

Review

# The mechanism of sister chromatid cohesion

Frank Uhlmann\*

*Chromosome Segregation Laboratory, Cancer Research UK London Research Institute, London WC2A 3PX, UK*

Received 8 January 2004

Available online 2 April 2004

## Abstract

Each of our cells inherit their genetic information in the form of chromosomes from a mother cell. In order that we obtain the full genetic complement, cells need to ensure that replicated chromosomes are accurately split and distributed during cell division. Mistakes in this process lead to aneuploidies, cells with supernumerous or missing chromosomes. Most aneuploid human embryos are not viable, and if they are, they develop severe birth defects. Aneuploidies later in human life are frequently found associated with the development of malignant cancer. DNA replication during S-phase is linked to segregation of the sister copies in mitosis by sister chromatid cohesion. A chromosomal protein complex, cohesin, holds replicated sister DNA strands together after their synthesis. This allows pairs of replication products to be recognised by the spindle apparatus in mitosis for segregation into opposite direction. At anaphase onset, cohesin is destroyed by a site-specific protease, separase. Here I review what we have learned about the molecular mechanism of sister chromatid cohesion. Cohesin forms a large proteinaceous ring that may hold sister chromatids by encircling and topological trapping. To understand how cohesin links newly synthesised replication products, biochemical assays to study the enzymology of cohesin will be required.

© 2004 Elsevier Inc. All rights reserved.

*Keywords:* Sister chromatid cohesion; Chromosome segregation; Cohesion

## Introduction

Cell division is the basis for growth and proliferation of all living organisms. The cell's genome contains the blueprint for growth, as well as the many other molecular tasks that cells have to fulfil to form a functional, healthy organism. A fundamental aspect of cell division is therefore the accurate transmission of the genome to daughter cells.

The DNA that contains the genomic information comes in several large pieces, the chromosomes. These are faithfully replicated once every cell generation, a sophisticated process that takes place during S-phase. The resulting copies, the sister chromatids, then have to be correctly distributed to two daughter cells during mitosis. To prevent any confusion as to which chromatids have to reach each daughter, the sister chromatids of each chromosome are tied together as pairs during the process of their synthesis. This tie, called sister chromatid cohesion, allows pairs of chro-

matids to be recognised in mitosis for alignment on the bipolar metaphase spindle. Each sister pair is oriented such that tension of the spindle anticipates the anaphase movement into opposite direction. Only when all chromosomes have reached such bioriented alignment, sister chromatid cohesion is destroyed by separase to trigger the final segregation into daughter cells (for reviews, see Refs. [1,2]).

## Sister chromatid cohesion, mediated by the cohesin ring

The molecular basis for sister chromatid cohesion is a chromosomal protein complex, called cohesin [3–5]. The cohesin complex consists of at least four subunits that together form a large proteinaceous ring (Fig. 1) [6–8]. To understand how cohesin holds together replicated sister chromatids and provides the strength to counteract the spindle force in mitosis, yet is swiftly released from chromatin after cleavage by separase, it is important to understand cohesin's mode of DNA binding. The ring shape of the cohesin complex has prompted the hypothesis that cohesin might bind by encircling and thereby topologically trapping DNA [2,7]. The diameter of the ring of close to 50

\* Chromosome Segregation Laboratory, Cancer Research UK London Research Institute, 44 Lincoln's Inn Fields, London WC2A 3PX, UK. Fax: +44-207-269-3581.

*E-mail address:* [frank.uhlmann@cancer.org.uk](mailto:frank.uhlmann@cancer.org.uk).

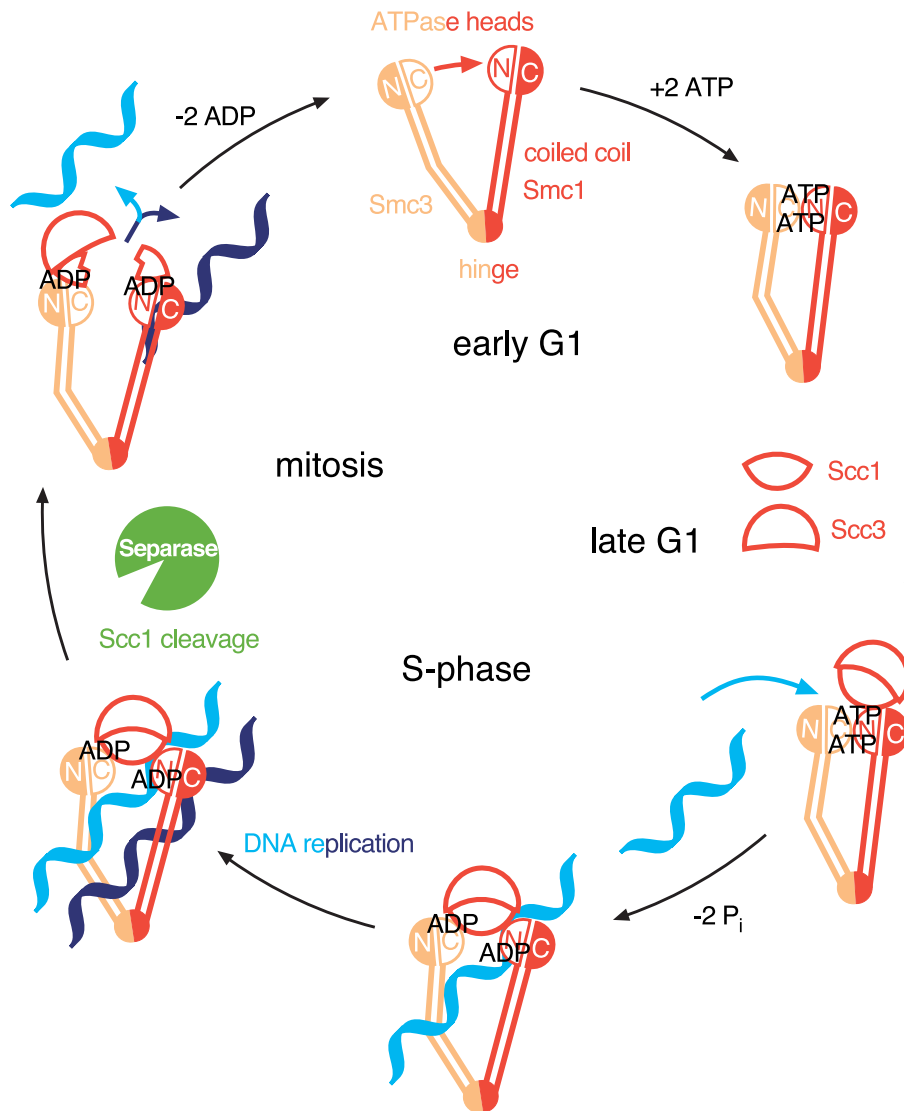


Fig. 1. A model for ATP hydrolysis-dependent binding of cohesin to DNA. The Smc1 and Smc3 subunits of cohesin form a proteinaceous ring in early G1, each consisting of a 50-nm-long stretch of coiled coil, held together at a hinge and at ATPase heads that dimerise upon ATP binding. In late G1, the Scc1 subunit is synthesised that binds to the Smc heads and recruits Scc3. Scc1 and Scc3 may make contact mainly with one of the Smc1 heads. This leaves a path open for DNA to reach the ATPase heads and stimulate hydrolysis of ATP. ATP hydrolysis in turn leads to separation of the Smc heads, thereby opening a gap in the Smc ring through which DNA may enter. ATP hydrolysis also changes the interaction of Scc1 with the Smc heads such that now Scc1 bridges the gap between Smc1 and Smc3. During DNA replication in S-phase, the replication fork may travel along DNA and through the cohesin ring, leaving the replication products (the sister chromatids) trapped inside. Once all pairs of sister chromatids are aligned in bioriented fashion in mitosis, separase is activated to cleave Scc1 and open the ring. The C-terminal Scc1 cleavage product keeps the cohesin ring open until it is degraded. ADP may dissociate from cohesin only after degradation of the Scc1 fragment. Then ATP can rebind and the Smc ring closes again in preparation for the following cell cycle.

nm would be sufficiently large to hold two sister DNA strands even when wrapped around histones. The circumference of the cohesin ring consists largely of flexible coiled coil of the subunits Smc1 and Smc3, and these two subunits bind each other in a head to head and tail to tail orientation (Fig. 1). The tails are linked with high affinity at a dimerisation interface, known as the hinge [7]. The Smc1 and Smc3 heads are ABC type ATPases that dimerise with each other after binding ATP [9,10]. Two further essential cohesin subunits, Scc1 and Scc3, associate with the Smc heads. Scc1 in particular shows interesting features, its N terminus binds to Smc3 while its C terminus binds Smc1

[7]. Thus, Scc1 could stabilise the interaction between the Smc heads. Scc1 associates with Smc heads that have bound either ATP or ADP [10,11]. This could be important because after ATP hydrolysis, the direct interaction between Smc1 and Smc3 heads is probably weakened, and any remaining link between the heads may depend on Scc1. At anaphase onset, it is the Scc1 subunit that is cleaved by separase to destroy sister chromatid cohesion [12–15].

Evidence has been accumulating that cohesin binds DNA by encircling and topological trapping. Cohesin is tightly bound to budding yeast chromatin from where it cannot be extracted by high salt treatment, under conditions that strip

off most DNA binding proteins. In contrast, when the DNA is restricted by nuclease treatment, cohesin is efficiently released from chromatin, as if it could now slip off the DNA ends [16]. As mentioned, Scc1 is cleaved in anaphase, and this in turn disrupts the interaction between the Smc heads in cohesin, thus opening the ring [10]. Importantly, not only cleavage of Scc1, also engineered cleavage of the coiled coil arm of Smc3 will release cohesin from DNA [8]. Thus, cleavage anywhere in the proposed topological surface between cohesin and DNA will destroy the cohesin DNA interaction, consistent with the idea that topological trapping is an essential aspect of cohesin's chromosome binding.

### Loading DNA into the cohesin ring

If cohesin indeed traps DNA within the ring, how does the DNA get inside? The similarity of the Smc heads to the ATPase domains of ABC transporters suggested that ATP hydrolysis by cohesin might be coupled to active transport of DNA into the ring [17]. In the case of ABC transporters, two ATPase domains are bound to a transmembrane channel, and the ATP hydrolysis-driven change in ATPase domain dimerisation is thought to drive the flow of solutes through the channel [18,19]. The cohesin ATPase domains are linked to the two stretches of Smc coiled coil, and the question therefore is whether an ATP hydrolysis-driven change in Smc head interactions, and possibly accompanying conformational changes, might be coupled to the transport of DNA into the ring.

Mutations in the Smc head domains that allow ATP binding but are predicted to prevent ATP hydrolysis have been constructed [10,11]. These mutant Smc subunits assemble with the other subunits into seemingly intact cohesin complexes, but such complexes do not bind chromatin *in vivo*. This finding is consistent with the idea that ATP hydrolysis is indeed required for DNA binding by cohesin. It fits well with the idea that ATP hydrolysis transports the DNA into the cohesin ring. It does of course not exclude that the Smc heads could use ATP hydrolysis not to pump the DNA into the ring, but to grab DNA and hold on to it in a different way, maybe by trapping it in a groove between the heads and Scc1 [20]. To better understand how cohesin binds DNA, it was therefore important to know which parts of cohesin are required for DNA binding.

A cohesin complex was constructed in budding yeast cells that included wild-type Smc heads but lacked the hinge, thereby producing an open gap in the ring [10]. Such a complex was not able to bind to chromatin despite the presence of intact Smc heads. This suggests that the Smc ATPase heads by themselves cannot bind to DNA, but that they only promote DNA binding when connected to a closed Smc ring. DNA transport by the ATPase heads into the ring would explain both the requirement of ATP hydrolysis as well as an intact cohesin ring for DNA binding.

Mutant analysis *in vivo*, as described, can provide important hints for possible scenarios of cohesin binding

to DNA. However, the behaviour of mutant proteins is difficult to fully interpret. For example, the Smc subunits that were designed to be defective in ATP hydrolysis, did they differ from wild-type proteins solely in their inability to hydrolyse ATP? Furthermore, the Smc subunits with intact ATPase heads but without hinge, was the ATPase activity really unaffected by the hinge truncation? These and many similar caveats to the analysis of mutant proteins *in vivo* call for the reconstitution of the binding reaction of cohesin to DNA *in vitro*, using purified components. Only this will allow important open questions to be addressed: When is ATP bound, hydrolysed, and when is the hydrolysed nucleotide released during the binding reaction? When and where does DNA contact cohesin? Can intermediates of the binding reaction be trapped? And maybe most importantly, can cohesin be observed bound to DNA by a microscopic technique that visualises whether DNA really is inside the cohesin ring after binding?

### *In vitro* studies on cohesin and related complexes

The purification of the necessary amounts of cohesin for biochemical studies is still a considerable challenge. Nevertheless, first experiments to investigate *in vitro* binding of purified human and budding yeast cohesin to DNA have been published [21,22]. In both sets of experiments, cohesin clearly showed a tendency to associate with DNA. However, the affinity to DNA was modest, no defined products of cohesin on DNA could be identified, and the binding that was observed occurred independently of the availability of hydrolysable ATP. This suggests that these *in vitro* reactions of cohesin with DNA did not fully recapitulate what happens when cohesin binds to DNA *in vivo*. Instead, the results so far might demonstrate an association of cohesin with DNA that normally precedes the ATP hydrolysis-dependent loading reaction of cohesin onto DNA. Why ATP hydrolysis-dependent DNA binding of cohesin could not be observed *in vitro* is difficult to know. The ATP-dependent activities of cohesin may be difficult to preserve during the purification procedure, or the correct DNA substrate for cohesin loading may not yet have been found. In addition, chromatin binding of cohesin *in vivo* requires the proteins Scc2 and Scc4, as well as a chromatin remodelling complex [16,23]. While the function of these accessory factors *in vivo* is not fully understood, it remains a possibility that DNA binding by cohesin *in vitro* also depends on some of them.

The mechanism of cohesin binding to DNA may share similarities with DNA binding of condensin, a chromosomal protein complex related to cohesin [24]. Condensin, like cohesin, consists of two Smc proteins, Smc2 and Smc4. The Smc2 and Smc4 ATPase heads dimerise with each other, and they bind three additional subunits, one of which shows homology to Scc1 [6,25]. Thus, just like with cohesin, Smc2 and Smc4 form a topological condensin ring. Condensin associates with DNA independently of ATP, but ATP hydro-

lysis is required for the actual binding reaction [26,27]. ATP-dependent condensin binding to DNA has been easier to reconstitute *in vitro* and has been studied in some detail. An interesting feature is that depending on ATP hydrolysis, DNA appears to be wrapped around condensin [27]. This wrapping generates torsion which introduces two positive supercoils into DNA per molecule of bound condensin. Whether DNA enters the condensin ring during the binding reaction has not yet been possible to determine. To address this, it would be interesting to compare the strength of the ATP-dependent and -independent modes of condensin binding to DNA.

When condensin was visualised bound to DNA by atomic force microscopy, it was seen touching the DNA via the Smc hinge [28]. This is exactly opposite from where the ATPase heads are thought to act in DNA binding. Notably, however, this mode of DNA binding was independent of ATP hydrolysis. It could therefore indicate that the first, ATP-independent contact of condensin, and maybe also of cohesin, with DNA is made at the Smc hinge. Intriguingly, a significant fraction of the condensin molecules were found bent back on themselves, the heads touching the hinge. Is this an indication that the heads pick up DNA from the hinge for transport, or does it mean that a more sophisticated collaboration of heads and hinge accomplishes eventual uptake of DNA into the ring? Further functional analysis *in vitro* coupled with visualisation of intermediates and products of the DNA binding reaction promise to hold crucial information.

Bacteria also contain chromatin-associated protein complexes based on Smc subunits. There is typically only one Smc subunit encoded by bacterial genomes, and this Smc subunit forms homodimeric protein rings [29]. Additional subunits, one of which again an Scc1-related subunit, associate with the Smc ring (the conserved Scc1-related subunits in all organisms have been given the generic name 'kleisins') [25,30]. These Smc complexes are required for bacterial chromatin compaction and chromosome segregation, and they probably function according to similar principles as cohesin and condensin [31–33]. Biochemical analysis of the bacterial Smc complexes has so far been largely restricted to studies of the Smc subunits only. This has shown that the ATPase activity within the Smc heads is significantly stimulated by the presence of DNA [34]. The ATPase activity of condensin is also stimulated by DNA [26]. These findings suggest that DNA directly or indirectly interacts with the Smc ATPase and induces it to hydrolyse ATP. If the bacterial Smc complex binds DNA according to a similar mechanism as its eukaryotic descendants, and there is no reason why this should not be so, they may provide a favourable model system to study this process.

### **A model for ATP hydrolysis-dependent transport of DNA into the cohesin ring**

Although important pieces of biochemical information are still missing from our understanding how cohesin binds

to DNA, the available evidence can be used to develop a model how it might work (Fig. 1). A model will only then be useful if it makes predictions that can be tested by further experimentation, so the consequences of the model will also be discussed.

The sequence of events in the budding yeast cell cycle starts in early G1 when Scc1 is absent from cells as it has been cleaved and degraded in the preceding mitosis [3,12]. In the absence of Scc1, Smc1 forms a dimer with Smc3. The Smc dimer is a closed ring by itself, as hydrodynamic analyses have revealed [10]. The ATPase heads tightly associate with each other, probably via bound ATP. Scc3 is not part of this complex because its association with the Smc proteins depends on Scc1 [7].

In late G1, Scc1 is synthesised and associates with the Smc ring. Scc1 also recruits Scc3, thus fully assembling the cohesin complex. The details of cohesin subunit interactions within the newly formed complex, before its binding to DNA, are important to know as they will have crucial implications for any mechanism of DNA transport into the ring. It is currently unclear whether Scc1 already bridges Smc1 and Smc3 heads before DNA binding. It is conceivable that Scc1 initially binds only one of the Smc heads, leaving the cleft between the heads accessible to DNA. One set of experiments to test this question came to the opposite conclusion, that Scc1 bridges Smc1 and Smc3 as soon as it is synthesised [11]. However, those experiments required that Scc1 be cleaved into two halves for analysis, and that may have had unwanted consequences on the interactions (see below). Clarification of the exact architecture of cohesin before it binds to DNA will clearly be important. It might become possible using more sophisticated biochemical experimentation. This could include site-specific cross-linking or fluorescence resonance energy transfer experiments to map distances of the individual parts of the complex.

As mentioned, binding of the cohesin complex to DNA will most likely require the hydrolysis of bound ATP, and DNA itself has the potential to stimulate ATP hydrolysis. A consequence of ATP hydrolysis is expected to be the weakening of the direct interaction between the Smc1 and Smc3 heads [9]. This would open a gap in the Smc ring through which the DNA can enter (Fig. 1). The requirement to separate the Smc heads to allow entry of DNA into the ring would be a gratifying explanation why ATP hydrolysis is required for DNA binding by cohesin. Once the Smc heads separate, the bridging role of Scc1 may become important to ensure that the cohesin ring stays topologically closed. ATP hydrolysis may not only lead to separation of the Smc heads, it may also induce conformational changes. Such changes may lead to stronger Scc1 binding to both Smc1 and Smc3, so that Scc1 now stably bridges the heads. It has been observed that an amino acid motif in the Smc heads that is required for their ATP-dependent dimerisation is also important for tight binding to Scc1 [10,11]. One possible interpretation of this finding is that direct ATP-dependent Smc head dimerisation and Scc1-mediated head

bridging after ATP hydrolysis are alternative and mutually exclusive forms of closing the cohesin ring. The transition between these states could be triggered when DNA contacts the Smc heads and induces ATP hydrolysis. The conformational changes during this transition may also constitute the transport reaction that forces the DNA into the ring (Fig. 1).

How can this model be tested? The model predicts that the Smc heads can be found in two modes of interaction, either direct and bound to ATP, or indirectly via the Scc1 bridge after ATP hydrolysis. EM pictures of human cohesin rings have been obtained that are consistent with both, Smc heads in direct contact or in some distance but bridged by the non-Smc components of the ring [6]. It would be interesting to correlate the state of ATP in the heads to the appearance of the ring. Furthermore, X-ray crystallography and high-resolution structure determination of the Smc heads in complex with Scc1 and bound to either ATP or ADP will be very revealing to describe any structural changes during ATP hydrolysis and how they might contribute to possible DNA transport.

Another important question to be addressed is how many molecules of ATP are hydrolysed for each DNA binding event. Purified cohesin is a very poor ATPase, suggesting that ATP turnover may be slow if cohesin is not engaged in an active DNA-binding reaction. One cycle of ATP binding and hydrolysis may indeed be sufficient to regulate DNA binding and dissociation of cohesin each cell generation. Cohesin could transport DNA into the ring in late G1 and remain DNA bound until in mitosis. Only after cleavage of Scc1 in anaphase might ADP be released from the Smc heads leading to disassembly of the cohesin complex. This scenario would be well adapted to the requirement for very stable association of cohesin with DNA. Alternatively, cycles of ATP binding and hydrolysis might occur more often, regulated by accessory factors. This might be of particular importance in higher eukaryotes when a portion of cohesin is released from chromosome arms without Scc1 cleavage in prophase.

### DNA replication and the establishment of sister chromatid cohesion

We would now like to know how cohesin cannot only bind one strand of DNA, but hold together two replicated sister chromatids. Are two rings required, each holding one sister chromatid, or one ring embracing both? No evidence could thus far be obtained for interactions between more than one cohesin ring on chromosomes [8,10], suggesting that both sister strands are trapped inside one ring. If this is true, the question becomes how a second strand of DNA enters the cohesin ring during DNA replication. As discussed above, it is unclear whether it is at all possible to transport DNA into the cohesin ring more than once per cell cycle. A second cycle of transport would require complete disassembly of the cohesin complex to release ADP and rebound ATP, and this might not be compatible with maintenance

of stable DNA binding. The problem of binding both replication products would be solved if the DNA replication fork simply sled through the cohesin rings that were put around DNA before S-phase. This would leave two replication products trapped inside the same ring without requiring any further DNA transport. It would at the same time provide an intrinsic solution to the crucial requirement to only ever establish sister chromatid cohesion between authentic replication products and never between any other two sequences of DNA.

A number of proteins in addition to cohesin are required for efficient cohesion establishment during S-phase [35–37], and it will be interesting to see whether they help the replication fork slide through cohesin rings. Alternatively, such protein factors could be regulators of a more sophisticated reaction in which cohesin is put around both sister chromatids subsequently as they emerge from the replication fork.

### Opening the ring in anaphase

Sister chromatid segregation at anaphase onset is triggered when separase is activated to cleave the Scc1 subunit in cohesin. If sister chromatids are indeed trapped inside the cohesin ring, how are they released after Scc1 cleavage? Because in the above model Scc1 is essential for bridging the two Smc heads after DNA has been transported, Scc1 cleavage might simply open up the ring [7]. On the other hand, in the absence of Scc1, the two Smc heads can interact directly by binding ATP [10]. Therefore, simply destroying Scc1 may not be good enough to ensure that the cohesin ring stays open long enough to release DNA. Scc1 is cleaved by separase at two specific sites [12], and the C-terminal cleavage product of Scc1 plays a critical role in preventing the Smc heads from interacting [10]. Thus, not cleavage alone, but production of a specific cleavage fragment helps to open the ring. Only after the Scc1 fragment is then removed can the Smc heads interact again. Removal of the Scc1 fragment might be required to release ADP and allow rebinding of ATP to the Smc1 head. The stability of the C-terminal Scc1 fragment is tightly controlled. After a short delay, it is quickly destroyed by targeted proteolysis [38]. If the fragment becomes too long-lived, the cohesin rings cannot close before cohesin needs to bind DNA again in the following cell cycle. This leads to defects in sister chromatid cohesion and consequently to severe chromosomal instability [38,39].

### Acknowledgments

I thank all the members of my laboratory as well as my colleagues in the field for many stimulating discussions. I thank EMBO for support through the Young Investigator Programme.

## References

- [1] K. Nasmyth, J.-M. Peters, F. Uhlmann, Splitting the chromosome: cutting the ties that bind sister chromatids, *Science* 288 (2000) 1379–1384.
- [2] K. Nasmyth, Disseminating the genome: joining, resolving, and separating sister chromatids during mitosis and meiosis, *Annu. Rev. Genet.* 35 (2001) 673–745.
- [3] C. Michaelis, R. Ciosk, K. Nasmyth, Cohesins: chromosomal proteins that prevent premature separation of sister chromatids, *Cell* 91 (1997) 35–45.
- [4] V. Guacci, D. Koshland, A. Strunnikov, A direct link between sister chromatid cohesion and chromosome condensation revealed through analysis of *MCD1* in *S. cerevisiae*, *Cell* 91 (1997) 47–57.
- [5] A. Losada, M. Hirano, T. Hirano, Identification of *Xenopus* SMC protein complexes required for sister chromatid cohesion, *Genes Dev.* 12 (1998) 1986–1997.
- [6] D.E. Anderson, A. Losada, H.P. Erickson, T. Hirano, Condensin and cohesin display different arm conformations with characteristic hinge angles, *J. Cell Biol.* 156 (2002) 419–424.
- [7] C.H. Haering, J. Löwe, A. Hochwagen, K. Nasmyth, Molecular architecture of SMC proteins and the yeast cohesin complex, *Mol. Cell* 9 (2002) 773–788.
- [8] S. Gruber, C.H. Haering, K. Nasmyth, Chromosomal cohesin forms a ring, *Cell* 112 (2003) 765–777.
- [9] K.-P. Hopfner, A. Karcher, D.S. Shin, L. Craig, L.M. Arthur, J.P. Carney, J.A. Tainer, Structural biology of Rad50 ATPase: ATP-driven conformational control in DNA double-strand break repair and the ABC-ATPase superfamily, *Cell* 101 (2000) 789–800.
- [10] S. Weitzer, C. Lehane, F. Uhlmann, A model for ATP hydrolysis-dependent binding of cohesin to DNA, *Curr. Biol.* 13 (2003) 1930–1940.
- [11] P. Arumugam, S. Gruber, K. Tanaka, C.H. Haering, K. Mechtler, K. Nasmyth, ATP hydrolysis is required for cohesin's association with chromosomes, *Curr. Biol.* 13 (2003) 1941–1953.
- [12] F. Uhlmann, F. Lottspeich, K. Nasmyth, Sister-chromatid separation at anaphase onset is promoted by cleavage of the cohesin subunit Scc1, *Nature* 400 (1999) 37–42.
- [13] F. Uhlmann, D. Wernic, M.-A. Poupard, E.V. Koonin, K. Nasmyth, Cleavage of cohesin by the CD clan protease separin triggers anaphase in yeast, *Cell* 103 (2000) 375–386.
- [14] T. Tomonaga, K. Nagao, Y. Kawasaki, K. Furuya, A. Murakami, J. Morishita, T. Yuasa, T. Sutani, S.E. Kearsey, F. Uhlmann, K. Nasmyth, M. Yanagida, Characterization of fission yeast cohesin: essential anaphase proteolysis of Rad21 phosphorylated in the S phase, *Genes Dev.* 14 (2000) 2757–2770.
- [15] I.C. Waizenegger, S. Hauf, A. Meinke, J.-M. Peters, Two distinct pathways remove mammalian cohesin complexes from chromosome arms in prophase and from centromeres in anaphase, *Cell* 103 (2000) 399–410.
- [16] R. Ciosk, M. Shirayama, A. Shevchenko, T. Tanaka, A. Toth, A. Shevchenko, K. Nasmyth, Cohesin's binding to chromosomes depends on a separate complex consisting of Scc2 and Scc4 proteins, *Mol. Cell* 5 (2000) 1–20.
- [17] F. Uhlmann, Chromosome cohesion and separation: from men and molecules, *Curr. Biol.* 13 (2003) R104–R114.
- [18] J. Chen, L. Gang, J. Lin, A.L. Davidson, F.A. Quijoch, A tweezers-like motion of the ATP-binding cassette dimer in an ABC transport cycle, *Mol. Cell* 12 (2003) 651–661.
- [19] H. Venter, R.A. Shilling, S. Velamakanni, L. Balakrishnan, H.W. van Veen, An ABC transporter with a secondary-active multidrug translocator domain, *Nature* 426 (2003) 866–870.
- [20] M. Milutinovich, D.E. Koshland, SMC complexes—Wrapped up in controversy, *Science* 300 (2003) 1101–1102.
- [21] A. Losada, T. Hirano, Intermolecular DNA interactions stimulated by the cohesin complex in vitro: implications for sister chromatid cohesion, *Curr. Biol.* 11 (2001) 268–272.
- [22] A. Kagansky, L. Freeman, D. Lukyanov, A. Strunnikov, Histone-tail independent chromatin-binding activity of recombinant cohesin holo-complex, *J. Biol. Chem.* (2003) (Epub ahead of print).
- [23] M.-A. Hakimi, D.A. Bochar, J.A. Schmiesing, Y. Dong, O.G. Barak, D.W. Speicher, K. Yokomori, R. Shiekhattar, A chromatin remodeling complex that loads cohesin onto human chromosomes, *Nature* 418 (2002) 994–997.
- [24] T. Hirano, The ABCs of SMC proteins: two-armed ATPases for chromosome condensation, cohesion, and repair, *Genes Dev.* 16 (2002) 399–414.
- [25] A. Schleiffer, S. Kaitna, S. Maurer-Stroh, M. Glotzer, K. Nasmyth, F. Eisenhaber, Kleisins: a superfamily of bacterial and eukaryotic SMC protein partners, *Mol. Cell* 11 (2003) 571–575.
- [26] K. Kimura, T. Hirano, ATP-dependent positive supercoiling of DNA by 13S condensin: a biochemical implication for chromosome condensation, *Cell* 90 (1997) 625–634.
- [27] D.P. Bazett-Jones, K. Kimura, T. Hirano, Efficient supercoiling of DNA by a single condensin complex as revealed by electron spectroscopic imaging, *Mol. Cell* 9 (2002) 1183–1190.
- [28] S.H. Yoshimura, K. Hizume, A. Murakami, T. Sutani, K. Takeyasu, M. Yanagida, Condensin architecture and interaction with DNA: regulatory non-SMC subunits bind to the head of SMC heterodimer, *Curr. Biol.* 12 (2002) 508–513.
- [29] T.E. Melby, C.N. Ciampaglio, G. Briscoe, H.P. Erickson, The symmetrical structure of structural maintenance of chromosomes (SMC) and MukB proteins: long, antiparallel coiled coils, folded at a flexible hinge, *J. Cell Biol.* 142 (1998) 1595–1604.
- [30] J. Mascarenhas, J. Soppa, A. Strunnikov, P.L. Graumann, Cell cycle-dependent localization of two novel prokaryotic chromosome segregation and condensation proteins in *Bacillus subtilis* that interact with SMC protein, *EMBO J.* 21 (2002) 3108–3118.
- [31] R.A. Britton, D. Chi-Hong Lin, A.D. Grossman, Characterization of a prokaryotic SMC protein involved in chromosome partitioning, *Genes Dev.* 12 (1998) 1254–1259.
- [32] S. Moriya, E. Tsujikawa, A.K. Hassan, K. Asai, T. Kodama, N. Ogasawara, A *Bacillus subtilis* gene-encoding protein homologous to eukaryotic SMC motor protein is necessary for chromosome partition, *Mol. Microbiol.* 29 (1998) 179–187.
- [33] A. Volkov, J. Mascarenhas, C. Andrei-Selmer, H.D. Ulrich, P.L. Graumann, A prokaryotic condensin/cohesin-like complex can actively compact chromosomes from a single position on the nucleoid and binds to DNA as a ring-like structure, *Mol. Cell Biol.* 23 (2003) 5638–5650.
- [34] M. Hirano, D.E. Anderson, H.P. Erickson, T. Hirano, Bimodal activation of SMC ATPase by intra- and inter-molecular interactions, *EMBO J.* 20 (2001) 3238–3250.
- [35] A. Tóth, R. Ciosk, F. Uhlmann, M. Galova, A. Schleiffer, K. Nasmyth, Yeast Cohesin complex requires a conserved protein, Eco1p (Ctf7), to establish cohesion between sister chromatids during DNA replication, *Genes Dev.* 13 (1999) 320–333.
- [36] R.V. Skibbens, L.B. Corson, D. Koshland, P. Hieter, Ctf7p is essential for sister chromatid cohesion and links mitotic chromosome structure to the DNA replication machinery, *Genes Dev.* 13 (1999) 307–319.
- [37] M.L. Mayer, S.P. Gygi, R. Aebersold, P. Hieter, Identification of RFC(Ctf18p, Ctf8p, Dcc1p): an alternative RFC complex required for sister chromatid cohesion in *S. cerevisiae*, *Mol. Cell* 7 (2001) 959–970.
- [38] H. Rao, F. Uhlmann, K. Nasmyth, A. Varshavsky, Degradation of a cohesin subunit by the N-end rule pathway is essential for chromosome stability, *Nature* 410 (2001) 955–959.
- [39] M.T. Hoque, F. Ishikawa, Cohesin defects lead to premature sister chromatid separation, kinetochore dysfunction and spindle-assembly checkpoint activation, *J. Biol. Chem.* 277 (2002) 42306–42314.