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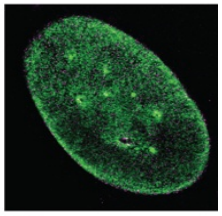

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REVIEW

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Closing the loops: chromatin loop dynamics after DNA damage

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ABSTRACT

Chromatin is a dynamic polymer in constant motion. These motions are heterogeneous between cells and within individual cell nuclei and are profoundly altered in response to DNA damage. The shifts in chromatin motions following genomic insults depend on the temporal and physical scales considered. They are also distinct in damaged and undamaged regions. In this review, we emphasize the role of chromatin tethering and loop formation in chromatin dynamics, with the view that pulsing loops are key contributors to chromatin motions. Chromatin tethers likely mediate micron-scale chromatin coherence predicted by polymer models and measured experimentally, and we propose that remodeling of the tethers in response to DNA breaks enables uncoupling of damaged and undamaged chromatin regions.

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Introduction

Chromatin is a dynamic polymer of DNA bound to histones and other associated proteins. At its most fundamental level, chromatin has a beads-on-string structure, with beads consisting of nucleosomes (i.e., histone octamers) separated by linker DNA. At the global scale of the nucleus, chromosomes in metazoan and plant nuclei occupy distinct territories during interphase. These territories are nonrandom, yet highly variable, even within homogeneous cell populations [1]. The concept of genomic territories applies across orders of magnitude down to budding yeasts, where genomic loci occupy sub-nuclear domains [2], and bacteria with chromosomal loci localizing to specific regions of the cell [3]. Higher-order chromatin organization between these extreme structural scales is more mysterious but increasingly understood thanks to advances in imaging and high-throughput chromosome conformation capture (Hi-C) methods [4–6]. Nucleosomes form heterogeneous clusters (or 'clutches') of approx. 100 nm in diameter [7–9], departing from the pervasive textbook notion of the '30 nm fiber'. At a yet higher, sub-micron scale, chromatin loops bring distant genomic loci in close contact [7,8,10–12]. Contacts from loops (or more accurately from 'loop domains' consisting of loops within loops) are identified as topologically associating domains (TADs) by chromosome conformation capture (Hi-C) [13] and represent interacting genomic regions in the range of 1 Mb. The direct visualization of a prototypical chromatin loop by FISH combined with super-resolution microscopy confirmed contact maps previously established by Hi-C but also revealed heterogeneity between individual loop structures [14].

While chromosomal loops have been reported in the early 1900's [15], it is only in the last decade that their role in chromosome compaction, transcriptional regulation, and formation of gene bodies has been appreciated. Chromatin loops are the means of condensing chromosomes into units suitable for high fidelity segregation in mitosis [16]. Loops also provide a mechanism to transform a linear array of genes and regulatory elements into three-dimensional structures that bring into proximity the suite of regulatory elements

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specific for a specific cell type [17]. This significantly expands the regulatory capacity of an organism. The impact of the loop structure is observed in the changes that accompany developmental progression as well as disease states [18]. Loops also provide a mechanism to compartmentalize functional domains. The most robust of these compartments is the nucleolus. The nucleolus is enriched in condensin, and while nucleolar formation is a multi-step process, DNA looping is sufficient to segregate the nucleolus from the remainder of the nucleus [19,20]. In a remarkable feat of genome paleontology, the 3D DNA loop structure responsible for chromosome territories and gene bodies was found to be preserved in a 52,000-year-old mammoth [21].

Chromatin loops are formed by tethers holding the DNA strands at their base. Cohesin is a key tether in mammalian and yeast interphase nuclei which generates loops by extruding chromatin [22–24]. Loop extrusion is restricted by CTCF which binds to boundary elements and insulators and brings the extrusion process to a halt. Whereas chromatin loops define TADs and are clearly visualized in contact maps derived from cell populations, loop positions vary at the single cell level [25–27]. This mirrors the high level of stochasticity in transcription (and in other genomic processes including the DNA damage response) between cells and over time [5,28], and the fact that loops will also arise through the natural fluctuations of the polymer chain [29,30]. The proportion of loops that arise through active loop extrusion or from stochastics of chain fluctuation remains to be determined.

In addition to internal tethers, external tethers anchor chromatin to structural hallmarks of the nucleus. Those hallmarks differ across species. In budding yeast, centromeres and telomeres are tethered at the nuclear periphery in the 'Rabl' configuration [31,32]. This conformation may limit chromosome entanglement [33] and dictates the overall topology of the yeast genome. In metazoan nuclei, domains of chromatin are also tethered at the nuclear periphery, to the lamina. These lamina-associated domains (LADs) help define global genome organization and correspond to regions with high chromatin compaction and low gene expression [34]. Chromatin association with internal nuclear domains, including the nucleolus and splicing factor speckles, further influences global genome organization in ways that are highly dynamic and cell type-dependent [4]. Structural elements residing in the interior of the nucleus, such as the internal pool of LaminA/C and the mitotic apparatus protein (NuMA), also bind chromatin [35]. These internal tethers affect chromatin organization and mobility, thereby impacting genome functions, and notably maintenance [36–43]. Motions of chromatin appear to be tuned to genomic functions including replication, transcription, and repair.

Multiple approaches have been developed to measure chromatin motions across scales. They have been reviewed recently [6,44–46] and are summarized in Figure 1. These approaches include labeling and tracking single nucleosomes [49–51]. Sparse labeling of 100–200 nucleosomes/nuclear plane can be achieved by incubating cells expressing HaloTag labeled histones with highly diluted fluorescent HaloTag ligands (HTL). Image sequences are generally captured using light sheet microscopy to minimize the out-of-focus background. Another classic approach is to track engineered chromatin loci [52]. Repeats of the lac operator (*LacO*) stably integrated in the genome are detected by the lac repressor labeled with GFP (LacI-GFP; [53]). The ANCHOR system is a good alternative based on the bacterial partitioning complex [54,55]. It enables tracking of individual genomic loci in which *ParS* sequences ('INT'; ~1 kb) have been inserted. ParB-GFP nucleates at the INT sequence, then spreads on flanking chromatin. Unlike the *LacO*/LacI-GFP system, ANCHOR does not inhibit transcription in the labeled region. Finally, CRISPR-based imaging is increasingly used to track endogenous genomic loci. It is a flexible approach based on nuclease-dead Cas9 (dCas9) to probe chromatin motions at specific genomic regions, defined by the sequences of the guide RNAs (sgRNAs). Early implementations using GFP-tagged dCas9 were restricted to genomic regions with large numbers of repeats (such as telomeres) or required an assortment of back-to-back sgRNAs. New strategies have multiplied the number of chromophores on the dCas9/sgRNA complex, for detection of shorter genomic loci with fewer sgRNAs [56–59].

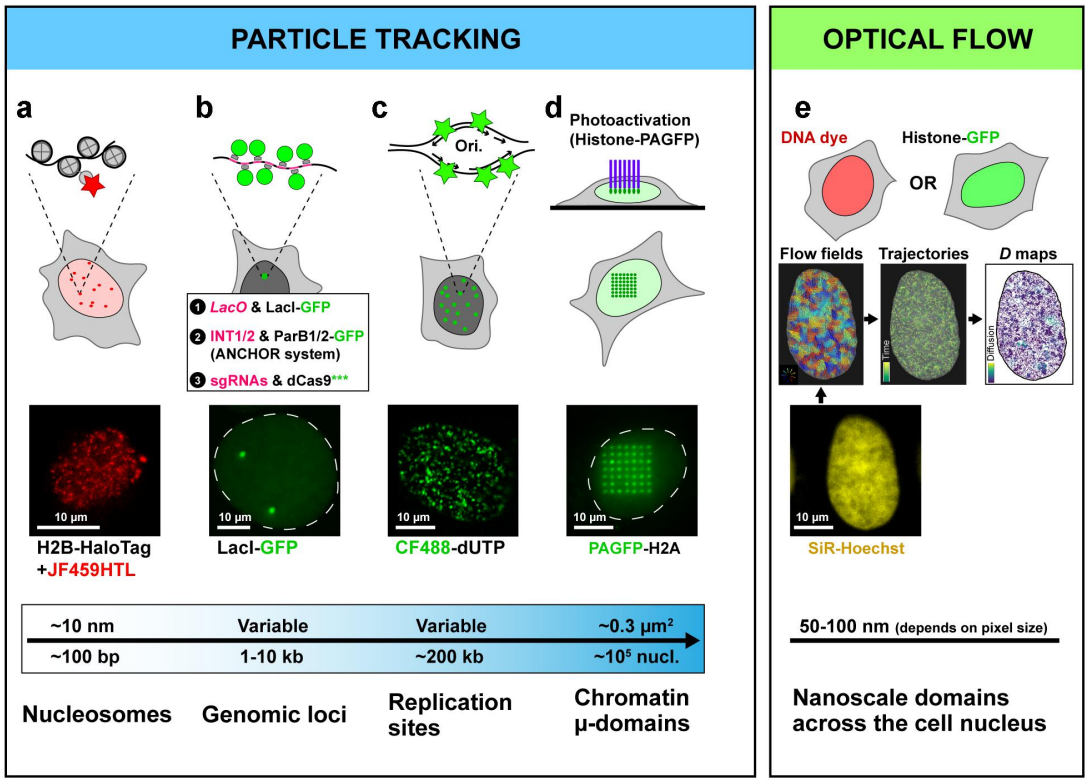

Figure 1. Methods to quantify chromatin motions across physical scales. (a) single-nucleosome labeling for single-particle tracking (SPT) at the nanoscale. HTL, HaloTag ligand. (b) SPT of chromatin loci. Labeling approaches include (1) LacI-GFP on lac operator (*LacO*) repeats, (2) the ANCHOR system, and (3) fluorescent nuclease-dead Cas9 (dCas9). (c) tracking of early DNA replication origins (Ori.) by pulse incorporation of fluorescent nucleotides. (d) Time-lapse imaging of photoactivated grids of chromatin microdomains. (e) optical flow measurements of densely labelled chromatin. This panel of methods is not exhaustive. For instance, multiple studies have followed the motions of specific chromatin domains or compartments, including centromeres, telomeres and DNA damage sites (ref [36,47,48], etc.).

To capture chromatin motions throughout the nucleus, early DNA replication origins can be labeled by pulse incorporation of fluorescent nucleotides and the signals used for single-particle tracking (SPT) [43,47,60]. At a larger, sub-micron, scale, photoactivated chromatin microdomains can be followed. An approach is to use diffractive optics producing tight grids of laser beamlets to activate photoactivatable fluorescent proteins tagged to histones. SPT of the photoactivated domains enables mapping of chromatin motions and analyses of motion correlations [43]. Finally, optical flow measurements are an alternative to SPT to study chromatin dynamics. The approach requires dense labeling of chromatin (DNA dyes or fluorescently tagged histones). It produces flow fields that are used to compute maps of biophysical characteristics, including diffusion constants and anomalous coefficients [61–63].

This review focuses on chromatin motions during the DNA damage response, with an emphasis on chromatin loops dynamics. Chromatin loops are considered to be major contributors of chromatin motions. They are constantly remodeled by dynamic tethering and extrusion, as well as by random fluctuations. We also discuss the convergence of experimental evidence for coordinated (coherent) chromatin motions, altered in response to DNA damage, and propose mechanisms incorporating the timescale of loop fluctuations, that together with repair factor nucleation, may explain this phenomenon.

Chromatin motions in response to DNA damage

DNA damage is a major disruptor of normal genome functions including transcription and DNA replication. Lesions affecting both DNA strands such as inter-strand cross-links and double-strand breaks (DSBs) are particularly deleterious as they can cause mutations, genome rearrangements, and cell death. Accordingly, cells have evolved elaborate mechanisms to cope with DSBs, which involve extensive chromatin remodeling at and around DSBs [64], a hierarchical recruitment of repair factors [65,66], and eventually the restoration of the initial chromatin state [67].

Remarkably, cell responses to DSBs appear to be highly heterogeneous, even within a homogeneous cell population. This is illustrated in Figure 2 with strikingly different kinetics of DSB repair foci accumulation and resolution in cells within a uniform irradiation field. Heterogeneity in genome organization and chromatin motions may contribute to these widely different outcomes. Even at steady state and within the same cell cycle phase, the mobilities of yeast genomic loci are highly heterogeneous [68,69]. Cell-to-cell variability in chromatin diffusion is also apparent in mammalian cells, where 3–5-fold differences in chromatin motions between cells with 'fast' vs 'slow' chromatin are typically observed [43]. In addition to this inter-cell heterogeneity, the measured values of chromatin motions are heterogeneous within a cell nucleus, due to the combination of measurement errors, the stochastic nature of polymer motions, and actual biological differences. By tracking tight grids of photoactivated chromatin microdomains (Figure 1d) [70], we estimated with repeated measurements that

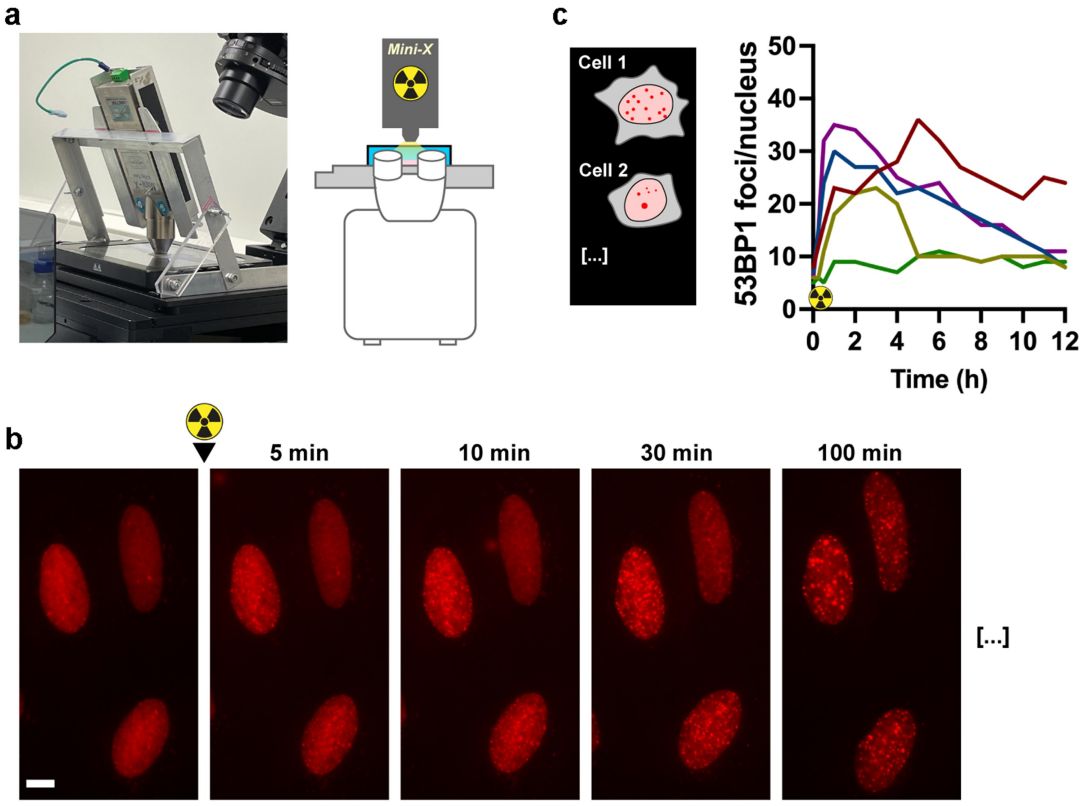

Figure 2. Stochasticity in DNA damage response outcomes. a) X-ray irradiation source mounted on a fluorescence microscope to monitor the DNA damage response in single cells with high temporal resolution. b) Time lapse images of a cell nucleus expressing a DSB reporter (mCherry fused to the C-terminal portion of 53BP1) showing radiation-induced formation of DNA damage foci. Scale bar, 10 µm. c) Heterogeneity in cell responses to X-ray irradiation (5 Gy). Counts of DSB foci following irradiation are shown for five cells from the same cell population.

biological differences contribute to about half of this spatial heterogeneity in chromatin motions [43].

Chromatin motions are profoundly impacted by DNA damage [45,52,71,72]. The effect of a DSB is particularly clear in yeast where a single break causes local and global acceleration of the polymer [68,73–79] (Figure 3a). Yeasts predominantly rely on homologous recombination repair (HRR), a pathway using the sister chromatid as a template in the repair process. Elevated motions of chromatin may facilitate homology search and strand invasion during HRR [73,80,81]. Recruitment of energy-consuming enzymes, such as Rad50, Rad51, and Rad54 enhance polymer fluctuations through the increase in molecular bombardment. Increasing motion through ATP consumption is analogous to using temperature to push molecules into a heightened state of motion. In addition, recent studies have found that Rad51 nuclear filaments may participate in the search and capture mechanisms analogous to microtubule dynamic instability in chromosome capture [82]. Rad51 coats single-stranded DNA

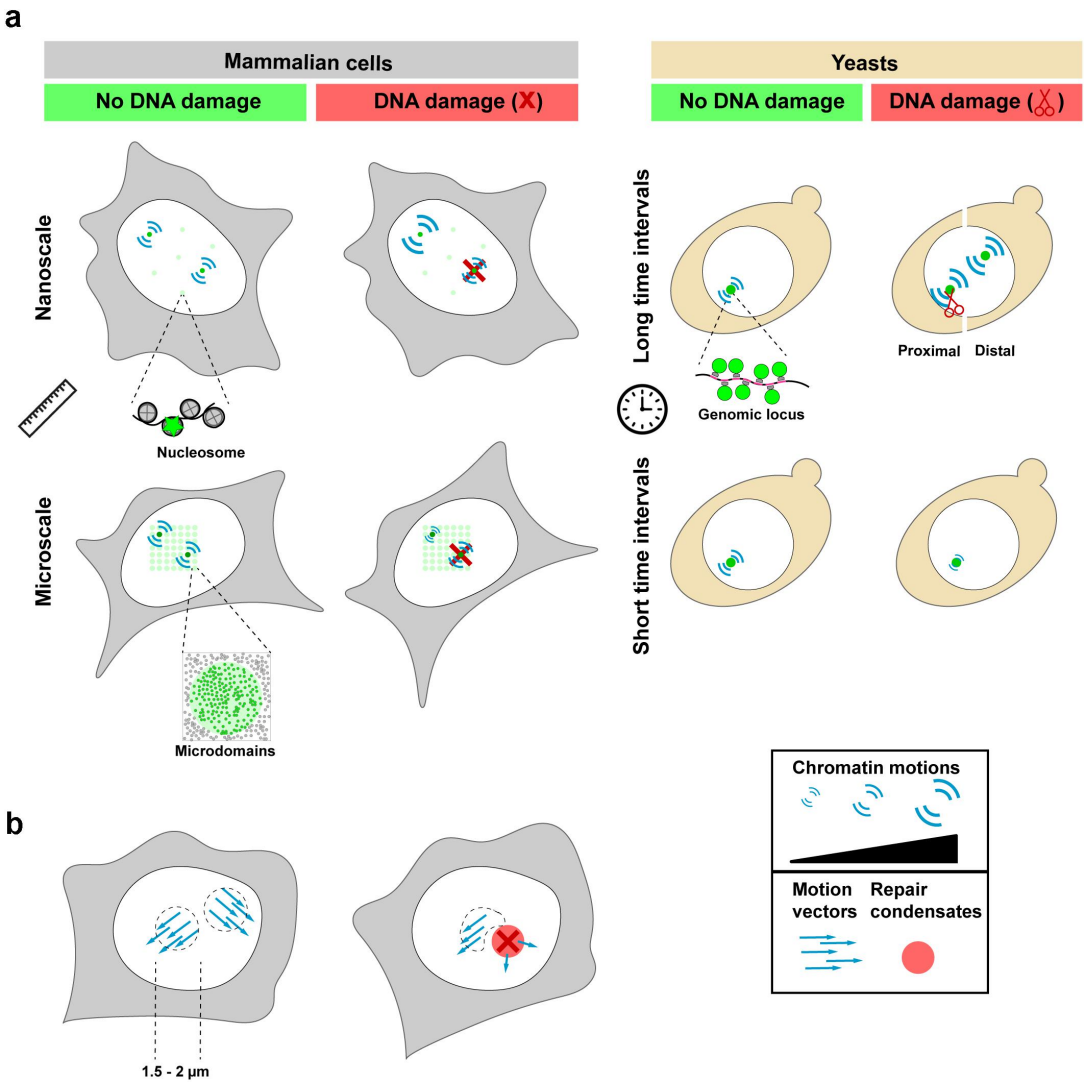

Figure 3. Context- and scale-dependent effects of DNA damage on chromatin motions. a) Effect of DNA damage ('X' or scissors/enzymatic) on chromatin dynamics in mammalian cells and yeasts as a function of (1) the position in the cell nucleus relative to DNA damage sites, (2) the physical scale of chromatin, and (3) the temporal scale considered. b) Chromatin cohesion (i.e., correlated motions) is reduced in response to DNA damage, potentially reflecting uncoupling of chromatin motions in damaged regions due to chromatin tether remodeling and/or the formation of repair factor condensates.

following the excision of the complementary strand at sites of breaks. The resulting filaments are dynamic polymers, that through extension, compaction, and bending, have the ability to significantly increase the search area.

The situation is more perplexing in animal cells which have a complex nuclear organization and context-dependent DSB repair pathway usage [66]. In specific situations, such as DNA breaks occurring in heterochromatin rich in repetitive elements, damaged DNA is actively relocated to the nuclear periphery or to nuclear regions that may be safer for repair and a shift from random to directed motions ensues (reviewed in [45,83,84]). All DSBs are not equal, and the nature of the break site(s) may influence the dynamics of damaged chromatin. For example, cells exposed to densely ionizing radiation suffer extensive damage, resulting in complex and clustered DSBs with different repair outcomes than cells with more uniformly distributed breaks [85,86]. DNA repair foci and dysfunctional telomeres (which resemble DSBs) are more mobile than non-damaged chromatin regions [47,87]. Similarly, UV-induced DNA damage and DNA damage caused by replication inhibition lead to faster motions of single nucleosomes [50,88]. This may reflect the global reduction in transcription activity during the DNA damage response; more specifically, the dissolution of RNA polymerase II 'transcription factories', which constrain chromatin at the nanoscale [50]. One might expect different behaviors of nucleosomes in damaged and non-damaged regions of the nucleus. There is evidence of transcription at DSBs actively contributing to the repair process [89]. Moreover, cohesin loading at DSBs [90,91] may locally reduce nucleosome motions. Cohesin recruitment at DSBs indeed ensures end-tethering, at least in yeast [92].

The picture is very different when considering chromatin at the microscale: measurements of microdomains of chromatin with the photoactivation method described above showed *decreased* chromatin motions after damage globally, yet faster motions near break sites [43,93]. This effect may be caused by an uncoupling of chromatin dynamics by the formation of DNA damage-induced 'damage compartments' [94]. Reduced motions in undamaged regions may reflect the demobilization of energy-intensive activities normally associated with transcription, to allocate resources for genome maintenance at DNA breaks. Higher chromatin motions in restricted regions around break sites may promote the repair process, as discussed above in yeast models.

The effect of DNA damage on chromatin mobility is not only location-dependent but also time scale-dependent, at least in yeast. By imaging chromatin loci with different time intervals, Mine-Hattab and colleagues [69] found that, in response to DNA damage, chromatin becomes more mobile at large time scales but less mobile at short time scales. This effect could be explained by Rad51 nucleoprotein filaments stiffening the polymer, and therefore restricting small chain fluctuations while increasing overall motions to enhance the 'search algorithm' in the context of homologous recombination. It remains to be seen if the same applies to mammalian cells that predominantly rely on non-homologous end joining for DNA repair, which is independent of Rad51. Hence, shifts in chromatin dynamics in response to DNA damage depend on (1) the location within the cell nucleus, (2) the physical scale of chromatin considered, and (3) the temporal scale from the analyses (Figure 3a).

DNA damage reduces chromatin coherence

Polymer models that include chromatin tethers predict coherent motions on the micron scale [95,96]. These predictions are consistent with experimental results. Optical flow measurements of labeled histones or DNA yield flow fields with long-range correlations over several micrometers [12,61,62,97]. Correlation analyses of particle tracking data also indicate correlated motions over 1.5–2 μm [43,98]. Coherence of chromatin motions is not restricted to individual chromosome territories; it was also measured between chromatin domains belonging to different chromosomes [43,61]. DNA damage reduces chromatin motion correlation [43,61], which may reflect uncoupling of chromatin motions at damaged sites (Figure 3b). This interpretation is consistent with Hi-C measurements showing reinforced contacts within TAD containing a DSB but reduced contacts between these DNA damage-containing TADs and undamaged neighboring regions [94].

While the molecular mechanisms for correlated chromatin motions are not clear yet, chromatin–chromatin interactions, both via 'sticky' nucleosome contacts and chromatin tethers, are likely determinants. Remodeling cross-links as a consequence of DNA damage would contribute to a reduction in chromatin motion correlation through the stoichiometric shift of tethers from their pan-genomic distribution to a more restricted distribution at sites of damage.

Correlated chromatin motions have also been measured at the nanoscale, with single nucleosome tracking [60]. By labeling nucleosomes with two different colors, it was possible to visualize and quantify correlated movements of neighboring nucleosomes [60]. At this scale, Nozaki *et al.* found that nucleosome motions are highly correlated within a distance of 150 nm, interpreted as nucleosome clusters [60], which have been identified in super resolution images of chromatin [7–9]. It will be very interesting to analyze the impact of DNA damage on chromatin coherence at the nanoscale, based on single-nucleosome tracking.

Loop dynamics, a framework to understand chromatin motions in the DNA damage response

As mentioned in the introduction, TAD boundaries vary from cells to cells [28], which reflects the dynamic nature of chromatin looping and heterogeneous epigenetic states. The kinetics of loop formation is likely critical for integrating the organization of chromosomes with key biological processes. Chromatin loops are constantly fluctuating in volume (expansions and contractions), physical size, and density (number of loops per kilobase pair). The kinetics of loop formation and fluctuations will depend on these parameters, as well as protein binding, phase state, proximity to tethers (centromeres or telomeres in yeast) and spatial position. The response to DNA damage is likely to encompass changes in loop dynamics that enhance recombinational or end-joining repair mechanisms.

Loop structure

To gain insights into loop size regulation and distribution, we have used bead-spring polymer chain models of chromatin and superimposed the activity of an SMC (structural maintenance of chromosome) complex on chromatin. We found that (1) the stiffness of the substrate (flexibility as well as compaction of the nucleosome polymer chain), (2) the spring properties of a chromatin cross-linking complex (tensile stiffness), and (3) the strength of internal or external anchors (tethers) cooperatively dictate loop size distributions and volumes within a chromatin domain (Figure 4). When DNA tethers are highly constrained, the loop sizes are determined by the stiffness of the condensin and/or cohesin spring. If the cross-linking complex is weak, DNA tethers prevent the cross-linkers from making a loop. If the cross-linking complex is strong, (greater than that of the DNA tethers) loops are able to form. When DNA tethers are loose or unconstrained, the regulation of loop size is conferred through chromatin stiffness. Floppy chromatin with short persistence length (Lp < 50 nm) will adopt a random coil, while stiffer chromatin (Lp > 200 nm) is the dominant determinant in loop size. Tethers provide additional inputs to the distribution of loops, and, unexpectedly, tethering strength affects how

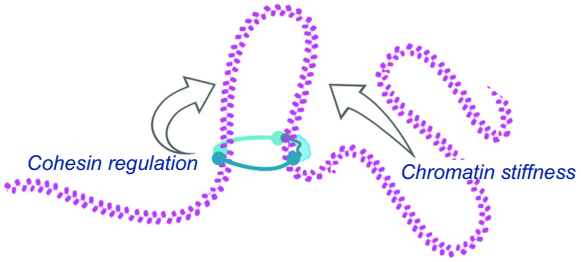

Figure 4. Determinants of chromatin loop structure. Chromatin is a long chain polymer of repeating nucleosome subunits (purple). The chain is inherently floppy, as defined by its short persistence length (Lp = 50 nm, the length scale over which the ends of the fluctuating chain are correlated). There are two modes of loop formation. One is from stochastic chain fluctuations (shown on the right), when distal regions of the chain come into contact. The second is when active cross-linkers, such as condensin and/or cohesin, actively extrude loops (shown on the left). The size, distribution, duration, and structure of loops are dictated by 1) stochastics of chain fluctuation that depend on the physical properties of the chain itself (stiffness, compaction), 2) the loop extruders (abundance, binding affinity and tensile strength) and 3) constraints through entanglements (internal tethers) or anchoring (external tethers such as nuclear lamina, microtubules) that provide resistance to the action of cross-linkers.

chromatin conforms within a topological domain [99]. In addition to modulation of the physical properties of the substrate (i.e., the polymer chain), loop size and regulation are modulated through the action of SMC complexes and boundary elements such as CTCF (CCCTC-binding factor). SMCs promote loop formation through the extrusion of one strand relative to another [24,100,101]. The site of extrusion is dictated through the action of CTCF bound to key regulatory sites unique to specific cell types [17,18]. Interestingly, CTCF binds in a tension-sensitive fashion [102], indicative of the ability of CTCF to sense force-induced features of the DNA chain that distinguish strands under strain.

Loop dynamics

Loop formation is likely a combination of active extrusion by condensin and cohesin, as well as loop structures that arise through random chain fluctuations and persist through the action of cross-linkers. It is also likely that the loop size and configuration (collapsed or extended) are dynamic. Measurements from fixed cells give us the average size, but in live cells as well as in polymer models, there is a genome-wide 'pulsing' of loops, which constantly elongate and shorten, as well as collapse or extend. These features are likely to have critical biological functions. Microtubules are the classic example for robust stochastics of polymer growth and shortening, providing the mechanism to ensure fidelity of chromosome segregation [103]. Another example of potential biological consequences of loop fluctuation come from studies of RecA binding to extended or compact substrates. RecA protein assembly at sites of DNA damage establishes a kinetic proofreading cascade that enables the cell to mount an SOS DNA repair response to ssDNA [104,105]. RecA proofreads the ssDNA through its binding fluctuations. There is a time-scale of protein nucleation at sites of ssDNA (damage) dependent on RecA concentration and a second time-scale dependent on the ssDNA polymer fluctuation (i.e. the tendency to adopt a random coil). At low RecA concentration, nucleation events will be rare, and filaments will not assemble. If polymer fluctuations are too rapid, ssDNA will collapse, and filaments will not assemble. The constant dynamic between chain fluctuation and protein binding to the chain provides a means of regulation. Tuning the timescales of protein nucleation (like RecA) and polymer fluctuation is critical to filament assembly and biological response. Loop formation in this example is central to the DNA damage response and action at the scale of nucleotides. Loops functioning at larger scales in mitotic chromosomes adopt a bottlebrush configuration that dictates mechanisms of chromosome compaction [106–108].

Network organization

Chromatin structure is highly influenced by the action of SMC proteins, in particular, cohesin and condensin. The physical behavior of these proteins is only now becoming clarified and quantified, performing actions such as crosslinking within and between chromosomes and loop extrusion within chromosomes. The polymer model framework of chromatin provides a means to explore the relationship between the nanoscale actions of these proteins and the resulting macroscale dynamic structure in the nucleus. Crosslinking action can be modeled by adding additional springs between pairs of beads that stochastically form (when they are within a prescribed distance) and break according to a prescribed mean timescale τ_{on}, which dictates the mean duration of a transient bond. To investigate chromatin network organization mediated by crosslinking, we added dynamic crosslinkers to the subset of beads within chromosome XII that comprise the nucleolus. The nucleolus was chosen for modeling due to the specificity of condensin binding at the rDNA repeats [109], and the repeat organization of rDNA genes, conducive to the repeating bead-spring configuration in polymer modeling. Whether these crosslinkers are present or not, and whether they are dynamic or fixed, result in different configurations of the nucleolus, as shown in Figure 5, and provide a physical basis for understanding heterogeneity

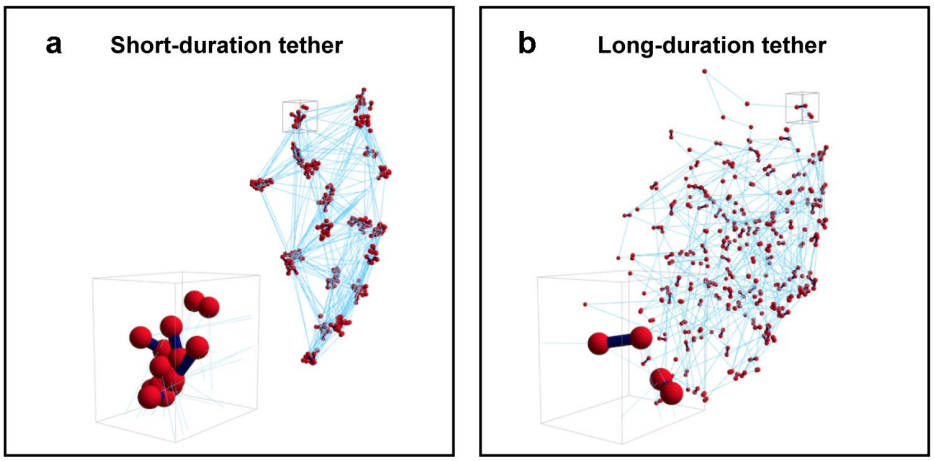

Figure 5. Network organization with different chromatin tether dynamics and properties. Snapshots of 3D simulations show bead distributions in models with different mean durations of the transient chromatin tethers. These durations were either short, with $\tau_{on} = 0.09$ s (a) or long, with $\tau_{on} = 90$ s (b). Red spheres represent bead positions, dark blue lines represent transient crosslinks between beads both inter- and intra-chain, light blue thin lines represent intra-chain connections between neighboring beads. The inserts are blowups of small volumes around bead clusters.

and dynamics in the nucleolar structure in live cells [19]. Hence, SMC activities and by extension loop dynamics profoundly influence chromatin networks.

Correlated motions

Can loop dynamics explain the coherent chromatin motions measured by particle tracking and optical flow analysis (see above)? Depending on the timescale of protein cross-linking vs. chain fluctuation, the network organization is very different. At fast cross-linking regimes, nodes are evident, whereas at slow cross-linking regimes, the network is homogenized. This has major consequences for the behavior of chains and genome regulation. At a high cross-linking density (Figure 5a) coherent chromatin motion will be isolated to nodes. That is, what is happening in one node will be shielded from others. In contrast, at low cross-linking density (homogeneous landscape; Figure 5b) coherent chromatin motion can percolate throughout the genome. In the case of damage, remodeling of cross-linkers to sites of damage, with cohesin recruitment to DSBs [90,91], would reduce coherent motion throughout the genome (as discussed above) while simultaneously increasing motion at repair domains. The distribution of cross-linkers may also dictate the motion of protein assemblies through the chromatin network, providing further means of channeling resources for repair of DNA damage [110].

Future perspectives

Most knowledge on chromatin loop dynamics comes from *in vitro* assays and *in silico* work. Moving forward, strategies to capture loop dynamics in living cells are needed. CRISPR-based imaging of chromatin loci (Figure 1b) now enable the visualization of defined pairs of genomic regions [111]. Fluctuations in the distance separating these loci can be measured precisely using paired particle tracking or other biophysical methods to probe distances, including FRET and molecular beacons. By labeling two regions close to CTCF binding sites, one may be able to visualize loop 'pulsing' (Figure 6a). However, an important caveat of CRISPR-based imaging is that it (still) entails relatively large molecular assemblies on the chromatin to reach the critical levels of chromophores needed for imaging. These dCas9 complexes add drag to chromatin motions and may cause steric interferences at the level of chromatin loops. The same consideration applies to other methods classically

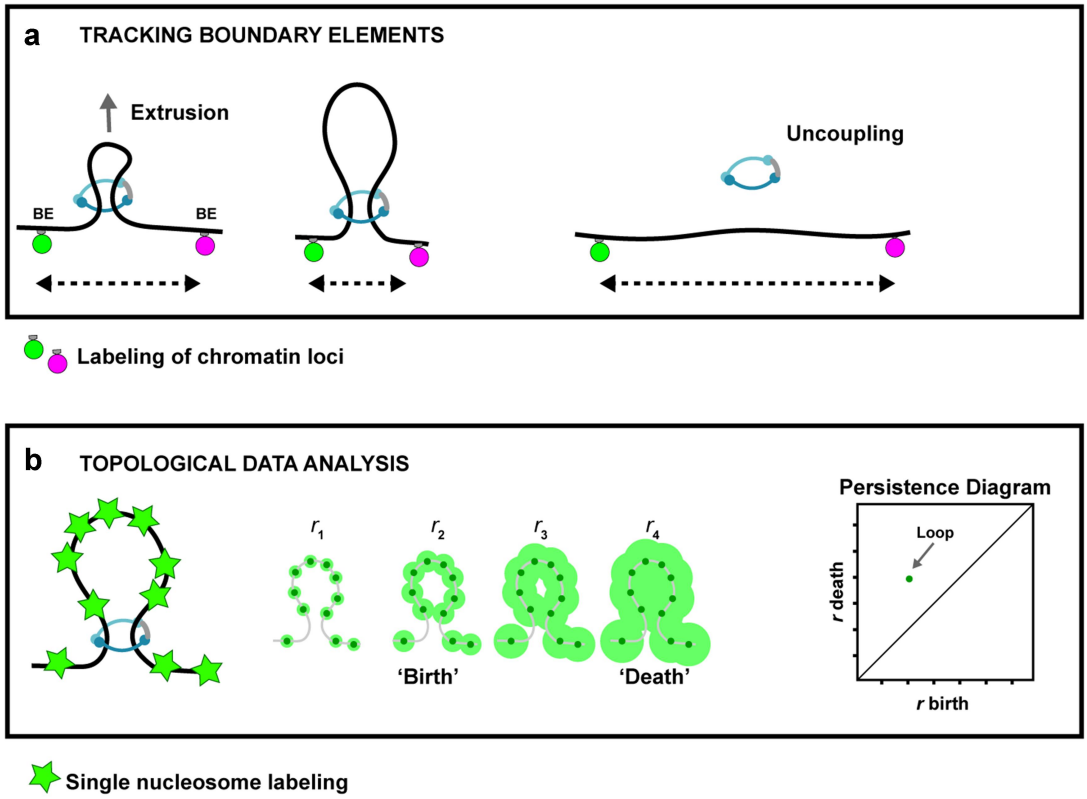

Figure 6. Strategies to study chromatin loop dynamics in live cells. a) Labeling chromatin loci (e.g., with dCas9-based approaches) close to loop boundary elements (BEs) may enable inference of loop dynamics with paired particle tracking, FRET, or a molecular beacons strategy. Loop extrusion would shorten the distance between BEs, whereas uncoupling of the tether would rapidly increase BE distances. b) Topological data analysis (TDA), which detects spatial patterns from image time series, is a promising new approach to study chromatin loop characteristics in live cell nuclei. r_n is the radius used to define points within the point cloud. See text for details.

used to track chromatin loci (*LacO* - LacI and INT – ParB). It will, therefore, be important to develop alternative approaches to visualize chromatin loops *in situ*, ideally via single-nucleosome tracking which can be done in quasi-native chromatin states. Toward this goal, we are now evaluating topological data analysis (TDA) as a new approach to measure loop formation and resolution [112] (Figure 6b). TDA provides a means to detect spatial patterns from time-series analysis. A loop can be considered as a topological feature. It is defined by a set of data points enclosing a hole within a cloud of single particles (point cloud). The persistence of a loop, a measure of its size and stiffness, is determined by its ability to maintain its topological identity as the radius used to define points within the point cloud increases. Persistence homology is the computational tool used to quantify the persistence of topological identity. We anticipate that the methodology will enable the field to assess the number, duration, and spatial distribution of loops in an individual cell. These features cannot be discerned from population studies of fixed specimens.

It will also be important to reconcile seemingly contradictory observations made at different physical and temporal scales with multi-scale imaging approaches. For example, it is possible to simultaneously track single nucleosomes within the context of larger chromatin domains [60]. Future studies combining micro-scale measurements of chromatin motions, for example, with the photoactivation paradigms [43,93,113,114] and nanoscale tracking of single nucleosomes will also be informative. Nested approaches are powerful to

probe different time scales in these experiments, whereby sequences of movies are collected to interrogate both the fast and slow kinetics of chromatin. These approaches require the acquisition of a large number of image frames, which can be enabled by AI-powered image restoration [51].

These technological advances to describe chromatin dynamics in the DNA damage response (and beyond) should enable the field to answer key open questions. These include the impact of chromatin motions across spatial and temporal scales on DNA repair outcomes, the mechanisms responsible for micro-scale chromatin coherence, and the roles of correlated chromatin motions in genomic functions.

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Disclosure statement

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Author contributions

PAV and K Bloom wrote the first draft of the manuscript and prepared the illustrations. JL and K Bonin critically reviewed and complemented the article. All authors have read and approved the final work.

Data availability statement

Data sharing is not applicable to this article as no new data were created or analyzed in this study.

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References

[1] Cremer T, Cremer M. Chromosome territories. Cold Spring Harb Perspect Biol. 2010;2(3):a003889. doi: 10.1101/cshperspect.a003889
[2] Berger AB, Cabal GG, Fabre E, et al. High-resolution statistical mapping reveals gene territories in live yeast. Nat Methods. 2008;5(12):1031–1037. doi: 10.1038/nmeth.1266
[3] Messelink JJB, van Teeseling MCF, Janssen J, et al. Learning the distribution of single-cell chromosome conformations in bacteria reveals emergent order across genomic scales. Nat Commun. 2021;12(1):1963. doi: 10.1038/s41467-021-22189-x
[4] Misteli T. The self-organizing genome: principles of genome architecture and function. Cell. 2020;183(1):28–45. doi: 10.1016/j.cell.2020.09.014
[5] Jerkovic I, Cavalli G. Understanding 3D genome organization by multidisciplinary methods. Nat Rev Mol Cell Biol. 2021;22(8):511–528. doi: 10.1038/s41580-021-00362-w
[6] Garcia Fernandez F, Huet S, Mine-Hattab J. Multi-scale imaging of the dynamic organization of chromatin. Int J Mol Sci. 2023;24(21):24(21. doi: 10.3390/ijms242115975
[7] Ricci MA, Manzo C, Garcia-Parajo MF, et al. Chromatin fibers are formed by heterogeneous groups of nucleosomes in vivo. Cell. 2015;160(6):1145–1158. doi: 10.1016/j.cell.2015.01.054
[8] Nozaki T, Imai R, Tanbo M, et al. Dynamic organization of chromatin domains revealed by super-resolution live-cell imaging. Mol Cell. 2017;67(2):282–293 e7. doi: 10.1016/j.molcel.2017.06.018
[9] Xu J, Ma H, Jin J, et al. Super-resolution imaging of higher-order chromatin structures at different epigenomic states in single mammalian cells. Cell Rep. 2018;24(4):873–882. doi: 10.1016/j.celrep.2018.06.085
[10] Bancaud A, Huet S, Daigle N, et al. Molecular crowding affects diffusion and binding of nuclear proteins in heterochromatin and reveals the fractal organization of chromatin. Embo J. 2009;28(24):3785–3798. doi: 10.1038/emboj.2009.340
[11] Miron E, Oldenkamp R, Brown JM, et al. Chromatin arranges in chains of mesoscale domains with nanoscale functional topography independent of cohesin. Sci Adv. 2020;6(39):6(39. doi: 10.1126/sciadv.aba8811
[12] Barth R, Bystricky K, Shaban HA. Coupling chromatin structure and dynamics by live super-resolution imaging. Sci Adv. 2020;6(27):6(27. doi: 10.1126/sciadv.aaz2196
[13] Lieberman-Aiden E, van Berkum NL, Williams L, et al. Comprehensive mapping of long-range interactions reveals folding principles of the human genome.

Science. 2009;326(5950):289–293. doi: 10.1126/science.1181369

[14] Parteka-Tojek Z, Zhu JJ, Lee B, et al. Super-resolution visualization of chromatin loop folding in human lymphoblastoid cells using interferometric photoactivated localization microscopy. Sci Rep. 2022;12(1):8582. doi: 10.1038/s41598-022-12568-9

[15] Hertwig O. Lehrbuch der Entwicklungsgeschichte des Menschen und der Wirbeltiere. 5th ed. Jena, Gustav Fischer; 1896. doi: 10.5962/bhl.title.3980

[16] Gibcus JH, Samejima K, Goloborodko A, et al. A pathway for mitotic chromosome formation. Science. 2018;359(6376). doi: 10.1126/science.aao6135

[17] Bond ML, Davis ES, Quiroga IY, et al. Chromatin loop dynamics during cellular differentiation are associated with changes to both anchor and internal regulatory features. Genome Res. 2023;33(8):1258–1268. doi: 10.1101/gr.277397.122

[18] Rittenhouse NL, Dowen JM. Cohesin regulation and roles in chromosome structure and function. Curr Opin Genet Dev. 2024;85:102159. doi: 10.1016/j.gde.2024.102159

[19] Hult C, Adalsteinsson D, Vasquez PA, et al. Enrichment of dynamic chromosomal crosslinks drive phase separation of the nucleolus. Nucleic Acids Res. 2017;45(19):11159–11173. doi: 10.1093/nar/gkx741

[20] Walker B, Taylor D, Lawrimore J, et al. Transient crosslinking kinetics optimize gene cluster interactions. PLOS Comput Biol. 2019;15(8):e1007124. doi: 10.1371/journal.pcbi.1007124

[21] Sandoval-Velasco M, Dudchenko O, Rodríguez JA, et al. Three-dimensional genome architecture persists in a 52,000-year-old woolly mammoth skin sample. Cell. 2024;187(14):3541–3562 e51. doi: 10.1016/j.cell.2024.06.002

[22] Arnould C, Rocher V, Finoux AL, et al. Loop extrusion as a mechanism for formation of DNA damage repair foci. Nature. 2021;590(7847):660–665. doi: 10.1038/s41586-021-03193-z

[23] Fudenberg G, Imakaev M, Lu C, et al. Formation of chromosomal domains by loop extrusion. Cell Rep. 2016;15(9):2038–2049. doi: 10.1016/j.celrep.2016.04.085

[24] Ganji M, Shaltiel IA, Bisht S, et al. Real-time imaging of DNA loop extrusion by condensin. Science. 2018;360(6384):102–105. doi: 10.1126/science.aar7831

[25] Finn EH, Pegoraro G, Brandao HB, et al. Extensive heterogeneity and intrinsic variation in spatial genome organization. Cell. 2019;176(6):1502–1515 e10. doi: 10.1016/j.cell.2019.01.020

[26] Su JH, Zheng P, Kinrot SS, et al. Genome-scale imaging of the 3D organization and transcriptional activity of chromatin. Cell. 2020;182(6):1641–1659 e26. doi: 10.1016/j.cell.2020.07.032

[27] Szabo Q, Donjon A, Jerkovic I, et al. Regulation of single-cell genome organization into TADs and chromatin nanodomains. Nat Genet. 2020;52(11):1151–1157. doi: 10.1038/s41588-020-00716-8

[28] Sood V, Misteli T. The stochastic nature of genome organization and function. Curr Opin Genet Dev. 2022;72:45–52. doi: 10.1016/j.gde.2021.10.004

[29] Vasquez PA, Bloom K. Polymer models of interphase chromosomes. Nucleus. 2014;5(5):376–390. doi: 10.4161/nucl.36275

[30] Vasquez PA, Hult C, Adalsteinsson D, et al. Entropy gives rise to topologically associating domains. Nucleic Acids Res. 2016;44(12):5540–5549. doi: 10.1093/nar/gkw510

[31] Taddei A, Gasser SM. Structure and function in the budding yeast nucleus. Genetics. 2012;192(1):107–129. doi: 10.1534/genetics.112.140608

[32] Lawrimore CJ, Lawrimore J, He Y, et al. Polymer perspective of genome mobilization. Mutat Res. 2020;821:111706. doi: 10.1016/j.mrfmmm.2020.111706

[33] Pouokam M, Cruz B, Burgess S, et al. The Rabl configuration limits topological entanglement of chromosomes in budding yeast. Sci Rep. 2019;9(1):6795. doi: 10.1038/s41598-019-42967-4

[34] van Steensel B, Belmont AS. Lamina-associated domains: links with chromosome architecture, Heterochromatin, and gene repression. Cell. 2017;169(5):780–791. doi: 10.1016/j.cell.2017.04.022

[35] Simon DN, Wilson KL. The nucleoskeleton as a genome-associated dynamic 'network of networks'. Nat Rev Mol Cell Biol. 2011;12(11):695–708. doi: 10.1038/nrm3207

[36] Bronshtein I, Kepten E, Kanter I, et al. Loss of lamin a function increases chromatin dynamics in the nuclear interior. Nat Commun. 2015;6(1):8044. doi: 10.1038/ncomms9044

[37] Taheri F, Isbilir B, Muller G, et al. Random motion of chromatin is influenced by Lamin a interconnections. Biophys J. 2018;114(10):2465–2472. doi: 10.1016/j.bpj.2018.04.037

[38] Graziano S, Kreienkamp R, Coll-Bonfill N, et al. Causes and consequences of genomic instability in laminopathies: replication stress and interferon response. Nucleus. 2018;9(1):258–275. doi: 10.1080/19491034.2018.1454168

[39] Abad PC, Lewis J, Mian IS, et al. NuMA influences higher order chromatin organization in human mammary epithelium. Mol Biol Cell. 2007;18(2):348–361. doi: 10.1091/mbc.e06-06-0551

[40] Vidi PA, Liu J, Salles D, et al. NuMA promotes homologous recombination repair by regulating the accumulation of the ISWI ATPase SNF2h at DNA breaks. Nucleic Acids Res. 2014;42(10):6365–6379. doi: 10.1093/nar/gku296

[41] Salvador Moreno N, Liu J, Haas KM, et al. The nuclear structural protein NuMA is a negative regulator of 53BP1 in DNA double-strand break repair. Nucleic Acids Res. 2019;47(6):2703–2715. doi: 10.1093/nar/gkz138

[42] Ray S, Abugable AA, Parker J, et al. A mechanism for oxidative damage repair at gene regulatory elements. Nature. 2022;609(7929):1038–1047. doi: 10.1038/s41586-022-05217-8
[43] Locatelli M, Lawrimore J, Lin H, et al. DNA damage reduces heterogeneity and coherence of chromatin motions. Proc Natl Acad Sci USA. 2022;119(29):e2205166119. doi: 10.1073/pnas.2205166119
[44] Shaban HA, Seeber A. Monitoring the spatio-temporal organization and dynamics of the genome. Nucleic Acids Res. 2020;48(7):3423–3434. doi: 10.1093/nar/gkaa135
[45] Mine-Hattab J, Chiolo I. Complex chromatin motions for DNA repair. Front Genet. 2020;11:800. doi: 10.3389/fgene.2020.00800
[46] Zhang M, Seitz C, Chang G, et al. A guide for single-particle chromatin tracking in live cell nuclei. Cell Biol Int. 2022;46(5):683–700. doi: 10.1002/cbin.11762
[47] Krawczyk PM, Borovski T, Stap J, et al. Chromatin mobility is increased at sites of DNA double-strand breaks. J Cell Sci. 2012;125(Pt 9):2127–2133. doi: 10.1242/jcs.089847
[48] Lottersberger F, Karssemeijer RA, Dimitrova N, et al. 53BP1 and the LINC complex promote microtubule-dependent DSB mobility and DNA repair. Cell. 2015;163(4):880–893. doi: 10.1016/j.cell.2015.09.057
[49] Hihara S, Pack C-G, Kaizu K, et al. Local nucleosome dynamics facilitate chromatin accessibility in living mammalian cells. Cell Rep. 2012;2(6):1645–1656. doi: 10.1016/j.celrep.2012.11.008
[50] Nagashima R, Hibino K, Ashwin SS, et al. Single nucleosome imaging reveals loose genome chromatin networks via active RNA polymerase II. J Cell Biol. 2019;218(5):1511–1530. doi: 10.1083/jcb.201811090
[51] Kefer P, Iqbal F, Locatelli M, et al. Performance of deep learning restoration methods for the extraction of particle dynamics in noisy microscopy image sequences. Mol Biol Cell. 2021;32(9):903–914. doi: 10.1091/mbc.E20-11-0689
[52] Seeber A, Hauer MH, Gasser SM. Chromosome dynamics in response to DNA damage. Annu Rev Genet. 2018;52(1):295–319. doi: 10.1146/annurev-genet-120417-031334
[53] Robinett CC, Straight A, Li G, et al. In vivo localization of DNA sequences and visualization of large-scale chromatin organization using lac operator/repressor recognition. J Cell Biol. 1996;135(6):1685–1700. doi: 10.1083/jcb.135.6.1685
[54] Saad H, Gallardo F, Dalvai M, et al. DNA dynamics during early double-strand break processing revealed by non-intrusive imaging of living cells. PlOS Genet. 2014;10(3):e1004187. doi: 10.1371/journal.pgen.1004187
[55] Germier T, Audibert S, Kocanova S, et al. Real-time imaging of specific genomic loci in eukaryotic cells using the ANCHOR DNA labelling system. Methods. 2018;142:16–23. doi: 10.1016/j.ymeth.2018.04.008
[56] Chen B, Gilbert LA, Cimini BA, et al. Dynamic imaging of genomic loci in living human cells by an optimized CRISPR/Cas system. Cell. 2013;155(7):1479–1491. doi: 10.1016/j.cell.2013.12.001
[57] Ma H, Tu LC, Naseri A, et al. Multiplexed labeling of genomic loci with dCas9 and engineered sgRNAs using CRISPRainbow. Nat Biotechnol. 2016;34(5):528–530. doi: 10.1038/nbt.3526
[58] Zhang Z, Rong X, Xie T, et al. Fluorogenic CRISPR for genomic DNA imaging. Nat Commun. 2024;15(1):934. doi: 10.1038/s41467-024-45163-9
[59] Thuma J, Chung YC, Tu LC. Advances and challenges in crispr-based real-time imaging of dynamic genome organization. Front Mol Biosci. 2023;10:1173545. doi: 10.3389/fmolb.2023.1173545
[60] Nozaki T, Shinkai S, Ide S, et al. Condensed but liquid-like domain organization of active chromatin regions in living human cells. Sci Adv. 2023;9(14):eadf1488. doi: 10.1126/sciadv.adf1488
[61] Zidovska A, Weitz DA, Mitchison TJ. Micron-scale coherence in interphase chromatin dynamics. Proc Natl Acad Sci USA. 2013;110(39):15555–15560. doi: 10.1073/pnas.1220313110
[62] Shaban HA, Barth R, Recoules L, et al. Hi-d: nanoscale mapping of nuclear dynamics in single living cells. Genome Biol. 2020;21(1):95. doi: 10.1186/s13059-020-02002-6
[63] Valades-Cruz CA, Barth R, Abdellah M, et al. Genome-wide analysis of the biophysical properties of chromatin and nuclear proteins in living cells with hi-D. Nat Protoc. 2024. doi: 10.1038/s41596-024-01038-3
[64] Dantuma NP, van Attikum H. Spatiotemporal regulation of posttranslational modifications in the DNA damage response. Embo J. 2016;35(1):6–23. doi: 10.15252/embj.201592595
[65] Ciccia A, Elledge SJ. The DNA damage response: making it safe to play with knives. Mol Cell. 2010;40(2):179–204. doi: 10.1016/j.molcel.2010.09.019
[66] Scully R, Panday A, Elango R, et al. DNA double-strand break repair-pathway choice in somatic mammalian cells. Nat Rev Mol Cell Biol. 2019;20(11):698–714. doi: 10.1038/s41580-019-0152-0
[67] Polo SE, Almouzni G. Chromatin dynamics after DNA damage: the legacy of the access–repair–restore model. DNA Repair (Amst). 2015;36:114–121. doi: 10.1016/j.dnarep.2015.09.014
[68] Lawrimore J, Barry TM, Barry RM, et al. Microtubule dynamics drive enhanced chromatin motion and mobilize telomeres in response to DNA damage. Mol Biol Cell. 2017;28(12):1701–1711. doi: 10.1091/mbc.e16-12-0846
[69] Mine-Hattab J, Recamier V, Izeddin I, et al. Multi-scale tracking reveals scale-dependent chromatin dynamics after DNA damage. Mol Biol Cell. 2017;28(23):3323–3332. doi: 10.1091/mbc.e17-05-0317

[70] Bonin K, Smelser A, Moreno NS, et al. Structured illumination to spatially map chromatin motions. J Biomed Opt. 2018;23(5):1–8. doi: 10.1117/1.JBO.23.5.056007

[71] Mine-Hattab J, Rothstein R. DNA in motion during double-strand break repair. Trends Cell Biol. 2013;23(11):529–536. doi: 10.1016/j.tcb.2013.05.006

[72] Garcia Fernandez F, Fabre E. The dynamic behavior of chromatin in response to DNA double-strand breaks. Genes (Basel). 2022;13(2):13(2. doi: 10.3390/genes13020215

[73] Mine-Hattab J, Rothstein R. Increased chromosome mobility facilitates homology search during recombination. Nat Cell Biol. 2012;14(5):510–517. doi: 10.1038/ncb2472

[74] Neumann FR, Dion V, Gehlen LR, et al. Targeted INO80 enhances subnuclear chromatin movement and ectopic homologous recombination. Genes Dev. 2012;26(4):369–383. doi: 10.1101/gad.176156.111

[75] Seeber A, Dion V, Gasser SM. Checkpoint kinases and the INO80 nucleosome remodeling complex enhance global chromatin mobility in response to DNA damage. Genes Dev. 2013;27(18):1999–2008. doi: 10.1101/gad.222992.113

[76] Dion V, Kalck V, Seeber A, et al. Cohesin and the nucleolus constrain the mobility of spontaneous repair foci. EMBO Rep. 2013;14(11):984–991. doi: 10.1038/embor.2013.142

[77] Herbert S, Brion A, Arbona JM, et al. Chromatin stiffening underlies enhanced locus mobility after DNA damage in budding yeast. Embo J. 2017;36(17):2595–2608. doi: 10.15252/embj.201695842

[78] Smith MJ, Bryant EE, Rothstein R. Increased chromosomal mobility after DNA damage is controlled by interactions between the recombination machinery and the checkpoint. Genes Dev. 2018;32(17–18):1242–-1251. doi: 10.1101/gad.317966.118

[79] Smith MJ, Bryant EE, Joseph FJ, et al. DNA damage triggers increased mobility of chromosomes in G1-phase cells. Mol Biol Cell. 2019;30(21):2620–2625. doi: 10.1091/mbc.E19-08-0469

[80] Challa K, Schmid CD, Kitagawa S, et al. Damage-induced chromatome dynamics link ubiquitin ligase and proteasome recruitment to histone loss and efficient DNA repair. Mol Cell. 2021;81(4):811–829 e6. doi: 10.1016/j.molcel.2020.12.021

[81] Garcia Fernandez F, Almayrac E, Carre Simon A, et al. Global chromatin mobility induced by a DSB is dictated by chromosomal conformation and defines the HR outcome. Elife. 2022;11:11. doi: 10.7554/eLife.78015

[82] Liu S, Mine-Hattab J, Villemeur M, et al. In vivo tracking of functionally tagged Rad51 unveils a robust strategy of homology search. Nat Struct Mol Biol. 2023;30(10):1582–1591. doi: 10.1038/s41594-023-01065-w

[83] Oshidari R, Mekhail K, Seeber A. Mobility and repair of damaged DNA: random or directed? Trends Cell Biol. 2020;30(2):144–156. doi: 10.1016/j.tcb.2019.11.003

[84] Merigliano C, Chiolo I. Multi-scale dynamics of heterochromatin repair. Curr Opin Genet Dev. 2021;71:206–215. doi: 10.1016/j.gde.2021.09.007

[85] Penninckx S, Pariset E, Cekanaviciute E, et al. Quantification of radiation-induced DNA double strand break repair foci to evaluate and predict biological responses to ionizing radiation. NAR Cancer. 2021;3(4):zcab046. doi: 10.1093/narcan/zcab046

[86] Falk M, Hausmann M. A paradigm revolution or just better resolution—will newly emerging superresolution techniques identify chromatin architecture as a key factor in radiation-induced DNA damage and repair regulation? Cancers (Basel). 2020;13(1):18. doi: 10.3390/cancers13010018

[87] Dimitrova N, Chen YC, Spector DL, et al. 53BP1 promotes non-homologous end joining of telomeres by increasing chromatin mobility. Nature. 2008;456(7221):524–528. doi: 10.1038/nature07433

[88] Iida S, Shinkai S, Itoh Y, et al. Single-nucleosome imaging reveals steady-state motion of interphase chromatin in living human cells. Sci Adv. 2022;8(22):eabn5626. doi: 10.1126/sciadv.abn5626

[89] Pessina F, Giavazzi F, Yin Y, et al. Functional transcription promoters at DNA double-strand breaks mediate RNA-driven phase separation of damage-response factors. Nat Cell Biol. 2019;21(10):1286–1299. doi: 10.1038/s41556-019-0392-4

[90] Unal E, Arbel-Eden A, Sattler U, et al. DNA damage response pathway uses histone modification to assemble a double-strand break-specific cohesin domain. Mol Cell. 2004;16(6):991–1002. doi: 10.1016/j.molcel.2004.11.027

[91] Strom L, Lindroos HB, Shirahige K, et al. Postreplicative recruitment of cohesin to double-strand breaks is required for DNA repair. Mol Cell. 2004;16(6):1003–1015. doi: 10.1016/j.molcel.2004.11.026

[92] Phipps J, Toulouze M, Ducrot C, et al. Cohesin complex oligomerization maintains end-tethering at DNA double-strand breaks. Nature Cell Biology. 2024. doi: 10.1038/s41556-024-01552-2

[93] Liu J, Vidi PA, Lelievre SA, et al. Nanoscale histone localization in live cells reveals reduced chromatin mobility in response to DNA damage. J Cell Sci. 2015;128(3):599–604. doi: 10.1242/jcs.161885

[94] Arnould C, Rocher V, Saur F, et al. Chromatin compartmentalization regulates the response to DNA damage. Nature. 2023;623(7985):183–192. doi: 10.1038/s41586-023-06635-y

[95] Shi G, Liu L, Hyeon C, et al. Interphase human chromosome exhibits out of equilibrium glassy dynamics.

Nat Commun. 2018;9(1):3161. doi: 10.1038/s41467-018-05606-6

[96] Salari H, Di Stefano M, Jost D. Spatial organization of chromosomes leads to heterogeneous chromatin motion and drives the liquid- or gel-like dynamical behavior of chromatin. Genome Res. 2022;32(1):28–43. doi: 10.1101/gr.275827.121

[97] Shaban HA, Barth R, Bystricky K. Formation of correlated chromatin domains at nanoscale dynamic resolution during transcription. Nucleic Acids Res. 2018;46(13):e77. doi: 10.1093/nar/gky269

[98] Bonin K, Smelser A, Salvador-Moreno N, et al. Structured illumination to spatially map chromatin motions. J Biomed Opt. 2018;23(5):1–8. doi: 10.1117/1.JBO.23.5.056007

[99] Kolbin D, Walker BL, Hult C, et al. Polymer modeling reveals interplay between physical properties of chromosomal DNA and the size and distribution of condensin-based chromatin loops. Genes (Basel). 2023;14(12):2193. doi: 10.3390/genes14122193

[100] Kim E, Kerssemakers J, Shaltiel IA, et al. DNA-loop extruding condensin complexes can traverse one another. Nature. 2020;579(7799):438–442. doi: 10.1038/s41586-020-2067-5

[101] Terakawa T, Bisht S, Eeftens JM, et al. The condensin complex is a mechanochemical motor that translocates along DNA. Science. 2017;358(6363):672–676. doi: 10.1126/science.aan6516

[102] Davidson IF, Barth R, Zaczek M, et al. CTCF is a dna-tension-dependent barrier to cohesin-mediated loop extrusion. Nature. 2023;616(7958):822–827. doi: 10.1038/s41586-023-05961-5

[103] Nicklas RB. Chance encounters and precision in mitosis. J Cell Sci. 1988;89(Pt 3):283–285. doi: 10.1242/jcs.89.3.283

[104] Tlusty T, Bar-Ziv R, Libchaber A. High-fidelity DNA sensing by protein binding fluctuations. Phys Rev Lett. 2004;93(25):258103. doi: 10.1103/PhysRevLett.93.258103

[105] Murugan A, Huse DA, Leibler S. Speed, dissipation, and error in kinetic proofreading. Proc Natl Acad Sci USA. 2012;109(30):12034–12039. doi: 10.1073/pnas.1119911109

[106] Lawrimore J, Bloom K. Shaping centromeres to resist mitotic spindle forces. J Cell Sci. 2022;135(4). doi: 10.1242/jcs.259532

[107] Sacristan C, Samejima K, Ruiz LA, et al. Vertebrate centromeres in mitosis are functionally bipartite structures stabilized by cohesin. Cell. 2024;187(12):3006–3023 e26. doi: 10.1016/j.cell.2024.04.014

[108] Bloom K. Chromosome segregation: brushing up on centromeres. Curr Biol. 2024;34(12):R565–R567. doi: 10.1016/j.cub.2024.04.074

[109] Johzuka K, Horiuchi T. The cis element and factors required for condensin recruitment to chromosomes. Mol Cell. 2009;34(1):26–35. doi: 10.1016/j.molcel.2009.02.021

[110] Wu ZP, Bloom KS, Forest MG, et al. Transient crosslinking controls the condensate formation pathway within chromatin networks. Phys Rev E. 2024;109(4):L042401. doi: 10.1103/PhysRevE.109.L042401

[111] Ma H, Tu LC, Chung YC, et al. Cell cycle– and genomic distance–dependent dynamics of a discrete chromosomal region. J Cell Biol. 2019;218(5):1467–1477. doi: 10.1083/jcb.201807162

[112] Kokkanti AT, Atanasiu AT, Kolbin D, et al. TopoLoop: a new tool for chromatin loop detection in live cells via single-particle tracking. J Chem Phys. 2024;161(20):204105. doi: 10.1063/5.0236090

[113] Kruhlak MJ, Celeste A, Dellaire G, et al. Changes in chromatin structure and mobility in living cells at sites of DNA double-strand breaks. J Cell Biol. 2006;172(6):823–834. doi: 10.1083/jcb.200510015

[114] Pang B, Qiao X, Janssen L, et al. Drug-induced histone eviction from open chromatin contributes to the chemotherapeutic effects of doxorubicin. Nat Commun. 2013;4(1):1908. doi: 10.1038/ncomms2921