Cell, Vol. 92, 307–313, February 6, 1998, Copyright ©1998 by Cell Press

Eukaryotic Transcription: An Interlaced Network of Transcription Factors and Chromatin-Modifying Machines

Review

James T. Kadonaga
Department of Biology, 0347
and Center for Molecular Genetics
University of California, San Diego
La Jolla, California 92093-0347

Introduction

Over the past several years, there have been many exciting advances in our understanding of the role of chromatin structure in the regulation of transcription by RNA polymerase II. It has been known that the packaging of genes into chromatin represses basal transcription and that transcriptional activators function, at least in part, to counteract chromatin-mediated repression. More recently, substantial effort has been devoted toward illuminating the mechanisms by which transcription factors function in the context of the repressed chromatin template. The objective of this review is to provide an overview of some current topics in the area of chromatin and transcription and then to discuss a few issues that pertain to chromatin remodeling machines. This essay is written for a general readership (i.e., nonspecialists). For more detailed information, it would be useful to consult other recent reviews in this area (for example, see Peterson and Tamkun, 1995; Brownell and Allis, 1996; Felsenfeld, 1996; Kingston et al., 1996; Peterson, 1996; Wolffe and Pruss, 1996; Grunstein, 1997; Hartzog and Winston, 1997; Pazin and Kadonaga, 1997a, 1997b; Tsukiyama and Wu, 1997).

A Network of Proposed Linkages between Chromatin and Transcription

An array of connections that have been suggested by experiments described in this review is depicted in Figure 1. It should be noted that there is not universal agreement with regard to some of these postulated linkages. Where differences exist, I have attempted to present alternate viewpoints. In addition, this figure is not meant to be comprehensive. The aim of the first part of this review is to provide a brief description of each of these different categories of factors and explanations for the connectivities that are outlined in Figure 1. In this manner, the figure will be used as a device, or framework, with which specific topics will be discussed in relation to one another.

Chromatin Remodeling Machines—Links to Chromatin and Transcription

These multisubunit molecular machines include the yeast SWI/SNF complex, yeast RSC complex, *Drosophila* NURF, *Drosophila* CHRAC, *Drosophila* ACF, *Drosophila* BRM complex, and mammalian BRG1- or hbrm-associated complexes (reviewed in Peterson and Tamkun, 1995; Kingston et al., 1996; Peterson, 1996; Hartzog and Winston, 1997; Pazin and Kadonaga, 1997a; Tsukiyama and Wu, 1997). These different protein complexes are placed in the same general category because they each contain a closely related subunit (SWI2/SNF2 in the yeast SWI/SNF complex; STH1 in RSC; ISWI in NURF, CHRAC, and ACF; *Drosophila* BRM; and mammalian BRG1 or hbrm) that is a member of a family of ribonucleoside triphosphate (NTP)–binding proteins (reviewed in Eisen et al., 1995). The presence of this NTP-binding subunit in the different factors suggests that it may carry out a related biochemical function in each of the complexes. For instance, it has been postulated that the NTP-binding subunit might act as a processive, ATP-driven DNA-translocating motor that disrupts histone–DNA interactions (Kornberg and Lorch, 1995; Pazin and Kadonaga, 1997a).

The yeast SWI/SNF complex provides an example of the connectivity of these complexes with chromatin and transcription, as shown in Figure 1. This protein complex has an apparent mass of about 2 MDa and comprises an estimated 11 polypeptides, which include SWI1/ADR6, SWI2/SNF2, SWI3, SNF5, SNF6, SNF11, TFG3/ANC1, and SWP73. Several of the genes that encode subunits of the yeast SWI/SNF complex were originally identified on the basis of their requirement for normal transcriptional activity of some but not all genes. A relation between the SWI/SNF complex and chromatin was then suggested by genetic studies (see, for instance, Hirschhorn et al., 1992; Kruger et al., 1995) as well as by biochemical experiments in which it was found that purified SWI/SNF complex is able to induce perturbation of histone–DNA interactions in a mononucleosome as well as to facilitate binding of GAL4-AH (a sequence-specific DNA-binding protein) to a mononucleosome (Côté et al., 1994). In the latter part of this review, chromatin remodeling machines will be discussed further.

RNA Polymerase II Complex (Holoenzyme)—Links to Transcription and Chromatin Remodeling Machines

Based on genetic and biochemical studies, it has been proposed that a fraction of the RNA polymerase II exists as a large complex that is commonly termed the "RNA polymerase II holoenzyme" (reviewed in Koleske and Young, 1995; Kingston et al., 1996; Orphanides et al., 1996; Struhl, 1996; Ptashne and Gann, 1997). This RNA polymerase II holoenzyme is a large protein conglomerate that contains "core" RNA polymerase II (the multiprotein enzyme that had traditionally been designated as "RNA polymerase II"), a subset of the basal transcription factors (such as TFIIB, TFIIE, TFIIF, and/or TFIIH), nine SRB (**s**uppressor of **R**NA polymerase **B**) proteins, as well as other known (such as GAL11, SIN4, RGR1, and ROX3) and unknown proteins.

A related complex, termed the mediator, was identified and purified on the basis of its requirement to achieve transcriptional activation in vitro (reviewed in Björklund and Kim, 1996). The mediator was found to contain SRB proteins as well as GAL11, SIN4, RGR1, and ROX3. It appears that a complex of the mediator and core RNA polymerase II, via an interaction between the mediator and the C-terminal domain of the largest subunit of the polymerase, is approximately equivalent

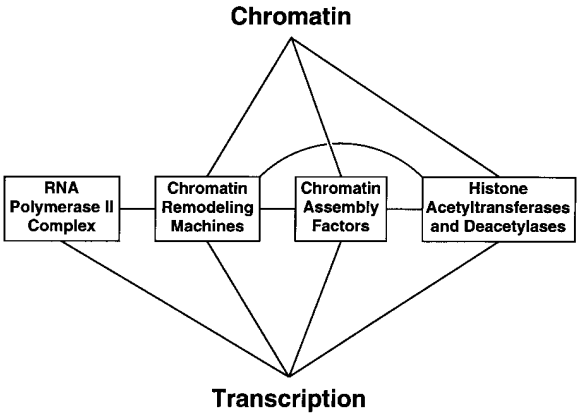

Figure 1. Schematic Diagram of Potential Linkages between Chromatin and Transcription

This figure depicts some linkages that have been suggested by experiments that are described in this review. As discussed in the text, there is not universal agreement with regard to some of these postulated connections. The arrangement of the factors and the nature of the connectivity are not meant to signify any preference or priority. In addition, this diagram is not meant to be comprehensive. For example, it does not include the relation between chromatin/DNA modification and gene regulation by DNA methylation.

to the holoenzyme. In this sense, the holoenzyme can be considered to be an RNA polymerase II–mediator complex (Kim et al., 1994).

The precise composition of the holoenzyme remains to be clarified, however, because different preparations of the holoenzyme that have been reported exhibit some variability in protein composition, especially for TFIIB, TFIID, TFIIE, and TFIIH. Nevertheless, the available data are consistent with the current hypothesis that a fraction of the RNA polymerase II associates with the mediator as well as other factors to give a large conglomerate that participates in the transcription process.

Then, what is the relation between the RNA polymerase II complex and chromatin remodeling machines that is depicted in Figure 1? An intriguing connection between the holoenzyme and the SWI/SNF complex was suggested by studies in which the SWI/SNF complex was found to be an integral and equimolar component of the mediator and holoenzyme (Wilson et al., 1996; reviewed in Kingston et al., 1996; Struhl, 1996; Ptashne and Gann, 1997). There is not, however, uniform agreement with regard to this conclusion, because components of SWI/SNF complexes were not found in other preparations of mediator or holoenzyme (Kim et al., 1994; Cairns et al., 1996; Pan et al., 1997). In addition, the recruitment of the yeast mediator/holoenzyme to a promoter with a GAL11-PHO4Δ2 fusion protein (which binds to DNA at PHO4 recognition sites and recruits the holoenzyme via the GAL11 fragment) was observed to induce nucleosome remodeling in vivo either in the presence or the absence of functionally active SWI/SNF complex (Gaudreau et al., 1997). These data collectively indicate that the nature of the interaction between holoenzyme and the SWI/SNF complex requires further clarification.

The association of the SWI/SNF complex with the RNA polymerase II holoenzyme also raises a question regarding the events that lead to transcriptional activation. That is, the collective results suggest the following: (1) the SWI/SNF-containing holoenzyme facilitates the binding of sequence-specific transcription factors to the promoter, and (2) the DNA-bound transcription factors function to recruit the SWI/SNF-containing holoenzyme to the promoter. Thus, with these two models for SWI/SNF function, it remains to be clarified whether (1) the promoter-associated holoenzyme recruits the promoter-binding transcription factors or (2) the promoter-associated transcription factors function to recruit the holoenzyme. Alternatively, the data might be accommodated by a model in which there is a concerted interaction of all of the factors (i.e., sequence-specific transcription factors and SWI/SNF-containing holoenyzme) with the promoter.

Histone Acetyltransferases and Deacetylases—Links to Transcription and Chromatin Remodeling Machines

The core histones, particularly H3 and H4, can be acetylated at the ϵ-amino groups of lysine residues in the N-terminal tails that extend outwardly from the globular core of the histone octamer. It has been postulated that the charge neutralization that occurs upon acetylation of the lysine side chains causes a reduction in the affinity of histone–DNA interactions and thus leads to increased access of transcription factors to the repressed chromatin template. Consistent with this hypothesis, there is a positive, but not universal, correlation between the extent of core histone acetylation and gene activity. For instance, a detailed analysis of the distribution of differently acetylated forms of histone H4 in *Drosophila* polytene chromosomes revealed that H4 acetylated at positions 5 or 8 is distributed throughout euchromatin, whereas H4 acetylated at position 12 is preferentially associated with β-heterochromatin, which is generally thought to be transcriptionally repressive (Turner et al., 1992). Hence, there does not appear to be a simple and general correlation between histone acetylation and transcriptional activity.

Recently, there has been considerable progress in the identification of enzymes that can catalyze the acetylation and deacetylation of histones (reviewed in Brownell and Allis, 1996; Kaufman, 1996; Roth and Allis, 1996; Wolffe and Pruss, 1996; Grunstein, 1997; Hartzog and Winston, 1997; Pazin and Kadonaga, 1997b). The enzymes that acetylate histones are usually referred to as histone acetyltransferases, or HATs, while the enzymes that deacetylate histones are called histone deacetylases, or HDACs. These studies revealed that HATs include the HAT1 protein, GCN5 protein, the TAFII250 subunit of TFIID, p300/CBP, P/CAF, and the SRC-1 family of coactivators. On the other hand, HDACs include a family of enzymes that are related to yeast RPD3 protein as well as a distinct protein termed HD2 (Lusser et al., 1997).

It is intriguing that the nuclear HATs include a basal transcription factor (TAFII250) and transcriptional coactivators (GCN5, p300/CBP, P/CAF, and SRC-1 proteins). In addition, the RPD3-related HDACs appear to participate in transcriptional repression (for recent review, see Pazin and Kadonaga, 1997b). These findings

provide evidence that the HAT and HDAC activities are an important component of transcriptional regulation. It does remain to be determined, however, whether it is the acetylation and deacetylation of core histones and/or other proteins that is responsible for the transcriptional effects. For instance, acetylation of HMG proteins, p53 (Gu and Roeder, 1997), and the basal transcription factors TFIIE and TFIIF (Imhof et al., 1997) has been observed. Hence, the biological function of HATs and HDACs may extend far beyond the acetylation and deacetylation of core histones.

Biochemical and genetic studies have led to the finding that yeast GCN5 protein is present in at least two distinct, multisubunit complexes that possess HAT activity (see Grant et al., 1997; Pollard and Peterson, 1997; Roberts and Winston, 1997; and references therein). For instance, a 1.8 MDa complex, termed SAGA (**S**PT-**A**DA-**G**CN5-**a**cetyltransferase), appears to contain GCN5, SPT3, SPT7, SPT8, SPT20/ADA5, ADA2, and ADA3 (Grant et al., 1997; Roberts and Winston, 1997). Genetic analysis of components of the SAGA and SWI/SNF complexes revealed a potential connection between these complexes. None of the known components of the SWI/SNF or SAGA complexes is essential for mitotic growth; however, synthetic lethality (or severe sickness) was seen between mutations in components of the SWI/SNF complex and mutations in components of the SAGA complex (Pollard and Peterson, 1997; Roberts and Winston, 1997). This synthetic lethality might be due to the independent and unrelated contributions of the SWI/SNF complex and the SAGA complex to an essential process, or alternatively, the two complexes might function more generally in a coordinate manner.

Chromatin Assembly Factors—Links to Transcription, Chromatin Remodeling Machines, and Histone Acetyltransferases and Deacetylases

Chromatin assembly is a fundamental biological process that is required for the duplication and the maintenance of the genome (for recent reviews, see Kaufman, 1996; Roth and Allis, 1996; Grunstein, 1997; Ito et al., 1997a). In actively dividing cells, chromatin assembly is required to package the newly synthesized DNA into chromatin, whereas in long-lived quiescent cells such as mammalian neurons, chromatin assembly is needed to maintain the integrity of the genome upon turnover of the histones. Although chromatin assembly occurs in either the presence or the absence of ongoing DNA replication, biochemical studies have shown that there is preferential assembly of chromatin onto newly replicated DNA relative to unreplicated DNA (Stillman, 1986; Smith and Stillman, 1989; Kamakaka et al., 1996). It has also been observed that core histones are acetylated (for instance, at positions 5, 8, and 12 of histone H4) immediately after synthesis in the cytoplasm and then become deacetylated after transport into the nucleus and assembly into chromatin. The specific function of this histone acetylation and deacetylation, such as whether it might be involved in protein stability and/or transport into the nucleus, remains to be elucidated.

The core chromatin assembly machinery that is required for the ATP-dependent assembly of periodic nucleosome arrays appears to comprise a protein complex termed ACF (**A**TP-utilizing **c**hromatin assembly and remodeling **f**actor) along with a core histone chaperone, such as CAF-1 (**c**hromatin **a**ssembly **f**actor-**1**) or NAP-1 (**n**ucleosome **a**ssembly **p**rotein-**1**) (Ito et al., 1997a, 1997b). ACF was purified from *Drosophila* embryos and found to be a multisubunit factor that contains ISWI, a protein that is in the same NTP-binding protein family as SWI2/SNF2 (Elfring et al., 1994; Eisen et al., 1995). Interestingly, ISWI is a subunit of at least three different protein complexes in *Drosophila*—NURF (**nu**cleosome **r**emodeling **f**actor; Tsukiyama et al., 1995), CHRAC (**chr**omatin-**a**ccessibility **c**omplex; Varga-Weisz et al., 1997), and ACF. ACF was also found to be able to mediate promoter-specific chromatin remodeling by GAL4-VP16 (a sequence-specific DNA-binding transcriptional activator) in an ATP-dependent manner (Ito et al., 1997b). Hence, the biochemical properties of ACF indicate that it can potentially participate in both chromatin assembly and the remodeling of nucleosomes that accompanies transcriptional activation.

The finding that ACF contains an ISWI subunit and can function as a chromatin remodeling factor suggests links among chromatin assembly, chromatin remodeling machines, and transcription (Figure 1). It has also been found that core histone-binding proteins (NAP-1 and nucleoplasmin) can stimulate the binding of transcription factors to mononucleosomes by a mechanism that appears to involve removal of histones H2A and H2B by the histone-binding proteins (Chen et al., 1994; Walter et al., 1995). Collectively, these findings suggest that chromatin assembly factors may participate in the transcription process.

The core chromatin assembly reaction that is mediated by ACF and a core histone chaperone does not appear to involve acetylation or deacetylation of core histones. It is interesting to note, however, that the core histone chaperone CAF-1 forms a complex, termed CAC (**c**hromatin **a**ssembly **c**omplex), with histone H3 and acetylated histone H4 (Verreault et al., 1996). In addition, the smallest subunit of CAF-1 appears to be identical to a protein that is associated with a histone deacetylase (Taunton et al., 1996; Tyler et al., 1996; Verreault et al., 1996; Kaufman et al., 1997) as well as closely related to a protein that is associated with the HAT1 histone acetyltransferase (Parthun et al., 1996; Tyler et al., 1996; Verreault et al., 1996; Kaufman et al., 1997). These data suggest that there are links between chromatin assembly and histone acetylation and deacetylation, but it also appears that the acetylation might be involved in aspects of the transport (including binding to CAF-1) or stability of the histones rather than in the intrinsic histone deposition process.

Recapitulation and Interlude

It is my hope that the preceding sections have provided some explanation of the connectivity of factors and processes that is outlined in Figure 1. When the entirety of the data is considered, it is evident that eukaryotic transcription is indeed an interlaced network of transcription factors and chromatin-modifying complexes. It now seems to be quite impossible to study transcriptional regulation in a manner that does not involve chromatin.

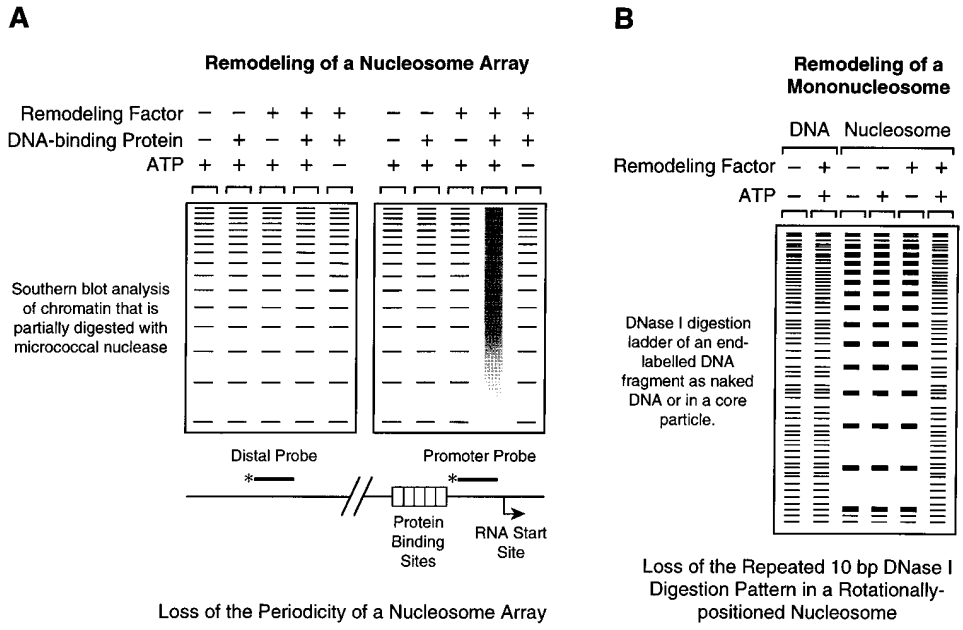

Figure 2. Two Chromatin Remodeling Assays

(A) Promoter-specific remodeling of a periodic nucleosome array by Southern blot analysis of micrococcal nuclease-digested chromatin.

(B) Perturbation of histone–DNA interactions in a rotationally positioned mononucleosome.

The remainder of this review will be a less structured discussion of some specific issues and questions that pertain to the chromatin remodeling machines. A comprehensive description of the properties and composition of these factors, which is not presented in the following section, can be found in several recent reviews (Peterson and Tamkun, 1995; Kingston et al., 1996; Peterson, 1996; Hartzog and Winston, 1997; Pazin and Kadonaga, 1997a; Tsukiyama and Wu, 1997).

What Is Chromatin Remodeling?

Thus far, I have used the term "chromatin remodeling" somewhat loosely. Hence, at this point, it is certainly reasonable to address the question of what is remodeling. To the best of my understanding, chromatin remodeling is any sort of detectable change in chromatin or mononucleosome structure. In addition, the terms "remodeling" and "reconfiguration" are generally used interchangeably.

How, then, is remodeling detected? According to the current rules that are applied in this area, a change in chromatin or mononucleosome structure, as detected by any one of several assays, qualifies as remodeling. For example, as depicted in Figure 2A, promoter-specific remodeling of a periodic nucleosome array can be detected by Southern blot analysis of micrococcal nuclease-digested chromatin. This assay has been used in the characterization of NURF and ACF (Tsukiyama and Wu, 1995; Ito et al., 1997b). In the hypothetical experiment shown in Figure 2A, the regularity of the nucleosome array is disrupted by the sequence-specific DNA-binding protein in a manner that is dependent upon an ATP-utilizing chromatin remodeling machine. This result reveals the loss of periodicity in the spacing of nucleosomes in the promoter region but does not indicate whether or not the nucleosomes are intact or disrupted. When the translational positions of the remodeled nucleosomes (i.e., the DNA sequences that are associated with specific nucleosomes) are additionally examined by indirect end-labeling analysis (which is something of a low-resolution nucleosome footprinting technique), nucleosomes are typically observed to be relocated to positions that flank the binding site(s) for the sequence-specific DNA binding protein (see, for example, Pazin et al., 1994; Tsukiyama et al., 1994). It should also be noted that the translational positioning of an array of nucleosomes can be altered without an apparent change in the regularity of the nucleosome array if multiple recognition sites for the DNA-binding protein are located at periodic intervals (in the linker regions between nucleosomes) that correspond to the nucleosome repeat length (see, for example, Pazin et al., 1997). Thus, it is useful to carry out both the micrococcal nuclease–Southern blot assay (Figure 2A) and indirect end-labeling analysis in the characterization of nucleosome arrays.

A widely used assay that detects changes in histone–DNA interactions in a mononucleosome is shown in Figure 2B. In this hypothetical experiment, the perturbation of histone–DNA interactions by an ATP-utilizing chromatin remodeling factor is detected by the loss of a characteristic 10 bp DNase I digestion ladder in a rotationally positioned mononucleosome. Rotational positioning of DNA relative to the core histone octamer is achieved in a population of nucleosomes when there is uniformity in the orientation of one face of the DNA relative to the surface of the globular core of the octamer. This effect is commonly observed with curved DNA fragments, which exhibit a preference to wrap around the octamer in an

orientation that is favored by the curvature of the DNA (i.e., the orientation that requires the least amount of bending of the DNA). When a nucleosome is treated with DNase I, the nuclease cleaves the DNA at approximately 10 bp intervals where the minor groove of the DNA (which is cleaved by DNase I) is facing away from the octamer. Thus, if a population of rotationally positioned nucleosomes is treated with DNase I, then a 10 bp ladder is seen with nucleosomes in the absence of the remodeling factor. As depicted in Figure 2B, the ATP-utilizing chromatin remodeling factor alters the histone–DNA interactions, as seen by the loss of the 10 bp DNase I ladder. In this specific instance, the DNase I digestion pattern of the remodeled chromatin resembles that of naked DNA, as is typically observed. These results do not indicate, however, whether or not the DNA has dissociated from the histone octamer.

DNase I digestion analysis of mononucleosomes has been used not only to detect the alteration of the 10 bp repeating ladder (Figure 2B) but also to investigate the ability of remodeling factors to facilitate the binding of transcription factors to mononucleosomes (i.e., DNase I footprinting with mononucleosomes). These types of experiments have been used to characterize the properties of yeast SWI/SNF complex, yeast RSC complex, *Drosophila* NURF, and human BRG1- and hbrm-containing complexes (see, for example, Côté et al., 1994; Kwon et al., 1994; Imbalzano et al., 1994; Tsukiyama and Wu, 1995; Cairns et al., 1996; Wang et al., 1996).

Some remodeling assays involve enhanced sensitivity of chromatin to nucleases. The hypersensitivity of specific regions in chromatin to digestion by nucleases such as DNase I has been used for many years in the study of gene regulation (for reviews, see Elgin, 1988; Gross and Garrard, 1988). Transcription factor-induced DNase I hypersensitive sites can also be seen with chromatin that is assembled in vitro (see, for instance, Pazin et al., 1996). In a different type of experiment, the general mobility of nucleosomes can be detected by monitoring the accessibility of nucleases to chromatin. For example, the nucleosome-remodeling factor CHRAC was identified and purified on the basis of its ability to enhance the ability of restriction enzymes (such as DraI) to digest chromatin (Varga-Weisz et al., 1995, 1997).

The assays that are currently being used to analyze nucleosome remodeling are, at present, somewhat crude. While the assays detect alterations in nucleosome structure, there is little learnt about the specific nature of the changes. In some instances, such as when remodeled nucleosomes are altered from the canonical nucleosomal form, we really do not yet know what remodeling is from a molecular point of view. Hence, the development of new and more informative assays to characterize chromatin remodeling is a high priority.

Some Issues Pertaining to the Function of Chromatin Remodeling Machines

In this section, I will address a few questions that I have seen arise in the discussion of chromatin remodeling machines. Most of the responses are in the realm of conjecture and speculation.

What Is the Function of the SWI2/SNF2-like Subunit in the Remodeling Machines?

The SWI2/SNF2-like proteins are members of a large group of NTP-binding proteins that includes many RNA and DNA helicases (see, for example, Eisen et al., 1995; Pazin and Kadonaga, 1997a). Unlike helicases, the SWI2/SNF2-like proteins do not appear to catalyze DNA unwinding. On the other hand, like helicases, these proteins may be molecular motors that possess an ATP-driven DNA-translocating activity. Helicases can translocate along DNA at rates of about 500 to 1000 nt/s, and thus, such a processive DNA-translocating activity in SWI/SNF and related complexes has tremendous potential for chromatin remodeling by disruption of histone–DNA interactions.

Why Are There Three Different Remodeling Complexes in Drosophila *(NURF, CHRAC, and ACF) that Each Contain an ISWI Subunit?*

Each of these protein complexes was identified and purified on the basis of a biochemical activity. NURF was found to be required for chromatin remodeling that is induced by the GAGA factor (a *Drosophila* sequence-specific DNA-binding protein) (Tsukiyama et al., 1994, 1995; Tsukiyama and Wu, 1995), as monitored by the loss of periodicity of nucleosome arrays (Figure 2A). CHRAC was identified on the basis of its ability to mobilize nucleosomes in a manner that allows enhanced access of a restriction enzyme to DNA packaged into chromatin (Varga-Weisz et al., 1995, 1997). ACF was purified as an ATP-utilizing chromatin assembly factor (Bulger et al., 1995; Ito et al., 1997b). It is possible that the common ISWI subunit serves a related biochemical function, such as a processive DNA-translocating motor, in each of these complexes. There does remain, however, essentially endless potential for the use of this putative ISWI "motor" to provide the mechanical energy that is needed for specialized biological functions of the different complexes. Aside from ISWI, the other subunits of these complexes appear to be distinct. For example, one of the subunits of CHRAC, but not of NURF or ACF, is topoisomerase II.

The presence of a common subunit in different complexes is not particularly unusual. For instance, TBP is present in multiple complexes that are involved in transcription by RNA polymerases I, II, and III. In addition, the smallest subunit of *Drosophila* CAF-1 (dCAF-1 p55) is (1) associated with a histone deacetylase (Taunton et al., 1996; Tyler et al., 1996), (2) a subunit of NURF (Martínez-Balbás et al., 1997), and (3) closely related to a protein that is associated with the HAT1 histone acetyltransferase (Parthun et al., 1996).

Why Is the Yeast SWI/SNF Complex Required for Full Transcriptional Activity of Some, but Not All Promoters?

One model for this promoter specificity of SWI/SNF action is that the complex is targeted to a selected subset of promoters. This targeting could, at least in theory, be achieved by interactions between SWI/SNF complex and sequence-specific promoter-binding factors and/or by sequence-specific DNA binding by the SWI/SNF itself (although there is no evidence for such sequence-specific DNA binding by SWI/SNF complex).

An alternate model for the variation in the effect of SWI/SNF complex at different promoters is that weak promoters require SWI/SNF function for full activity, whereas strong promoters do not require SWI/SNF at all. There is some evidence that supports this hypothesis. For instance, when two of the four GAL4 binding sites in the *GAL1,10* UAS are removed, the SWI/SNF-independent *GAL1* promoter becomes dependent on SWI/SNF (Burns and Peterson, 1997; Gaudreau et al., 1997). In addition, when two low-affinity GAL4 binding sites in the *GAL1,10* UAS are converted to high-affinity consensus GAL4 recognition sites, the SWI/SNF-dependent weak promoter (with the nonconsensus GAL4 sites) becomes independent of SWI/SNF complex (Burns and Peterson, 1997). Thus, there does appear to be an inverse correlation between promoter strength and the requirement for the SWI/SNF complex for full transcriptional activation. These results suggest that promoter specificity of the SWI/SNF complex may be due, at least in part, to the promoter strength.

Summary and Perspectives

In conclusion, this review covers only a fraction of our current knowledge of chromatin remodeling machines and the broad relation between chromatin and transcription. It is my hope that this essay is nonetheless able to convey some useful information. In the future, it seems likely that there will be significant advances in our understanding of both the biological function of chromatin remodeling machines and the molecular mechanisms by which these factors function. There is, however, a significant need for improved experimental strategies and assays for the analysis of chromatin remodeling as well as for the characterization of the biological activities of different remodeling machines. The existing assays generally provide little information regarding the nature of the changes in nucleosome structure that occur upon remodeling. Moreover, with some of the simplified assays, the activities of different remodeling complexes, which likely have distinct biological functions, appear to be indistinguishable. Thus, while progress is imminent, it will still require some ingenuity to be achieved.

Acknowledgments

I am grateful to Jessica Tyler and Lee Kraus for critical reading of the manuscript. I also thank the many helpful individuals who have kindly and generously provided preprints of their papers prior to publication. I apologize for any errors or omissions in this review. Studies of chromatin structure and transcriptional regulation in the laboratory of J. T. K. are supported by the National Institutes of Health (GM46995). Studies of chromatin assembly with ACF are supported by the National Science Foundation (MCB 9631121).

References

Björklund, S., and Kim, Y.-J. (1996). Mediator of transcriptional regulation. Trends Biochem. Sci. *21*, 335–337.

Brownell, J.E., and Allis, C.D. (1996). Special HATs for special occasions: linking histone acetylation to chromatin assembly and gene activation. Curr. Opin. Genet. Dev. *6*, 176–184.

Bulger, M., Ito, T., Kamakaka, R.T., and Kadonaga, J.T. (1995). Assembly of regularly spaced nucleosome arrays by dCAF-1 and a 56 kDa histone-binding protein. Proc. Natl. Acad. Sci. USA *92*, 11726–11730.

Burns, L.G., and Peterson, C.L. (1997). The yeast SWI-SNF complex facilitates binding of a transcriptional activator to nucleosomal sites in vivo. Mol. Cell. Biol. *17*, 4811–4819.

Cairns, B.R., Lorch, Y., Li, Y., Zhang, M., Lacomis, L., Erdjument-Bromage, H., Tempst, P., Du, J., Laurent, B., and Kornberg, R.D. (1996). RSC, an essential, abundant chromatin-remodeling complex. Cell *87*, 1249–1260.

Chen, H., Li, B., and Workman, J.L. (1994). A histone-binding protein, nucleoplasmin, stimulates transcription factor binding to nucleosomes and factor-induced nucleosome disassembly. EMBO J. *13*, 380–390.

Côté, J., Quinn, J., Workman, J.L., and Peterson, C.L. (1994). Stimulation of GAL4 derivative binding to nucleosomal DNA by the yeast SWI/SNF complex. Science *265*, 53–60.

Eisen, J.A., Sweder, K.S., and Hanawalt, P.C. (1995). Evolution of the SNF2 family of proteins: subfamilies with distinct sequences and functions. Nucleic Acids Res. *23*, 2715–2723.

Elfring, L.K., Deuring, R., McCallum, C.M., Peterson, C.L., and Tamkun, J.W. (1994). Identification and characterization of *Drosophila* relatives of the yeast transcriptional activator SNF2/SWI2. Mol. Cell. Biol. *14*, 2225–2234.

Elgin, S.C.R. (1988). The formation and function of DNase I hypersensitive sites in the process of gene activation. J. Biol. Chem. *263*, 19259–19262.

Felsenfeld, G. (1996). Chromatin unfolds. Cell *86*, 13–19.

Gaudreau, L., Schmid, A., Blaschke, D., Ptashne, M., and Hörz, W. (1997). RNA polymerase II holoenzyme recruitment is sufficient to remodel chromatin at the yeast *PHO5* promoter. Cell *89*, 55–62.

Gross, D.S., and Garrard, W.T. (1988). Nuclease hypersensitive sites in chromatin. Annu. Rev. Biochem. *57*, 159–197.

Grant, P.A., Duggan, L., Côté, J., Roberts, S.M., Brownell, J.E., Candau, R., Ohba, R., Owen-Hughes, T., Allis, C.D., Winston, F., et al. (1997). Yeast Gcn5 functions in two multisubunit complexes to acetylate nucleosomal histones: characterization of an Ada complex and the SAGA (Spt/Ada) complex. Genes Dev. *11*, 1640–1650.

Grunstein, M. (1997). Histone acetylation in chromatin structure and transcription. Nature *389*, 349–352.

Gu, W., and Roeder, R.G. (1997). Activation of p53 sequence-specific DNA binding by acetylation of the p53 C-terminal domain. Cell *90*, 595–606.

Imbalzano, A.N., Kwon, H., Green, M.R., and Kingston, R.E. (1994). Facilitated binding of TATA-binding protein to nucleosomal DNA. Nature *370*, 481–485.

Hartzog, G.A., and Winston, F. (1997). Nucleosomes and transcription: recent lessons from genetics. Curr. Opin. Genet. Dev. *7*, 192–198.

Hirschhorn, J.N., Brown, S.A., Clark, C.D., and Winston, F. (1992). Evidence that SNF2/SWI2 and SNF5 activate transcription in yeast by altering chromatin structure. Genes Dev. *6*, 2288–2298.

Imhof, A., Yang, X.-J., Ogryzko, V.V., Nakatani, Y., Wolffe, A.P., and Ge, H. (1997). Acetylation of general transcription factors by histone acetyltransferases. Curr. Biol. *7*, 689–692.

Ito, T., Tyler, J.K., and Kadonaga, J.T. (1997a). Chromatin assembly factors: a dual function in nucleosome formation and mobilization? Genes Cells *2*, 593–600.

Ito, T., Bulger, M., Pazin, M.J., Kobayashi, R., and Kadonaga, J.T. (1997b). ACF, an ISWI-containing and ATP-utilizing chromatin assembly and remodeling factor. Cell *90*, 145–155.

Kamakaka, R.T., Bulger, M., Kaufman, P.D., Stillman, B., and Kadonaga, J.T. (1996). Post-replicative chromatin assembly by *Drosophila* and human chromatin assembly factor-1. Mol. Cell. Biol. *16*, 810–817.

Kaufman, P.D. (1996). Nucleosome assembly: the CAF and the HAT. Curr. Opin. Cell Biol. *8*, 369–373.

Kaufman, P.D., Kobayashi, R., and Stillman, B. (1997). Ultraviolet radiation sensitivity and reduction of telomeric silencing in *Saccharomyces cerevisiae* cells lacking chromatin assembly factor-I. Genes Dev. *11*, 345–357.

Kim, Y.-J., Björklund, S., Li, Y., Sayre, M.H., and Kornberg, R.D. (1994). A multiprotein mediator of transcriptional activation and its interaction with the C-terminal repeat domain of RNA polymerase II. Cell *77*, 599–608.

Kingston, R.E., Bunker, C.A., and Imbalzano, A.N. (1996). Repression and activation by multiprotein complexes that alter chromatin structure. Genes Dev. *10*, 905–920.

Koleske, A.J., and Young, R.A. (1995). The RNA polymerase II holoenzyme and its implications for gene regulation. Trends Biochem. Sci. *20*, 113–116.

Kornberg, R.D., and Lorch, Y. (1995). Interplay between chromatin structure and transcription. Curr. Opin. Cell Biol. *7*, 371–375.

Kruger, W., Peterson, C.L., Sil, A., Coburn, C., Arents, G., Moudrianakis, E.N., and Herskowitz, I. (1995). Amino acid substitutions in the structured domains of histones H3 and H4 partially relieve the requirement of the yeast SWI/SNF complex for transcription. Genes Dev. *9*, 2770–2779.

Kwon, H., Imbalzano, A.N., Khavari, P.A., Kingston, R.E., and Green, M.R. (1994). Nucleosome disruption and enhancement of activator binding by a human SWI/SNF complex. Nature *370*, 477–481.

Lusser, A., Brosch, G., Loidl, A., Haas, H., and Loidl, P. (1997). Identification of maize histone deacetylase HD2 as an acidic nucleolar phosphoprotein. Science *277*, 88–91.

Martínez-Balbás, M.A., Tsukiyama, T., Gdula, D., and Wu, C. (1997). *Drosophila* NURF-55, a WD repeat protein involved in histone metabolism. Proc. Natl. Acad. Sci. USA, in press.

Orphanides, G., Lagrange, T., and Reinberg, D. (1996). The general transcription factors of RNA polymerase II. Genes Dev. *10*, 2657–2683.

Pan, G., Aso, T., and Greenblatt, J. (1997). Interaction of elongation factors TFIIS and elongin A with a human RNA polymerase II holoenzyme capable of promoter-specific initiation and responsive to transcriptional activators. J. Biol. Chem. *272*, 24563–24571.

Parthun, M.R., Widom, J., and Gottschling, D.E. (1996). The major cytoplasmic histone acetyltransferase in yeast: links to chromatin replication and histone metabolism. Cell *87*, 85–94.

Pazin, M.J., and Kadonaga, J.T. (1997a). SWI2/SNF2 and related proteins: ATP-driven motors that disrupt protein–DNA interactions? Cell *88*, 737–740.

Pazin, M.J., and Kadonaga, J.T. (1997b). What's up and down with histone deacetylation and transcription? Cell *89*, 325–328.

Pazin, M.J., Kamakaka, R.T., and Kadonaga, J.T. (1994). ATP-dependent nucleosome reconfiguration and transcriptional activation from preassembled chromatin templates. Science *266*, 2007–2011.

Pazin, M.J., Sheridan, P.L., Cannon, K., Cao, Z., Keck, J.G., Kadonaga, J.T., and Jones, K.A. (1996). NF-kB-mediated chromatin reconfiguration and transcriptional activation of the HIV-1 enhancer in vitro. Genes Dev. *10*, 37–49.

Pazin, M.J., Bhargava, P., Geiduschek, E.P., and Kadonaga, J.T. (1997). Nucleosome mobility and the maintenance of nucleosome positioning. Science *276*, 809–812.

Peterson, C.L. (1996). Multiple switches to turn on chromatin? Curr. Opin. Genet. Dev. *6*, 171–175.

Peterson, C.L., and Tamkun, J.W. (1995). The SWI-SNF complex: a chromatin remodeling machine? Trends Biochem. Sci. *20*, 143–146.

Pollard, K.J., and Peterson, C.L. (1997). Role for *ADA*/*GCN5* products in antagonizing chromatin-mediated transcriptional repression. Mol. Cell. Biol. *11*, 6212–6222.

Ptashne, M., and Gann, A. (1997). Transcriptional activation by recruitment. Nature *386*, 569–577.

Roberts, S.M., and Winston, F. (1997). Essential functional interactions of SAGA, a *Saccharomyces cerevisiae* complex of Spt, Ada, and Gcn5 proteins, with the Snf/Swi and Srb/mediator complexes. Genetics *147*, 451–465.

Roth, S.Y., and Allis, C.D. (1996). Histone acetylation and chromatin assembly: a single escort, multiple dances? Cell *87*, 5–8.

Smith, S., and Stillman, B. (1989). Purification and characterization of CAF-I, a human cell factor required for chromatin assembly during DNA replication in vitro. Cell *58*, 15–25.

Stillman, B. (1986). Chromatin assembly during SV40 DNA replication in vitro. Cell *45*, 555–565.

Struhl, K. (1996). Chromatin structure and the RNA polymerase II connection: implications for transcription. Cell *84*, 179–182.

Taunton, J., Hassig, C.A., and Schreiber, S.L. (1996). A mammalian histone deacetylase related to the yeast transcriptional regulator Rpd3p. Science *272*, 408–411.

Tsukiyama, T., and Wu, C. (1995). Purification and properties of an ATP-dependent nucleosome remodeling factor. Cell *83*, 1011–1020.

Tsukiyama, T., and Wu, C. (1997). Chromatin remodeling and transcription. Curr. Opin. Genet. Dev. *7*, 182–191.

Tsukiyama, T., Becker, P.B., and Wu, C. (1994). ATP-dependent nucleosome disruption at a heat-shock promoter mediated by binding of GAGA transcription factor. Nature *367*, 525–532.

Tsukiyama, T., Daniel, C., Tamkun, J., and Wu, C. (1995). ISWI, a member of the SWI2/SNF2 ATPase family, encodes the 140 kDa subunit of the nucleosome remodeling factor. Cell *83*, 1021–1026.

Turner, B.M., Birley, A.J., and Lavender, J. (1992). Histone H4 isoforms acetylated at specific lysine residues define individual chromosomes and chromatin domains in *Drosophila* polytene nuclei. Cell *69*, 375–384.

Tyler, J.K., Bulger, M., Kamakaka, R.T., Kobayashi, R., and Kadonaga, J.T. (1996). The p55 subunit of *Drosophila* chromatin assembly factor 1 is homologous to a histone deacetylase-associated protein. Mol. Cell. Biol. *16*, 6149–6159.

Varga-Weisz, P.D., Blank, T.A., and Becker, P.B. (1995). Energy-dependent chromatin accessibility and nucleosome mobility in a cell-free system. EMBO J. *14*, 2209–2216.

Varga-Weisz, P.D., Wilm, M., Bonte, E., Dumas, K., Mann, M., and Becker, P.B. (1997). Chromatin-remodelling factor CHRAC contains the ATPases ISWI and topoisomerase II. Nature *388*, 598–602.

Verreault, A., Kaufman, P.D., Kobayashi, R., and Stillman, B. (1996). Nucleosome assembly by a complex of CAF-1 and acetylated histones H3/H4. Cell *87*, 95–104.

Walter, P.P., Owen-Hughes, T.A., Côté, J., and Workman, J.L. (1995). Stimulation of transcription factor binding and histone displacement by nucleosome assembly protein 1 and nucleoplasmin requires disruption of the histone octamer. Mol. Cell. Biol. *15*, 6178–6187.

Wang, W., Côté, J., Xue, Y., Zhou, S., Khavari, P.A., Biggar, S.R., Muchardt, C., Kalpana, G.V., Goff, S.P., Yaniv, M., et al. (1996). Purification and biochemical heterogeneity of the mammalian SWI-SNF complex. EMBO J. *15*, 5370–5382.

Wilson, C.J., Chao, D.M., Imbalzano, A.N., Schnitzler, G.R., Kingston, R.E., and Young, R.A. (1996). RNA polymerase II holoenzyme contains SWI/SNF regulators involved in chromatin remodeling. Cell *84*, 235–244.

Wolffe, A.P., and Pruss, D. (1996). Targeting chromatin disruption: transcription regulators that acetylate histones. Cell *84*, 817–819.