL	0.095	0.650	0.047	0.511	0.003	0.275
PYGM	0.000	0.051	0.000	0.055	0.000	0.045
QARS	0.238	0.736	0.007	0.387	0.011	0.335
QPCTL	0.106	0.619	0.022	0.436	0.031	0.373
QPRT	0.278	0.677	0.003	0.221	0.012	0.219
QTRT1	0.026	0.485	0.024	0.457	0.100	0.498
RAB26	0.004	0.300	0.000	0.048	0.000	0.048
RAB28	0.038	0.498	0.138	0.580	0.202	0.565
RAB30	0.293	0.764	0.008	0.345	0.075	0.437
RAB3D	0.259	0.733	0.021	0.382	0.004	0.183
RAB3IL1	0.041	0.452	0.389	0.713	0.202	0.541
RAB42	0.146	0.429	0.036	0.107	0.140	0.257
RAB8A	1.239	0.935	0.038	0.493	0.671	0.866
RABAC1	0.028	0.502	0.008	0.397	0.662	0.789
RABL6	0.130	0.664	0.127	0.614	0.010	0.334
RAC3	0.001	0.271	0.000	0.197	0.000	0.085
RAD54L	0.432	1.173	0.167	0.623	0.048	0.401
RASL10B	0.881	0.869	0.025	0.402	0.000	0.100
RAVER2	0.465	0.802	0.043	0.435	0.018	0.274
RBM14	0.081	0.562	0.035	0.434	0.177	0.545
RBM47	0.306	0.726	0.395	0.728	0.045	0.293
RCC2	0.854	0.891	0.241	0.700	0.034	0.416
RCN1	0.656	0.840	0.251	0.707	0.010	0.339
RCOR2	0.202	0.679	0.127	0.576	0.003	0.181
RECQL4	0.329	0.771	0.035	0.457	0.010	0.299
REEP4	0.525	0.849	0.623	0.866	0.045	0.422
REEP5	0.163	0.688	0.026	0.464	1.241	1.045
REEP6	0.032	0.313	0.029	0.290	0.048	0.208
RFK	0.046	0.575	0.008	0.354	0.071	0.428
RFX5	1.397	0.946	0.421	0.790	0.035	0.403
RHBDL1	0.039	0.362	0.003	0.086	0.032	0.183
RHOB	0.000	0.133	0.000	0.206	0.404	0.747
RHOBTB1	0.054	0.513	0.003	0.228	0.039	0.318
RHOBTB2	0.463	0.811	0.013	0.397	0.037	0.386
RHOT1	0.309	0.748	0.046	0.431	0.214	0.554
RHOV	0.966	0.935	0.019	0.260	0.431	0.605
RMI2	0.082	0.515	0.002	0.178	0.062	0.354
RNASEH1	0.169	0.670	0.016	0.404	0.229	0.611
RNASET2	0.232	0.686	0.277	0.672	0.012	0.264
RNF11	0.829	1.112	0.002	0.236	0.183	1.328
RNF130	0.968	1.125	0.415	0.753	0.037	0.358
RNF182	0.008	0.354	0.015	0.280	0.049	0.287
RNF212	0.169	0.662	0.023	0.410	0.020	0.317
ROBO1	0.032	0.534	0.130	0.597	0.004	0.254
ROMO1	0.004	0.468	0.212	0.686	0.350	1.159
ROR2	0.132	0.654	0.020	0.399	0.005	0.221
RORC	0.134	0.551	0.007	0.153	0.107	0.329
RPA3	0.042	0.502	0.002	0.273	0.059	0.428
RPIA	0.377	0.770	1.376	0.926	0.047	0.405
RPL14	0.021	0.480	0.227	0.694	0.149	0.569
RPL18A	0.017	0.493	0.384	0.782	0.101	0.521
RPL24	0.015	0.470	0.362	0.775	0.309	0.697
RPL26	0.018	0.473	0.229	0.697	0.184	0.599
RPL30	0.023	0.500	0.587	0.862	0.346	0.723
RPL36A	0.023	0.380	0.086	0.434	0.271	0.587
RPL36A-HNRNPH	0.011	0.391	0.015	0.423	0.002	0.211
RPL37	0.015	0.495	0.373	0.779	0.184	0.609
RPL37A	0.003	0.380	0.942	0.892	0.485	0.802
RPL39	0.007	0.419	0.478	0.821	0.214	0.622
RPL39L	0.311	1.359	0.041	0.498	0.014	0.341
RPL41	0.030	0.520	0.076	0.564	0.029	0.412
RPL7L1	0.357	0.799	0.037	0.494	0.453	0.772
RPRD1A	0.223	0.735	0.465	0.802	0.036	0.389
RPRM	0.000	0.122	0.003	0.252	0.000	0.049
RPS10	0.012	0.456	0.162	0.649	0.092	0.516
RPS12	0.003	0.352	0.473	0.821	0.276	0.673
RPS13	0.023	0.493	0.324	0.754	0.193	0.606
RPS15A	0.011	0.461	0.299	0.738	0.264	0.665
RPS23	0.118	0.697	0.096	0.595	0.014	0.364
RPS25	0.016	0.467	0.345	0.761	0.145	0.567
RPS27L	0.133	0.674	0.017	0.439	0.359	0.720
RPS6KA1	0.635	0.846	0.114	0.582	0.019	0.341
RPS7	0.017	0.468	0.267	0.722	0.109	0.531
RPSA	0.147	0.680	0.012	0.431	0.270	0.665
RSC1A1	0.025	0.479	0.389	0.717	0.210	0.530
RTF1	0.721	0.915	0.580	0.840	0.005	0.229
RWDD2B	0.108	0.625	0.005	0.326	0.056	0.406
S100A3	0.042	0.430	0.030	0.377	0.209	1.664
S100A4	0.152	0.700	0.000	0.269	0.045	0.442
SAMD1	0.068	0.543	1.210	0.930	0.013	0.291
SAMD11	0.000	0.151	0.075	0.529	0.000	0.116
SAMD5	0.021	0.437	0.214	0.631	0.010	0.213
SAP30	0.004	0.369	0.108	0.596	0.020	0.358
SAPCD2	0.108	0.621	0.045	0.490	0.000	0.168
SCAMP1	1.094	0.894	0.174	0.569	0.045	0.288
SCAND1	0.024	0.479	0.040	0.476	0.503	1.186
SCARA3	0.003	0.352	0.000	0.251	0.000	0.142
SCARA5	0.018	0.422	0.015	0.354	0.022	0.302

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

SCARB1	0.323	0.788	0.098	0.593	0.000	0.191
SCCPDH	0.000	0.283	0.002	0.316	0.010	0.328
SCD	0.012	0.493	0.000	0.199	0.000	0.162
SCD5	0.353	0.745	0.330	0.691	0.006	0.140
SCLY	0.061	0.541	0.102	0.557	0.014	0.297
SCMH1	0.639	0.831	0.070	0.492	0.005	0.215
SCN1B	0.299	0.721	0.111	0.506	0.007	0.116
SCRN2	0.323	0.709	0.039	0.411	0.017	0.269
SDC1	0.643	0.860	0.026	0.434	0.086	0.462
SEH1L	0.590	0.836	0.038	0.484	0.080	0.477
SELENBP1	0.101	0.630	0.000	0.198	0.005	0.288
SEMA3A	1.002	1.062	0.071	0.532	0.003	0.247
SEMA3B	0.270	0.722	0.025	0.350	0.020	2.115
SEPT5	0.122	0.625	0.013	0.341	0.002	0.156
SERPINF1	0.025	0.092	0.114	0.261	0.082	0.037
SESN3	0.005	0.409	0.027	0.452	0.004	0.247
SF3B14	0.011	0.452	0.025	0.472	0.096	0.508
SFXN2	0.658	0.806	0.175	0.556	0.013	0.091
SFXN3	1.091	1.045	0.016	0.418	0.294	0.665
SFXN4	0.370	0.802	1.178	1.035	0.013	0.330
SGPP1	0.013	0.456	0.006	0.346	0.039	0.389
SGSM3	0.316	0.771	0.010	0.356	0.246	0.606
SH3BGRL	0.811	0.928	0.003	0.267	0.065	0.396
SH3BP5	0.051	0.570	0.000	0.230	0.001	0.178
SH3GLB2	0.363	0.804	0.022	0.452	0.047	0.426
SH3PXD2B	0.790	0.844	0.041	0.442	0.006	0.218
SHMT1	0.250	0.754	0.069	0.526	0.023	0.358
SHMT2	0.551	0.861	0.150	0.633	0.012	0.343
SIAE	0.048	0.475	0.045	0.389	0.015	0.209
SIGMAR1	0.666	0.850	0.013	0.431	0.025	0.394
SIK1	0.001	0.337	0.043	0.503	0.003	0.271
SIVA1	0.008	0.438	0.097	0.593	0.277	0.667
SKP2	0.055	0.607	0.235	0.696	0.002	0.244
SLC12A7	0.700	0.903	0.508	0.821	0.013	0.315
SLC17A5	0.022	0.270	0.054	0.334	0.112	0.331
SLC17A9	1.183	1.057	0.853	0.853	0.025	0.282
SLC18B1	0.181	0.681	0.117	0.585	0.016	0.314
SLC19A1	0.690	0.858	0.119	1.350	0.032	0.355
SLC1A3	0.016	1.798	0.406	0.773	0.007	0.272
SLC22A18	0.244	0.614	0.034	0.313	0.239	0.522
SLC25A1	0.042	0.540	0.002	0.302	0.007	0.299
SLC25A10	0.248	0.744	0.074	0.552	0.005	0.296
SLC25A11	0.111	0.653	0.040	0.496	0.243	0.635
SLC25A13	0.795	0.864	0.074	0.538	0.039	0.401
SLC25A23	0.203	0.738	0.002	0.290	0.000	0.139
SLC25A29	0.049	0.507	0.015	0.378	0.010	0.266
SLC25A33	0.004	0.378	0.100	1.373	0.445	0.745
SLC25A39	0.262	0.753	0.046	0.513	0.012	0.350
SLC27A2	0.043	0.458	0.003	0.251	0.002	0.115
SLC27A5	0.004	0.310	0.129	0.583	0.001	0.151
SLC29A1	0.223	0.730	0.092	0.568	0.003	0.253
SLC29A2	0.771	1.181	0.256	0.657	0.005	0.154
SLC2A13	0.067	0.514	0.077	0.453	0.028	0.255
SLC2A4RG	0.673	0.899	0.012	0.417	0.008	0.318
SLC37A4	0.068	0.548	0.087	0.533	0.011	0.273
SLC38A10	0.727	0.890	0.023	0.395	0.098	0.440
SLC38A5	0.194	0.702	0.412	0.792	0.037	0.405
SLC39A6	0.088	0.663	0.321	0.744	0.022	0.380
SLC39A9	1.257	1.012	0.012	0.390	0.252	1.234
SLC44A2	0.501	0.818	0.007	0.301	0.002	0.131
SLC4A2	0.528	0.855	0.051	0.521	0.020	0.378
SLC50A1	0.043	0.570	0.002	0.319	0.095	0.497
SLC5A6	1.169	0.928	0.486	1.129	0.041	0.387
SLC7A2	0.026	0.541	0.280	0.715	0.033	0.392
SLC9A3R1	0.086	0.610	0.012	0.421	0.131	0.547
SLC9A3R2	0.013	0.405	0.079	0.531	0.130	1.407
SLIRP	0.066	0.585	0.034	0.491	0.171	0.590
SLX1A	0.017	0.418	0.025	0.437	0.228	0.615
SLX1B	0.019	0.427	0.020	0.413	0.278	0.650
SMARCA1	0.160	0.646	0.006	0.257	0.010	0.202
SMARCA4	0.682	0.866	0.579	0.853	0.016	0.349
SMARCC1	0.632	0.867	0.256	0.700	0.034	0.387
SMARCC2	0.524	0.822	0.664	0.837	0.025	0.355
SMARCD3	0.084	0.537	1.064	0.883	0.044	0.344
SMIM1	0.077	0.416	0.003	0.113	0.003	0.000
SMN1	0.023	0.494	1.459	0.918	0.773	0.817
SMOC1	1.244	0.957	1.623	0.963	0.003	0.240
SMPD2	0.377	0.771	0.153	0.574	0.049	0.313
SNCA	1.343	0.940	0.014	0.283	0.010	0.147
SNCG	1.411	0.982	0.009	0.387	0.131	0.540
SNRNP25	0.604	0.825	0.046	0.505	0.050	0.433
SNRNP40	0.537	0.825	0.028	0.478	0.289	0.677
SNRPB	0.003	0.358	0.624	0.872	0.804	0.909
SNRPF	0.018	0.454	0.689	0.886	0.125	0.533
SNRPN	0.062	0.575	0.021	0.443	0.038	0.402
SNTA1	0.025	0.475	0.033	0.439	0.014	0.303
SNX5	0.684	0.851	0.483	0.810	0.021	0.354
SOAT1	0.078	0.590	0.060	0.482	0.029	0.326
SOD1	0.006	0.414	0.164	0.651	0.233	0.636
SORD	0.140	0.680	0.092	0.574	0.001	0.216
SOX12	0.275	0.740	0.177	0.624	0.002	0.176
SPAG5	0.977	1.084	0.365	0.761	0.031	0.394
SPATA24	0.134	0.718	0.024	0.354	0.013	0.239
SPC24	0.057	0.621	0.003	0.342	0.005	0.282
SPC25	0.121	0.660	0.856	0.879	0.023	0.285
SPCS3	0.184	0.732	0.010	0.398	0.267	0.649
SPG21	0.344	0.804	0.240	0.689	0.035	0.402
SPINK13	0.025	0.000	0.095	0.178	0.286	0.366
SPINK4	0.907	1.035	0.000	0.111	0.005	2.663
SPOCK3	0.033	0.263	0.039	0.184	0.037	0.055
SPR	0.292	0.739	1.213	0.919	0.040	0.417
SPTBN2	0.308	0.766	0.022	0.426	0.001	0.181
SPTLC3	0.276	0.685	0.026	0.242	0.089	0.282
SPTSSA	0.003	0.347	0.010	0.382	0.037	0.386
SQLE	0.025	0.497	0.007	0.351	0.022	0.338

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

SREBF1	0.068	0.579	0.001	0.292	0.000	0.160
SRI	0.004	0.371	0.058	0.519	0.201	0.598
SRSF8	1.485	0.914	0.022	0.397	0.229	0.588
SSB	0.069	0.635	0.060	0.533	0.040	0.421
SSBP4	0.023	0.437	0.075	0.512	0.013	0.290
SSH3	1.111	1.054	0.018	0.308	0.176	0.493
ST3GAL4	0.203	0.709	0.042	0.485	0.002	0.218
ST6GALNAC4	0.226	0.748	0.046	0.519	0.014	0.353
ST6GALNAC6	0.310	0.766	0.006	0.357	0.002	0.220
STAC	1.301	0.986	0.181	0.642	0.044	0.362
STAC2	0.018	0.105	0.047	0.147	0.060	0.050
STARD4	0.068	0.546	0.007	0.302	0.249	0.592
STK25	0.333	0.790	0.024	0.459	0.027	0.388
STMN1	0.339	0.797	0.003	0.330	0.013	0.344
STOX2	0.037	0.414	0.109	0.466	0.139	0.410
STRA13	0.007	0.436	0.054	0.532	0.047	0.444
STRA6	0.502	0.758	0.006	0.178	1.691	0.958
STT3A	1.243	1.021	0.191	0.659	0.041	0.420
STUB1	0.186	0.702	0.019	0.452	0.063	0.470
STX10	0.657	0.866	0.017	0.431	0.074	0.469
SUCLG2	0.018	0.472	0.025	0.423	0.262	0.632
SULF2	0.094	1.470	0.096	0.543	0.011	0.276
SULT1A3	1.280	0.951	0.206	0.611	0.031	0.317
SULT1A4	1.312	1.015	0.037	0.386	0.087	0.399
SUMO3	0.021	0.467	0.869	1.066	0.173	0.575
SURF1	0.061	0.527	0.014	0.375	0.222	0.585
SVIP	0.216	0.701	0.042	0.450	0.387	0.707
SWI5	0.118	0.625	0.035	0.497	0.852	0.872
SYBU	0.650	1.200	0.096	0.435	0.043	0.231
SYNC	0.146	0.557	0.044	0.285	0.020	2.377
T	0.005	0.370	0.003	0.310	0.000	0.071
TACC2	0.822	0.879	0.039	0.316	0.034	0.202
TADA3	0.939	0.906	0.045	0.497	0.982	0.911
TAF12	0.037	0.497	1.258	1.011	1.053	1.031
TAF7L	0.340	0.648	0.367	0.632	0.032	0.138
TAP2	0.841	1.137	0.119	1.351	0.042	0.383
TAZ	0.042	0.498	0.321	0.709	0.370	0.693
TBC1D16	0.839	1.140	0.371	0.747	0.010	0.252
TBC1D4	1.584	0.953	0.087	0.534	0.013	0.300
TBC1D7	0.995	1.109	0.011	0.300	0.857	0.889
TBCD	0.910	0.923	0.093	0.552	0.010	0.293
TBXA2R	0.058	0.462	0.005	0.194	0.058	0.299
TCEA1	1.152	0.938	0.049	0.500	0.073	0.455
TCF12	0.097	0.663	0.098	0.582	0.006	0.298
TCF25	1.215	1.011	0.097	0.572	0.013	0.338
TCF3	0.146	0.663	0.483	0.817	0.016	0.348
TCFL5	0.047	0.523	0.052	0.496	0.044	0.399
TCIRG1	0.507	0.804	0.020	0.406	0.304	0.658
TCOF1	0.825	1.051	0.228	1.226	0.034	0.381
TDP1	0.466	0.814	0.014	0.361	0.145	0.503
TEAD2	0.533	0.852	0.072	0.532	0.002	0.214
TECR	1.374	1.016	0.054	0.529	0.035	0.419
TENC1	0.198	1.347	0.001	0.228	0.012	0.279
TFAP4	1.441	0.935	0.241	1.219	0.015	0.309
TFR2	0.344	0.739	0.207	0.604	0.013	0.197
TGIF2	0.851	0.982	0.040	0.412	0.002	0.120
THEM6	0.044	0.493	0.113	0.557	0.042	0.373
THRA	0.398	0.786	0.006	0.329	0.001	0.139
THYN1	0.211	0.698	0.090	0.543	0.041	0.388
TIFA	0.441	0.728	0.040	0.311	0.394	0.651
TK1	1.221	0.920	0.015	0.437	0.031	0.404
TLCD1	0.084	0.577	0.046	0.486	0.002	0.196
TMA7	0.039	0.536	0.004	0.371	0.075	0.486
TMBIM4	0.019	0.384	0.033	0.400	0.133	0.480
TMED10	0.018	0.502	0.135	0.614	0.040	0.413
TMEM102	0.330	0.720	0.033	0.354	0.018	0.208
TMEM106C	1.064	0.883	0.019	0.457	0.203	0.609
TMEM120A	0.013	0.427	0.027	0.439	0.093	0.468
TMEM126B	0.065	0.602	0.042	0.497	0.118	0.522
TMEM129	0.857	0.943	0.038	0.440	0.192	0.548
TMEM144	1.274	0.840	0.036	0.129	0.071	0.115
TMEM14A	0.084	0.593	0.002	0.275	0.076	0.444
TMEM160	0.194	0.708	0.047	0.488	0.060	0.430
TMEM161A	0.137	0.653	0.051	0.510	0.010	0.311
TMEM187	0.047	0.429	0.007	0.194	0.323	0.607
TMEM205	0.039	0.543	0.006	0.391	0.003	0.269
TMEM219	0.148	0.692	0.045	0.490	0.289	0.685
TMEM223	0.061	0.567	0.024	0.464	0.256	0.657
TMEM231	0.082	0.549	0.086	0.472	0.032	0.287
TMEM42	0.007	0.383	0.047	0.461	1.262	1.060
TMEM45A	0.169	0.658	0.023	0.399	0.101	0.459
TMEM48	0.275	0.769	0.427	0.790	0.013	0.323
TMEM60	0.079	0.554	0.005	0.270	1.051	1.102
TMEM97	0.821	0.888	0.096	1.423	0.041	0.324
TMTC2	0.533	0.787	0.129	0.507	0.014	0.165
TMTC4	0.260	0.725	0.003	0.201	0.013	0.200
TNFAIP8	0.565	1.215	0.045	0.362	1.357	1.042
TNFRSF11B	0.915	0.870	0.000	4.943	0.016	0.172
TNNC1	0.210	0.551	0.115	0.293	0.048	0.000
TNNT1	0.474	1.268	0.016	0.392	0.137	0.515
TNS3	0.924	0.904	0.126	0.566	0.010	0.253
TOB1	0.015	0.468	0.160	0.640	0.319	0.690
TOP1MT	0.356	0.788	1.496	1.018	0.040	0.399
TOP2A	0.628	0.826	0.235	0.696	0.005	0.296
TOR3A	1.504	0.962	0.289	0.732	0.018	0.342
TP53	0.249	1.358	0.068	0.498	0.005	0.228
TPD52L1	0.625	0.814	0.011	0.337	0.003	0.179
TPGS2	0.634	0.856	0.008	0.391	0.022	0.371
TPMT	0.811	0.853	0.034	0.406	0.307	0.633
TRABD2A	0.452	0.833	0.063	0.524	0.000	0.134
TRADD	0.075	0.515	0.005	0.168	0.911	0.834
TRAF7	0.434	0.822	0.025	0.462	0.077	0.477
TRAP1	0.106	0.672	0.202	0.677	0.007	0.329
TRAPPC6A	0.044	0.393	0.019	0.227	0.698	0.790
TREX1	1.658	0.982	0.013	0.369	0.271	0.620

Table S1B. The list of genes that were downregulated by Rtf1, Paf1, or Ski8 knockdown (cont.)

TRIAP1	0.107	0.637	0.004	0.357	0.857	1.025
TRIM14	1.075	0.928	0.681	0.820	0.019	0.247
TRIM62	0.264	0.633	0.047	0.330	0.081	0.294
TRIM65	0.455	0.817	0.113	0.566	0.049	0.401
TRIML2	0.137	0.581	0.042	0.352	0.270	1.293
TRIQK	0.027	0.339	0.037	0.267	0.106	0.308
TRMT61B	0.062	0.545	0.047	0.453	0.610	0.782
TROAP	0.907	0.933	0.155	0.634	0.013	0.348
TRPT1	1.084	1.047	0.023	0.439	0.585	0.785
TSC22D3	0.037	0.404	0.151	0.537	0.205	0.504
TSEN15	0.082	0.604	0.035	0.447	0.357	0.689
TSPAN13	0.058	0.447	0.014	0.248	0.398	0.638
TSPAN6	0.119	0.649	0.020	0.420	0.059	0.426
TSPAN8	0.008	2.584	0.021	0.000	0.146	0.205
TSPO	0.072	0.605	0.044	0.505	0.033	0.395
TSPYL4	0.450	0.739	0.013	0.310	1.600	0.946
TST	0.871	0.887	0.022	0.406	1.422	1.024
TSTA3	0.168	0.694	0.002	0.311	0.025	0.392
TTC13	0.214	0.668	0.113	0.519	0.033	0.305
TTC39C	0.137	0.610	0.064	0.442	0.012	0.196
TTF2	1.289	0.982	0.310	0.723	0.024	0.346
TTK	1.502	0.984	1.591	0.991	0.030	0.372
TTLL12	0.383	0.803	0.460	0.810	0.040	0.419
TTYH3	0.305	0.766	0.002	0.284	0.000	0.137
TUBA1A	0.485	0.843	0.014	0.426	0.389	0.736
TUBB	1.126	1.035	0.089	0.579	0.037	0.429
TUSC3	0.105	0.648	0.076	0.552	0.008	0.315
TWF2	0.299	0.771	0.008	0.396	0.033	0.405
TWIST1	0.149	0.614	0.125	0.508	0.020	0.177
TWIST2	0.049	0.489	0.610	0.818	0.125	0.486
TXNL4A	0.134	0.664	0.000	0.112	0.146	0.555
TYMP	0.004	0.306	1.157	0.884	0.094	0.424
TYMS	0.134	0.702	0.028	0.478	0.002	0.245
U2SURP	1.312	0.931	0.044	0.479	0.252	0.628
UAP1	0.247	0.758	0.035	0.476	0.559	1.174
UAP1L1	0.025	0.412	0.076	0.451	0.252	0.571
UBAC1	0.271	0.766	0.227	0.687	0.049	0.439
UBL5	0.004	0.444	0.783	0.857	1.531	0.979
UBQLN4	0.023	0.495	0.287	1.188	0.167	0.574
UCP2	1.567	0.980	0.010	0.384	0.045	0.398
UGP2	1.302	0.977	0.041	0.488	0.934	0.934
UGT8	0.309	0.767	0.096	0.511	0.004	0.178
UHRF1	0.607	0.871	0.031	0.468	0.250	0.635
ULK1	0.700	0.810	0.046	0.410	0.065	1.680
UNC13B	0.079	0.614	0.101	0.560	0.025	0.353
UNC93B1	0.281	0.683	0.272	1.254	0.030	0.315
UNG	0.811	0.865	0.840	0.856	0.027	0.372
UQCR11	0.022	0.492	0.067	0.545	0.293	0.683
UQCRB	0.066	0.628	0.047	0.496	0.083	0.471
UQCRQ	0.014	0.473	0.180	0.660	0.187	0.597
UROD	0.442	0.828	0.015	0.434	0.364	0.721
UROS	0.105	0.605	0.007	0.363	0.083	0.466
USH1C	0.000	0.153	0.000	0.052	0.000	0.038
USMG5	0.010	0.431	0.106	0.595	0.144	0.564
USP28	0.006	0.299	0.495	0.786	0.115	0.420
UST	0.050	0.509	0.005	0.253	0.002	0.113
VARS	0.068	0.609	0.005	0.372	0.010	0.331
VAV3	0.095	0.617	0.002	0.238	0.002	0.144
VEGFB	1.171	0.957	0.000	0.126	0.045	0.320
VSIG10	0.809	0.913	0.246	0.558	0.049	0.169
VWA5A	0.150	0.645	0.017	0.350	0.358	0.676
WDR34	0.105	0.628	0.014	0.425	0.108	0.513
WDR54	0.233	0.759	0.015	0.417	0.916	0.868
WDR59	0.756	0.866	0.840	0.883	0.032	0.341
WDR61	0.079	0.602	0.000	0.078	0.161	0.563
WDR76	1.149	0.938	0.044	0.407	0.698	0.847
WFDC1	0.741	1.100	0.887	0.848	0.020	0.233
WNT10B	0.240	0.712	0.797	0.879	0.005	0.239
WRB	0.008	0.349	0.051	0.446	0.172	0.522
XXYLT1	0.208	0.716	0.130	0.593	0.007	0.273
YDJC	0.041	0.525	0.108	0.585	0.045	0.425
ZBED3	0.163	0.601	0.009	0.279	0.014	0.207
ZBTB42	0.028	0.354	0.116	0.481	0.025	0.201
ZCCHC24	0.195	0.695	0.040	0.456	0.297	0.648
ZDHHC2	0.051	0.525	0.005	0.298	0.018	0.296
ZDHHC24	0.059	0.563	0.008	0.366	0.125	0.506
ZMYM3	0.353	0.782	0.062	0.485	0.015	0.287
ZNF556	0.241	0.706	0.461	0.770	0.007	0.144
ZNF76	0.963	0.912	0.010	0.338	0.408	0.712
ZNF883	1.351	0.943	0.225	0.607	0.019	0.224
ZNHIT1	0.024	0.500	0.362	0.767	0.523	1.087
ZP3	0.003	0.294	0.238	0.672	0.198	0.557
ZRANB2	0.733	0.865	0.038	0.484	0.245	0.630