Chromatin-dependent regulation of nuclear size and mechanics

Wessel Rodenburg[1,2], Marian Barandica Graciani[1], Sven Ebben[1] & Jorine Eeftens[2,1]

[1]Institute for Molecules and Materials, Radboud University, Nijmegen, The Netherlands
[2]Radboud Institute for Molecular Life Sciences, Radboud University, Nijmegen, The Netherlands

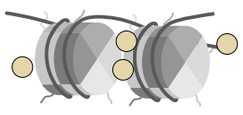
Radboud University

Connecting chromatin to mechanics

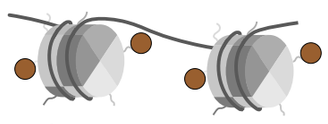
- Nuclear morphology and mechanics impact cell function.
- Chromatin is recognized as an important regulator, but the effect of chromatin composition is underexplored.

Goal: quantify how chromatin composition affects nuclear size and mechanics

Changing chromatin composition with small molecules:



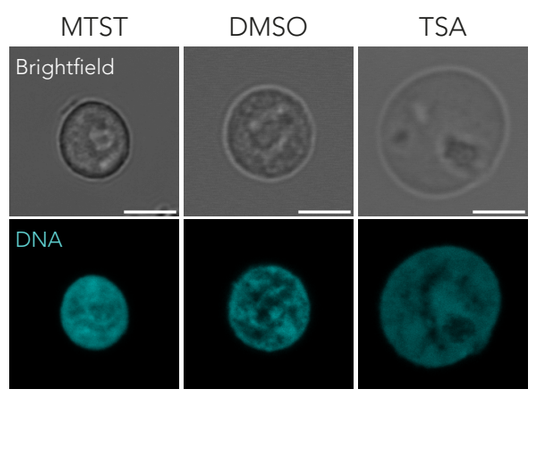
Methylstat (MTST): increases histone methylation

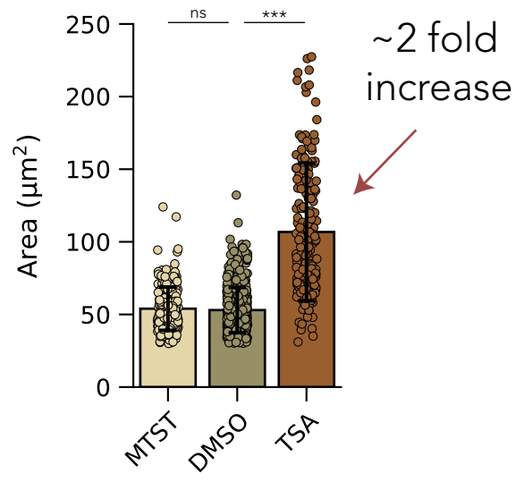


Trichostatin-A (TSA): increases histone acetylation

Histone modifications affect nuclear size

Quantifying size of isolated nuclei treated with MTST/TSA:

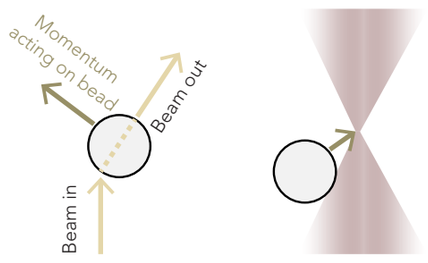


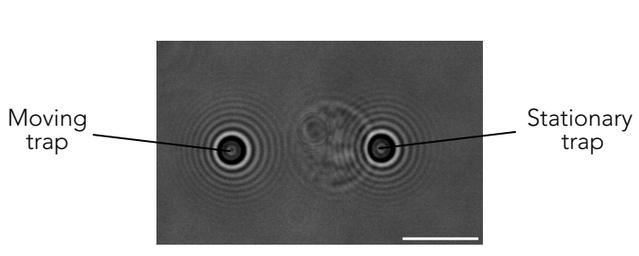


Chromatin composition changes the size of isolated nuclei

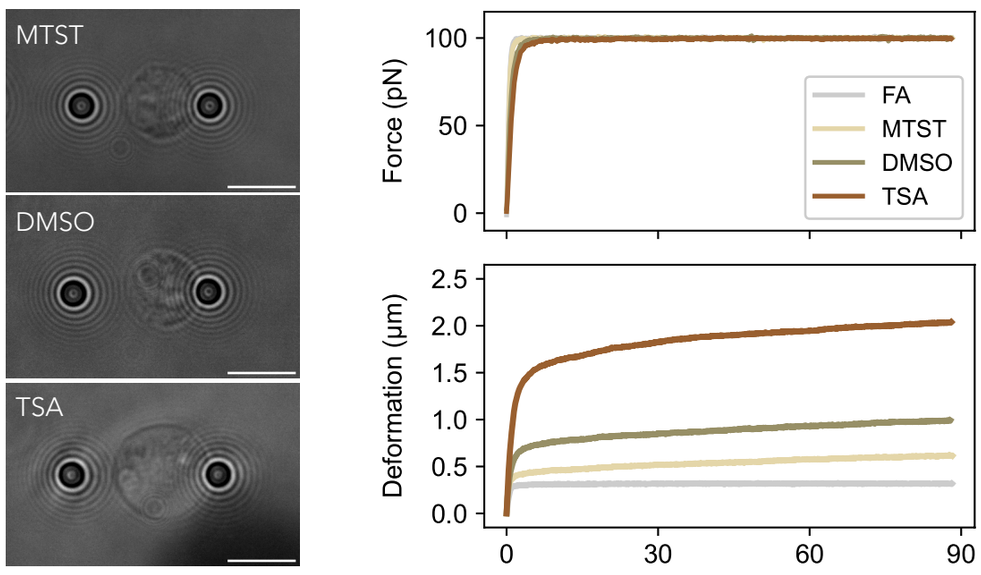
Histone modifications affect nuclear mechanics

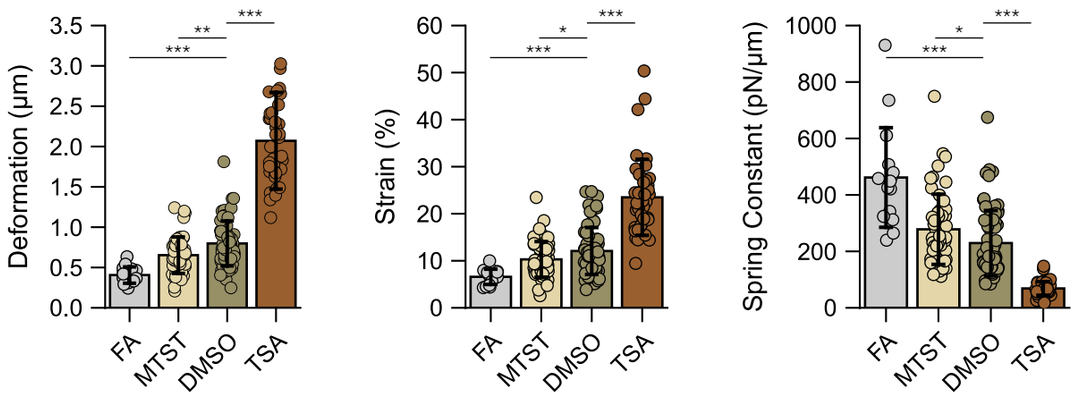
Measuring nuclear deformation with optical tweezers:
- Beads are trapped in a focused laserbeam
- Apply calibrated forces by displacing the laser





Quantifying chromatin effect on nuclear deformability:
- Apply 100 pN force and record deformation over time
- Compare nuclei treated with MTST, DMSO, TSA or crosslinking reagent (FA)





MTST:
- ↓ nuclear deformability
- ↑ stiffness

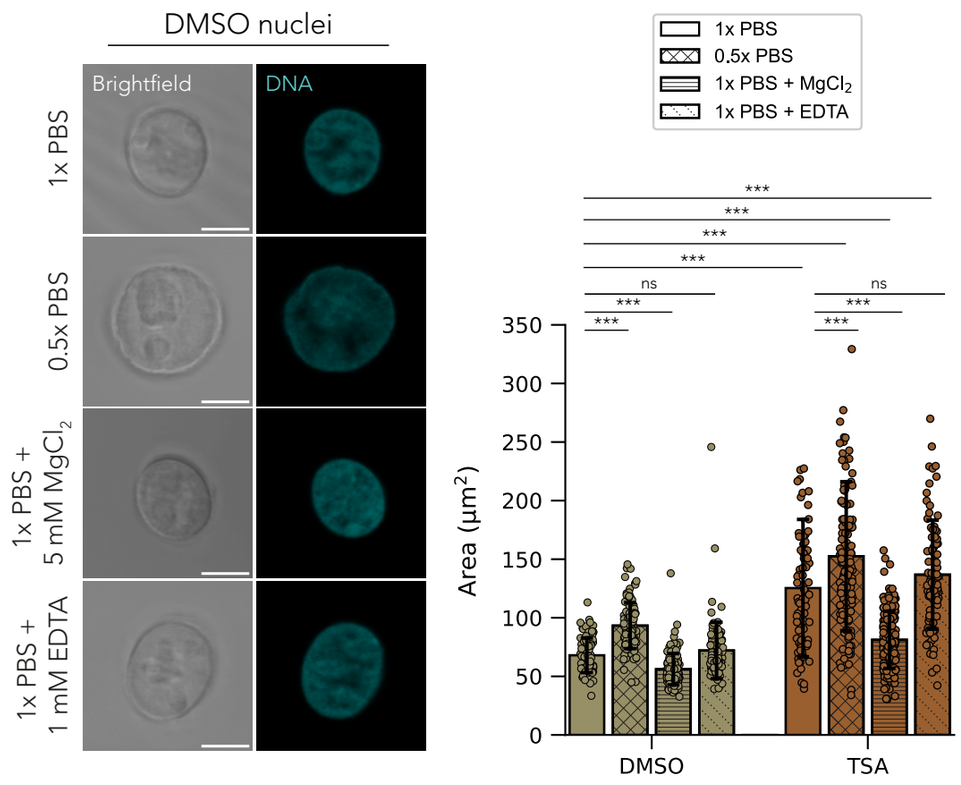
TSA:
- ↑ nuclear deformability
- ↓ stiffness

Quantifying chromatin effect on nuclear fluidity:
- Extract viscoelastic properties with creep compliance
- Exponent indicates fluidity (0=pure solid, 1=pure liquid)

All conditions have similar exponent: β = 0.2, consistent with predominantly solid behavior.

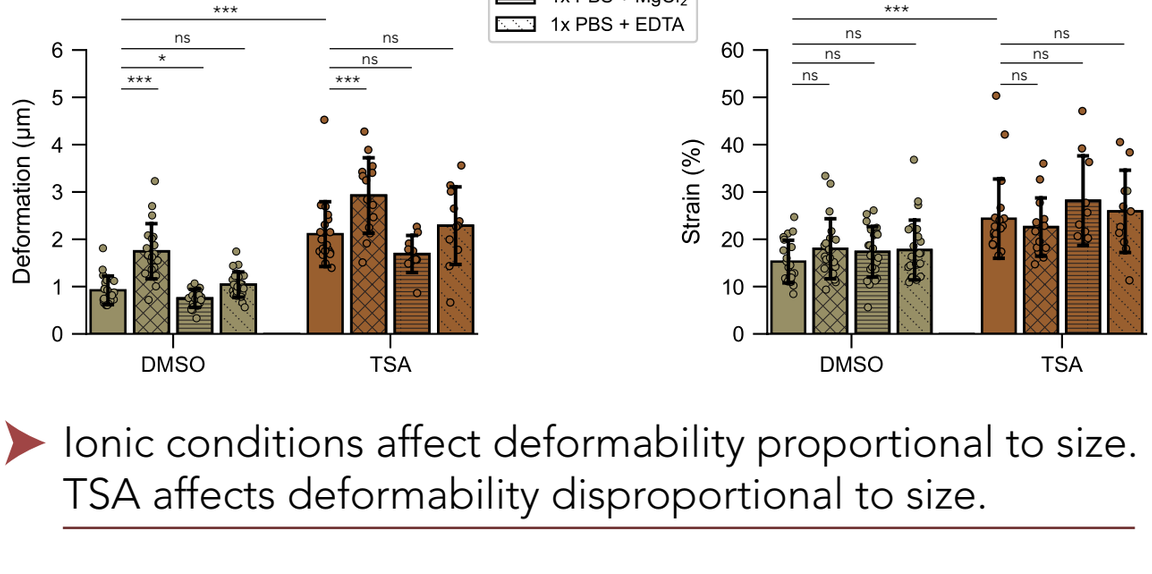
Chromatin and osmotics have differential effects on nuclear size and mechanics

Quantifying size of isolated nuclei in different buffers:
- Nuclear size also depends on ionic environment
- Quantified nuclear size of DMSO or TSA treated nuclei in different osmotic conditions



TSA increases size independent of osmotic condition

Quantifying the osmotics-chromatin balance in mechanics:
- Apply 100 pN force, record deformation over time
- DMSO and TSA nuclei in different osmotic conditions



Ionic conditions affect deformability proportional to size. TSA affects deformability disproportional to size.

Conclusions

- We can quantify the effect of chromatin composition on nuclear mechanics with optical tweezers.
- Osmotic condition changes mechanics proportional to size.
- Changes in histone modifications alter mechanics independent of nuclear size.
- **Chromatin composition intrinsically changes nuclear mechanics.**