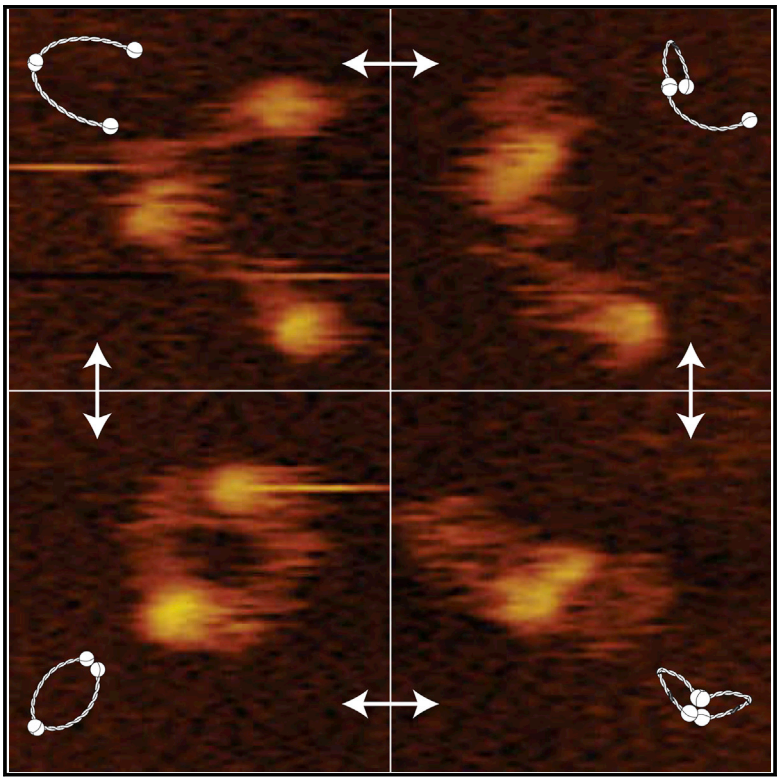
Report

Cell Reports

Condensin Smc2-Smc4 Dimers Are Flexible and Dynamic

Graphical Abstract



Authors

Jorine M. Eeftens, Allard J. Katan, Marc Kschonsak, ..., Essam M. Dief, Christian H. Haering, Cees Dekker

Correspondence

christian.haering@embl.de (C.H.H.), c.dekker@tudelft.nl (C.D.)

In Brief

Eeftens et al. probe the topology and dynamics of condensin's Smc2-Smc4 dimers with high-speed AFM in liquid. They find that the Smc2-Smc4 coiled coils are remarkably flexible and do adopt various conformations that interconvert dynamically over time.

Highlights

- The conformation and dynamics of SMC dimers were imaged with high-speed AFM
- The heads of Smc2 and Smc4 engage with each other and with the hinge dynamically
- The coiled coils of Smc2 and Smc4 are flexible and show extensive fluctuations in time

Eeftens et al., 2016, Cell Reports *14*, 1813–1818
March 1, 2016
http://dx.doi.org/10.1016/j.celrep.2016.01.063

CellPress

Cell Reports
Report

Condensin Smc2-Smc4 Dimers Are Flexible and Dynamic

Jorine M. Eeftens,[1,3] Allard J. Katan,[1,3] Marc Kschonsak,[2] Markus Hassler,[2] Liza de Wilde,[1] Essam M. Dief,[1] Christian H. Haering,[2,*] and Cees Dekker[1,*]
[1]Department of Bionanoscience, Kavli Institute of Nanoscience Delft, Delft University of Technology, Delft 2628 CJ, the Netherlands
[2]Cell Biology and Biophysics Unit, European Molecular Biology Laboratory (EMBL), 69117 Heidelberg, Germany
[3]Co-first author
*Correspondence: christian.haering@embl.de (C.H.H.), c.dekker@tudelft.nl (C.D.)
http://dx.doi.org/10.1016/j.celrep.2016.01.063

SUMMARY

Structural maintenance of chromosomes (SMC) protein complexes, including cohesin and condensin, play key roles in the regulation of higher-order chromosome organization. Even though SMC proteins are thought to mechanistically determine the function of the complexes, their native conformations and dynamics have remained unclear. Here, we probe the topology of Smc2-Smc4 dimers of the *S. cerevisiae* condensin complex with high-speed atomic force microscopy (AFM) in liquid. We show that the Smc2-Smc4 coiled coils are highly flexible polymers with a persistence length of only ~4 nm. Moreover, we demonstrate that the SMC dimers can adopt various architectures that interconvert dynamically over time, and we find that the SMC head domains engage not only with each other, but also with the hinge domain situated at the other end of the ~45-nm-long coiled coil. Our findings reveal structural properties that provide insights into the molecular mechanics of condensin complexes.

INTRODUCTION

Cohesin and condensin protein complexes play central roles in many aspects of chromosome biology, including the segregation of sister chromatids during cell divisions, compaction of chromosomes, and regulation of gene expression during interphase (reviewed in Aragon et al., 2013; Hirano, 2006). Although functionally different, cohesin and condensin have similar architectures: both complexes are composed of two different SMC subunits and a subunit of the kleisin protein family. Together, these three proteins form a ring-like structure that is conserved from bacteria to eukaryotes. The protein chain of each SMC protein folds back onto itself to form an ~45-nm-long antiparallel coiled coil, which connects a globular "hinge" domain at one end to an ATPase "head" domain, created by the association of N- and C-terminal protein sequences, at the other end (Figure 1A). Two SMC proteins form a heterodimer by the association of their hinge domains: Smc1-Smc3 in the case of cohesin and Smc2-Smc4 in the case of condensin (Anderson et al., 2002). In addition, the head domains of the two SMC subunits can associate in the presence of ATP. The functional roles of ATP binding-mediated dimerization and hydrolysis-dependent dissociation of the two head domains have remained largely unclear. Both cohesin and condensin have been suggested to bind to chromosomes by encircling chromatin fibers topologically within their SMC-kleisin rings (Cuylen et al., 2011; Haering et al., 2008).

The conformation and dynamics of SMC dimers are of great importance, since they are thought to mechanistically determine the biological function of all SMC protein complexes. Accordingly, there have been numerous efforts to gain insight into the configuration of the SMC dimers. Electron microscopy (EM) images of cohesin complexes suggest that the Smc1–Smc3 coiled coils emerge from the hinge domain in an open conformation, resulting in V- or O-shaped arrangements with the two coils separated along most of their lengths (Anderson et al., 2002; Haering et al., 2002; Huis in 't Veld et al., 2014). V-shaped conformations were also observed for condensin's Smc2-Smc4. However, in a large fraction of molecules the Smc2-Smc4 coiled coils seemed to align, resulting in rod- or I-shaped rather than V-shaped conformations (Anderson et al., 2002; Yoshimura et al., 2002). Support to the notion that condensin's SMC coiled coils tightly associate with each other came from a recent crystal structure of the Smc2-Smc4 hinge domains and parts of the adjacent coiled coils, as well as from chemical cross-linking experiments (Barysz et al., 2015; Soh et al., 2015). Small Angle X-ray Scattering (SAXS) experiments implied that also the SMC subunits of cohesin and prokaryotic SMC complexes form I-shaped molecules in solution (Soh et al., 2015). These contradicting results indicate that it is still unclear which configurations SMC dimers adopt in vivo, and under which circumstances conformational changes might occur. The major disadvantages of all methods that have so far been used to study the configuration of SMC molecules are that they probed the protein structure either in highly artificial environments (e.g., dried in vacuum or packed into a crystal lattice) or in a kinetically trapped state (e.g., by cross-linking).

Atomic force microscopy (AFM) has proved to be a powerful tool to visualize biomolecules and to study their mechanical properties at nanometer resolution without the need for labeling. Importantly, it can also be carried out in aqueous solution under physiological conditions. Recent technical advances have made it possible to observe single molecules in action with high-speed AFM, reaching frame rates of up to 20 frames per second and thereby allowing imaging in real time (Ando et al., 2001; Katan and Dekker, 2011). Here, we use high-speed AFM in liquid, in