



## Nuclear positioning as an integrator of cell fate

Maria Almonacid, Marie-Emilie Terret, Marie-Hélène Verlhac

### ► To cite this version:

Maria Almonacid, Marie-Emilie Terret, Marie-Hélène Verlhac. Nuclear positioning as an integrator of cell fate. *Current Opinion in Cell Biology*, 2019, 56, pp.122 - 129. <10.1016/j.ceb.2018.12.002>. <hal-03003451>

**HAL Id: hal-03003451**

**<https://hal.science/hal-03003451v1>**

Submitted on 25 Nov 2020

**HAL** is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



HAL Authorization



# Nuclear positioning as an integrator of cell fate

Maria Almonacid, Marie-Emilie Terret and Marie-Hélène Verlhac

Cells are the building units of living organisms and consequently adapt to their environment by modulating their intracellular architecture, in particular the position of their nucleus. Important efforts have been made to decipher the molecular mechanisms involved in nuclear positioning. The LINC complex at the nuclear envelope is a very important part of the molecular connectivity between the cell outside and the intranuclear compartment, and thus emerged as a central player in nuclear mechanotransduction. More recent concepts in nuclear mechanotransduction came from studies involving nuclear confined migration, compression or swelling. Also, the effect of nuclear mechanosensitive properties in driving cell differentiation raises the question of nuclear mechanotransduction and gene expression and recent efforts have been done to tackle it, even though it remains difficult to address in a direct manner. Eventually, an original mechanism of nucleus positioning, mechanotransduction and regulation of gene expression in the non-adherent, non-polarized mouse oocyte, highlights the fact that nuclear positioning is an important developmental issue.

## Address

CIRB, Collège de France, and CNRS-UMR7241 and INSERM-U1050, Equipe Labellisée FRM, Paris F-75005, France

Corresponding author:

Almonacid, Maria ([maria-elsa.almonacid@college-de-france.fr](mailto:maria-elsa.almonacid@college-de-france.fr))

**Current Opinion in Cell Biology** 2019, **56**:122–129

This review comes from a themed issue on **Cell architecture**

Edited by **Johanna Ivaska** and **Manuel Théry**

<https://doi.org/10.1016/j.ceb.2018.12.002>

0955-0674/© 2018 Elsevier Ltd. All rights reserved.

## Introduction

As the largest organelle in cells, the nucleus is often the organizing center of other subcellular structures and compartments. Neurons are tangible examples of this, with their nucleus location defining the neuronal cell body. In eukaryotic cells, the nucleus is positioned in an active manner, resulting in its location associated with different biological processes, such as the site of cell division, the differentiation status or the direction of migration [1,2]. Nuclear position in differentiated cells, such as neurons, myofibers, or epithelial cells, strongly

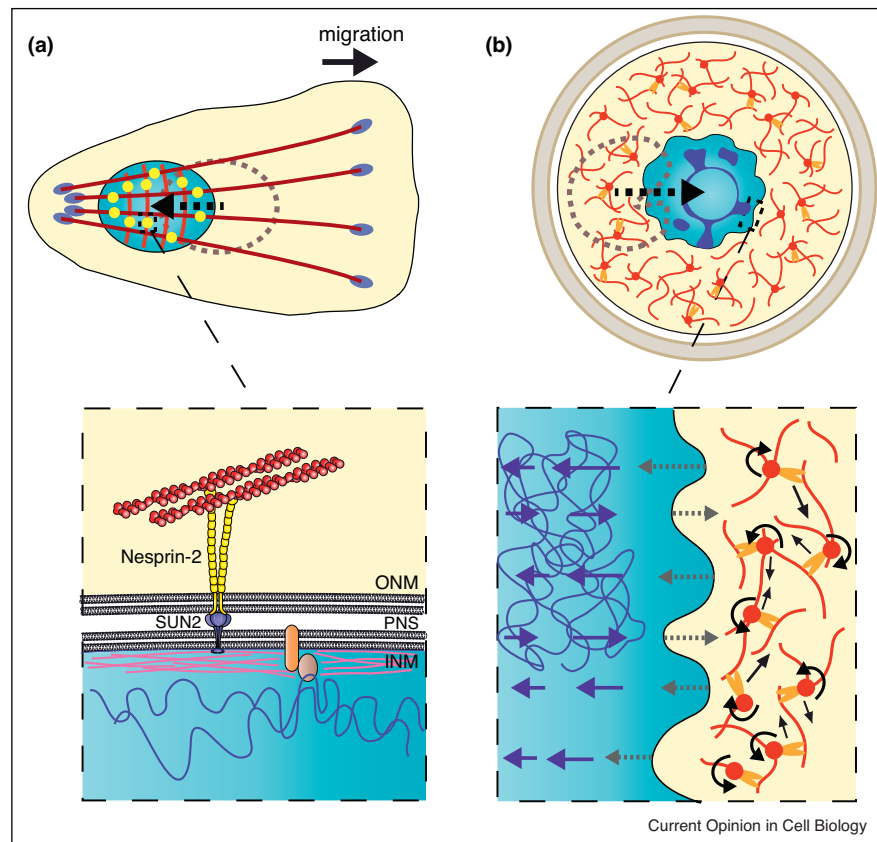
impacts their function, as its deregulation can result in cell dysfunction and disease, such as muscular disorders such as Emery-Dreifuss muscular dystrophy and dilated cardiomyopathy, and central nervous system disorders such as lissencephaly [3,4]. In this review, we will address the link between nuclear positioning and mechanotransduction in two very different models: polarized migrating cells and the non-polarized mouse oocyte. Indeed, a recent work has highlighted the existence of mechanosensitive mechanisms in the nucleus, revealing its responsiveness to mechanical forces, even if the molecular players involved need further investigation. We will also discuss the impact on gene expression, enabling us to raise potential developmental consequences.

## Molecular connectivity from outside the cell to the nucleus

Cells evolve in a dynamic environment where they are subjected to various mechanical influences, from their two-dimensional substrate in the case of monolayers *in vitro*, to the context of the surrounding tissue in an organism. Variations in the mechanical environment influence cell–cell as well as cell–extracellular matrix interactions and *in fine*, cell morphology and function. Thus, cells have to adapt to extracellular inputs and as a consequence, rearrange their intracellular components, including the nucleus, the largest organelle. Indeed, many studies suggest now that the nucleus is able to sense extracellular forces. The idea of a coupling between the extracellular environment and the nucleus was first proposed by the Ingber lab [5]. Such a coupling implies connectivity from the plasma membrane to the nucleus (for reviews, see Refs. [6,7]). Indeed, forces exerted on mechanosensitive receptors such as integrins or cadherins at the cell surface are relayed by cytoskeletal filaments in the cytoplasm to the nucleus [8].

Key players in this process are LINC complexes that make the connection between the cytoplasmic-skeleton and the nuclear interior. The role of LINC (LInker of the Nucleoskeleton and Cytoskeleton) complexes in nucleo-cytoplasmic coupling during nuclear positioning emerged from studies performed in *Caenorhabditis elegans* [9,10], but LINC complexes have been documented in all eukaryotic cells [2]. This force transmission system is associated with the nuclear envelope [2]. LINC complexes are composed of outer nuclear membrane (ONM) KASH domain (Klarsicht, ANC-1, Syne Homology) proteins and inner nuclear membrane (INM) SUN domain (Sad1, UNC-84) proteins (Figure 1a). SUN domain proteins are tethers for the KASH domain within the perinuclear space (PNS) between the inner and the outer

Figure 1



Nuclear mechanotransduction mechanisms during nuclear positioning in 2D migrating cells and mouse oocytes.

**(a): Up:** Nuclear positioning in a 2D migrating cell. The black arrow represents the direction of cell migration. The black dotted arrow corresponds to the displacement between initial (brown dotted outline) and final nucleus position. The nucleus is in blue and the cytoplasm in yellow. Two dorsal actin networks are represented: the perinuclear actin cap that flattens the nucleus, consisting of actin filament bundles (dark brown) connected to focal adhesions (purple pads), and TAN lines (red) involved in cell migration. Both are connected to the nucleus through the LINC complex (yellow dots).

**Zoomed inset:** Molecular connectivity at the nuclear envelope involved in nuclear mechanotransduction by TAN lines. Nesprin-2 interacts with actin filaments at the outer nuclear envelope. The KASH domain of Nesprin-2 interacts with the SUN domain protein SUN2 in the perinuclear space (PNS). SUN2 interacts with the nuclear lamina underlying the inner nuclear membrane (INM). At the INM, LEM domain proteins (LAP2, Emerin, MAN1) interact with the BAF protein that directly binds DNA. Red: actin filaments, yellow: Nesprin-2, purple: SUN2, pink: nuclear lamina, orange: LEM domain proteins, brown: BAF protein, dark blue: chromatin.

**(b): Up:** Nuclear positioning in the unpolarized mouse oocyte. The oocyte is surrounded by its protective shell, the zona pellucida (in grey). The black dotted arrow corresponds to the displacement between initial (brown dotted outline) and final nucleus position. The nucleus is in blue and the cytoplasm in yellow. The cytoplasm is filled with the cytoplasmic actin meshwork (actin filaments and actin-coated vesicles, in red). Myosin Vb, which drives the dynamics of the meshwork, is in orange. Inside the nucleus is represented the very peculiar ring-like structure of chromatin (dark blue) in fully grown mouse oocytes, with few contacts with the nuclear envelope. The empty hole corresponds to the space occupied by the nucleolus (not represented).

**Zoomed inset:** Nuclear mechanotransduction by active fluctuations at the nuclear envelope. In the cytoplasm, the dynamics of the cytoplasmic actin meshwork (black arrows) generates advection within the cytoplasm (black vortices). This global cytoplasmic activity generates fluctuations at the nuclear envelope (grey dotted arrows), which in turn promote global agitation in the nucleoplasm (blue arrows) and increase chromatin dynamics (dark blue: chromatin).

nuclear membranes (Figure 1a). The C-terminal KASH domain contains a trans-membrane segment followed by a short extension into the PNS that interacts with KASH binding sites on individual SUN protein trimers [11]. The cytoplasmic domains of KASH proteins can bind various cytoskeletal components. In SUN proteins, the C-terminal region includes the SUN domain that extends

into the PNS. The N-terminal domain is associated with the nuclear lamina or other nuclear components (Figure 1a) [2].

The last element of this trans-cellular network is the nuclear lamina, consisting of A-type and B-type lamins. These intermediate filaments constitute a scaffold

underlying the INM (Figure 1a) [12]. A-type lamins are the main determinants of NE mechanical properties [12,13], supporting the role of the final output in this mechanotransductive chain.

Altogether, the multiple players of this connective entity between plasma membrane and nucleus allow an effective coupling between the nucleus and the cell nearest environment.

### Mechanisms of nucleus positioning involving LINC complexes

The mechanisms of nucleus positioning have been studied in various cell types. In polarized cells, these mechanisms often depend on the connectivity between the nucleus and the cytoskeleton and thus on LINC complexes.

As nuclear positioning defects are associated to muscular and central nervous system disorders, nuclear positioning has been well investigated during myogenesis and neurogenesis. Myogenesis involves successive nuclear movements, mostly microtubules-dependent [14]. In mammalian neuro-epithelia, nuclear shuttling from a basal to an apical and from an apical to basal position, known as Interkinetic Nuclear Movement (IKNM), essential for the regulated proliferation of neuro-epithelial cells, is also microtubules-dependent [15].

Nuclear positioning has also been studied during cell migration (for a review, see Ref. [16]). The mechanisms documented thus far are mostly actin-dependent. Before migration, cells undergo changes in their architecture, that is, organelles and cytoskeleton rearrangements allow the cell to polarize in the direction of migration. In this regard, the position of the nucleus plays a central role. Lots of concepts on cell migration come from studies on 2D substrates. In most 2D cultured cells, nuclei localize near the cell centroid, but move to the cell rear upon initiation of migration, creating a leading edge/centrosome/nucleus axis in the direction of migration (Figure 1a) [17,18]. Such a nuclear translocation is achieved by coupling of the nuclear envelope to an actin retrograde flow through the LINC complex. This nuclear movement involves a structure called TAN (Transmembrane Actin-associated Nuclear) lines that are dorsal actin cables containing the KASH domain protein Nesprin-2, SUN2 and actin and that tether the nucleus to the actin cytoskeleton (Figure 1a, Table 1) [17,18]. During migration, the position of the nucleus is maintained by a Myosin II-dependent mechanism, both pushing and pulling the nucleus (Table 1) [19].

The use of 3D substrates highlighted novel aspects of nuclear positioning during cell migration. During 3D cell migration, cells often position their nuclei at the rear, and the nucleus becomes an obstacle to penetrate through narrow

space as it is the case during metastasis. Intrinsic mechanical properties of the nuclei become then very important [20<sup>••</sup>,21<sup>•</sup>,22<sup>•</sup>]. Nuclear translocation through tiny space is Myosin II-dependent, as shown for breast cancer cells (Table 1) [23]. Alternatively, immune cells can insert their nuclei in the leading edge to pass through endothelial layers during Transendothelial Migration (TEM). In those trans-migrating cells, Myosin II contractility is involved in nuclear squeezing across the endothelial barrier [24].

### Nuclear positioning and mechanotransduction

Nuclear positioning mechanisms based on the molecular connectivity from the plasma membrane to the nucleus involve mechanotransduction (for reviews, see Refs. [25,26]). Thanks to this molecular coupling, the nucleus is wired to mechanical changes outside the cell such as extracellular matrix adhesiveness or mechanics. As an illustration, an intact cytoskeleton is required to regulate nuclear morphology [27–30] and expression of dominant negative forms of the LINC complex components SUN or KASH impair force transmission from the cytoskeleton to the nucleus and impair nuclear deformation [31].

However, recent findings suggest that the LINC complex is not always required for mechanotransduction to the nucleus. Whereas in adherent cells, a commonly observed perinuclear actin cap flattening the nucleus is LINC complex-dependent [27], Thiam *et al.* reported a perinuclear actin network in dendritic cells that facilitates nuclear migration through narrow constrictions by disrupting Lamin A/C and thus favoring nuclear deformability. This mechanism does not rely on the LINC complex (Table 1) [20<sup>••</sup>]. Remarkably, nuclear translocation through tiny spaces challenges nuclear integrity. It induces transient nuclear envelope rupture, rapidly repaired by the ESCRT III machinery to avoid further DNA damage [21<sup>•</sup>,22<sup>•</sup>]. This is an example of mechanotransduction impacting nuclear content and genome integrity and whether this could affect other cellular responses such as transcription would be an interesting matter of investigation.

On stiff substrates, focal adhesions and stress fibers apply forces to the nucleus and flatten it. This increases nuclear membrane curvature, stretching nuclear pores and leading to increased import of the transcriptional regulator YAP. Recently, Elosegui-Artola *et al.* found that direct application of a force on the nucleus through AFM (Atomic Force Microscopy) mimicked this effect in a LINC-independent manner [32<sup>••</sup>].

The nucleus itself has mechanosensitive properties that relay the mechanotransduction from outside of the cell and from the cytoplasm. This was experimentally demonstrated by magnetic tweezers experiments on isolated nuclei: application of a repeated force triggered nuclear stiffening, showing that the nucleus itself can produce a

**Table 1****Features of nuclear mechanotransduction mechanisms during nuclear positioning in polarized migrating cells and mouse oocytes**

	Polarized migrating cell (2D)	Polarized migrating cell (3D)	Mouse oocyte
Cytoplasmic-skeleton	TAN lines (actin)	Perinuclear actin network	Cytoplasmic actin meshwork (actin filaments and actin-coated vesicles)
Molecular motor	Myosin II	Myosin II	Myosin Vb
Driving force	Coupling of the nuclear envelope to an actin retrograde flow	Nuclear deformation: perinuclear actin nucleation induces nuclear lamina rupture	Advection induced by the cytoplasmic actin meshwork generates nuclear envelope fluctuations
Nuclear envelope connectivity	LINC complex (Nesprin-2 and SUN2)	LINC independent	Not reported
Effects on gene expression	Contacts chromatin/lamina	Not reported	Nucleoplasmic advection

mechanical response to applied forces [33]. The lamina and chromatin are the major determinants of nuclear mechanics. Lamin A is a key element regulating nuclear shape and rigidity [34,35] and depending on its state, chromatin can either stiffen or soften nuclei. Heterochromatin is more densely packed and makes nuclei stiffer, whereas euchromatin contributes to the viscous part of nuclear mechanics [36,37]. This is particularly striking during cellular differentiation, where both Lamin A accumulation underneath the INM and formation of peripheral heterochromatin increase stiffness at the periphery [36,37]. Another important parameter in nuclear mechanics is nuclear membrane curvature and tension. Trying to elucidate the inflammatory response to wounding in zebrafish, a recent study discovered that in HeLa cells, pro-inflammatory phospholipase A2 (PLA2) translocates from the nucleoplasm to the INM when membrane tension increases following nuclear swelling [38<sup>••</sup>]. This mechanotransduction mechanism triggers a chemoattractive inflammatory signal that recruits leucocytes. Interestingly, the actin cap [27] plays here a protective role against nuclear swelling. This mechanism could also have potential implications in other situations putting nuclei at stress such as Transendothelial Migration or migration through narrow spaces.

A comprehensive study from the Discher lab reported the importance of nuclear mechanosensitive properties in driving cell differentiation [34]. Artificial changes in matrix stiffness modulate Lamin A levels from tissue culture cells, with high stiffness indirectly raising nuclear rigidity and regulating stress fiber genes as well as nuclear localization of transcriptional regulators such as YAP. Altogether, extracellular matrix stiffness regulation tunes the commitment of undifferentiated cells toward either bone-like or fat-like fates. Studying the effects of mechanotransduction on gene expression is thus the subject of extensive investigation.

### Mechanotransduction and gene expression

The continuum of force transmission existing in polarized cells, from outside the cell up to the intra-nuclear

compartment, through the connection between the cytoplasmic-skeleton and the nuclear envelope, is an intuitive way to regulate gene expression in response to extracellular inputs. This is a meaningful point, since chromatin is one of the last players of this pathway through its association to the nuclear envelope by the Barrier to Autointegration Factor (BAF) protein (Figure 1a) [39,40]. Indeed, chromatin organization inside the nucleus can be modified by external perturbation, like stretching the cell with glass pipettes or load compression [41]. Modulating chromatin organization is a way to regulate gene expression. The spatial location of the DNA within the nucleus is nonrandom. This organization constitutes the '4D nucleome' (the 3D chromatin architecture and its change over time) and impacts transcription and cellular functions [42–46]. In differentiated somatic cells, condensed and transcriptionally inactive heterochromatin is preferentially located at the nuclear periphery, close to the nuclear lamina, whereas de-condensed and transcriptionally active euchromatin is found away from the nuclear envelope [47,48]. Experimentally tethering genetically modified loci to the nuclear periphery can result in their repression, supporting the idea that Lamin A at the nuclear periphery represses transcription (Table 1) [49,50].

Several recent studies address more or less directly the link between mechanotransduction and gene expression. The Shivashankar lab established correlations between LINC complex connections, nuclear architecture, chromatin organization and gene expression in *Drosophila* cells [51]. They also established correlations between cell geometry, 3D organization of chromosome territories (the physical space occupied by individual chromosomes) inside the nucleus and gene expression in micro-patterned cells [52]. Another work demonstrated that mechanical strain applied on epidermal multipotent stem cells led to the enrichment of the nuclear membrane protein Emerin at the ONM and its decrease at the INM, inducing heterochromatin rearrangements and decrease of nuclear G-actin, altogether resulting in the downregulation of lineage commitment genes [53]. A more direct evidence of external force on transcription was provided by a single-cell study,



where application of a local shear stress by magnetic beads at the cell surface via integrins induced upregulation of transcription of a multi-copy insertion of a Bacterial Artificial Chromosome (BAC) [54\*\*].

### The original case of nucleus positioning inside a non-polarized cell, the mouse oocyte

Mouse oocytes grow within the ovary until reaching the big size of 80  $\mu\text{m}$  diameter. By the end of their growth, the nucleus moves from a peripheral to a central position [55]. Unlike somatic cells, mouse oocytes lack the key players allowing them to adapt their geometry relative to their environment [56]: they neither have centrosomes nor associated microtubules [57], and they also lack cortical and intracellular polarity [58–60]. However, fully grown mouse oocytes display a stereotypical spatial organization, with their big nucleus (30  $\mu\text{m}$  diameter, the size of a somatic cell) at the center, suggesting that these cells have their own mechanisms to define their center. Consequently, they present an original mode of nuclear positioning, not relying on microtubules and centrosomes, or on Myosin II induced contractility.

Instead, nuclear positioning is driven by the MyosinVb-dependent dynamics of actin-coated vesicles (Table 1). More precisely, the activity of those actin vesicles increases close to the cortex, generating a pressure gradient and a propulsion force sufficient to move the oocyte nucleus (Figure 1b). Actin vesicles dynamics also move the whole cytoplasm (fluids and organelles) by advection (Table 1), thus promoting its fluidization, favoring nucleus directional movement toward the center (Figure 1b) [61]. This mechanism is robust which could be related to the physical nature of the gradient. Indeed, even fluorescent latex beads aggregates, when injected to the periphery of the cell can be pushed toward the center [61]. Therefore, we suspect that any object placed within the gradient would find its way to the center. The size of the object may be a critical parameter and further work will help fully characterize this gradient of pressure and its properties. The non-specific nature of the gradient also suggests that nuclear centering mechanism in mouse oocytes is probably LINC complex independent.

As a sequel of this work, a preprint article reports a potential role for nuclear centering in mouse oocytes [62]. During the process of nuclear positioning, cytoplasmic activity generated by the dynamics of actin vesicles, promotes nuclear envelope fluctuations and chromatin motion, thereby modulating gene expression (Figure 1b, Table 1). This mechanotransduction effect is developmentally important, since transcriptome composition in mammalian fully grown oocytes sustains further embryo development. Indeed transcription is strongly decreased at the end of oocyte growth [62] and during meiotic divisions to resume after fertilization at the two-cell embryo stage only. Hence, mRNA maternal stores

support these essential steps in the early life of a new organism. This unprecedented model for mechanotransduction, where the activity from the cytoplasm alone is able to generate fluctuations at the nuclear envelope and promotes global agitation inside the nucleus, is fully consistent in this mechanically isolated cell. This is also reminiscent of a study from the Lecuit lab, where during early *Drosophila* embryogenesis, microtubule forces are transmitted to the nucleoplasm and enhance chromatin dynamics. The global agitation inside the nucleus could contribute to the induction of zygotic gene expression [37,63].

Such a novel mode of mechanotransduction in mouse oocytes results from the non-conventional mechanism of nuclear positioning. The mouse oocyte by its very high cortex tension at that stage [64,65] as well as the presence of its protective shell, the zona pellucida, is somehow rather isolated in terms of forces from the outside, thus all the connectivity from the cell outside up to the nuclear compartment, as described in polarized cells, is not relevant in that case. Furthermore, as discussed above, we suspect nuclear positioning in mouse oocytes to be LINC complex independent. Interestingly, LINC-independent mechanisms for mechanotransduction have also been reported in polarized cells, such as dendritic cells migrating through narrow constrictions [20\*\*]. Also, nuclear architecture in fully grown, developmentally competent mouse oocytes is very different from the one in somatic cells. The nucleus itself is the size of a somatic cell and contacts between lamina and chromatin are reduced at this stage. Instead, at the end of oocyte growth, chromatin wraps around the big nucleolus as a ring-like structure (Figure 1b) [66]. Thus, unlike in somatic cells, contacts with lamina and DNA/chromatin may not be essential for gene expression regulation by mechanotransduction.

### Conclusion and perspectives

Nucleus positioning is involved in many biological functions and nuclear positioning defects can lead to dysfunction and disease [1,2]. Understanding the role of nuclear positioning is also an important developmental issue. Noteworthy, the first nuclear movement in the life of an organism is the migration of the female and male pronuclei toward each other, often at zygote center, to allow the fusion of parental genomes. Also, in many organisms, nuclear positioning in the oocyte predefines the future axes of the embryo and in mouse oocytes, unpublished data suggest it regulates the composition of the transcriptome [62]. During development, nuclear positioning could impact not only the geometry of cell divisions but also cell fate. Nuclear positioning could affect cell fate determination through associated mechanotransduction and gene expression regulation. However, studying the link between mechanotransduction and gene expression *in vivo* is a hard task. Integrative approaches are strongly needed for such projects. There

is a real conceptual challenge to connect two different fields, cytoskeleton/mechanobiology and nuclear organization/gene regulation. For cell biologists, it implies to get familiar with complex approaches as Chromosome Conformation Capture to analyze genome organization or DNA adenine methyltransferase identification (DamID) to analyze genome-wide contacts with lamina. It also requires to circumvent several technical challenges such as direct measurements of nuclear forces and gene expression visualization in live cells. Recent developments in the labeling of specific endogenous genomic loci or transcripts using CRISPR–Cas9 and related systems in live will help overcome these critical issues [67,68]. All these efforts will contribute to better understand the impact of nuclear positioning on essential developmental processes.

### Conflict of interest statement

Nothing declared.

### Acknowledgements

The authors would like to apologize to all authors whose work could not be cited due to space constraints. This work was supported by grants from the Fondation pour la Recherche Médicale (FRM Label to MHV-DEQ20150331758), from the ANR (ANR-14-CE11 to MHV, ANR-16-CE13 to MET) and from the Labex Memolife (to MHV). This work has received support under the program «Investissements d'Avenir» launched by the French Government and implemented by the ANR, with the references: ANR-10-LABX-54 MEMO LIFE, ANR-11-IDEX-0001-02 PSL\* Research University.

### References and recommended reading

Papers of particular interest, published within the period of review, have been highlighted as:

- of special interest
- of outstanding interest

1. Bone CR, Starr DA: **Nuclear migration events throughout development.** *J Cell Sci* 2016, **129**:1951-1961.
2. Gundersen GG, Worman HJ: **Nuclear positioning.** *Cell* 2013, **152**:1376-1389.
3. Zhou C, Rao L, Warren DT, Shanahan CM, Zhang Q: **Mouse models of nesprin-related diseases.** *Biochem Soc Trans* 2018, **46**:669-681.
4. Banerjee I, Zhang J, Moore-Morris T, Pfeiffer E, Buchholz KS, Liu A, Ouyang K, Stroud MJ, Gerace L, Evans SM *et al.*: **Targeted ablation of nesprin 1 and nesprin 2 from murine myocardium results in cardiomyopathy, altered nuclear morphology and inhibition of the biomechanical gene response.** *PLoS Genet* 2014, **10**:e1004114.
5. Maniotis AJ, Chen CS, Ingber DE: **Demonstration of mechanical connections between integrins, cytoskeletal filaments, and nucleoplasm that stabilize nuclear structure.** *Proc Natl Acad Sci U S A* 1997, **94**:849-854.
6. Aureille J, Belaadi N, Guilluy C: **Mechanotransduction via the nuclear envelope: a distant reflection of the cell surface.** *Curr Opin Cell Biol* 2017, **44**:59-67.
7. Lee YL, Burke B: **LINC complexes and nuclear positioning.** *Semin Cell Dev Biol* 2018, **82**:67-76.
8. Wang N, Tytell JD, Ingber DE: **Mechanotransduction at a distance: mechanically coupling the extracellular matrix with the nucleus.** *Nat Rev Mol Cell Biol* 2009, **10**:75-82.
9. Starr DA, Han M: **Role of ANC-1 in tethering nuclei to the actin cytoskeleton.** *Science* 2002, **298**:406-409.
10. Lee KK, Starr D, Cohen M, Liu J, Han M, Wilson KL, Gruenbaum Y: **Lamin-dependent localization of UNC-84, a protein required for nuclear migration in *Caenorhabditis elegans*.** *Mol Biol Cell* 2002, **13**:892-901.
11. Sosa BA, Rothballer A, Kutay U, Schwartz TU: **LINC complexes form by binding of three KASH peptides to domain interfaces of trimeric SUN proteins.** *Cell* 2012, **149**:1035-1047.
12. Osmanagic-Myers S, Dechat T, Foisner R: **Lamins at the crossroads of mechanosignaling.** *Genes Dev* 2015, **29**:225-237.
13. Dahl KN, Kahn SM, Wilson KL, Discher DE: **The nuclear envelope lamina network has elasticity and a compressibility limit suggestive of a molecular shock absorber.** *J Cell Sci* 2004, **117**:4779-4786.
14. Roman W, Gomes ER: **Nuclear positioning in skeletal muscle.** *Semin Cell Dev Biol* 2018, **82**:51-56.
15. Hu DJ-K, Baffet AD, Nayak T, Akhmanova A, Doye V, Vallee RB: **Dynein recruitment to nuclear pores activates apical nuclear migration and mitotic entry in brain progenitor cells.** *Cell* 2013, **154**:1300-1313.
16. Calero-Cuenca FJ, Janota CS, Gomes ER: **Dealing with the nucleus during cell migration.** *Curr Opin Cell Biol* 2018, **50**:35-41.
17. Gomes ER, Jani S, Gundersen GG: **Nuclear movement regulated by Cdc42, MRCK, myosin, and actin flow establishes MTOC polarization in migrating cells.** *Cell* 2005, **121**:451-463.
18. Luxton GWG, Gomes ER, Folker ES, Vintinner E, Gundersen GG: **Linear arrays of nuclear envelope proteins harness retrograde actin flow for nuclear movement.** *Science* 2010, **329**:956-959.
19. Wu J, Kent IA, Shekhar N, Chancellor TJ, Mendonca A, Dickinson RB, Lele TP: **Actomyosin pulls to advance the nucleus in a migrating tissue cell.** *Biophys J* 2014, **106**:7-15.
20. Thiam H-R, Vargas P, Carpi N, Crespo CL, Raab M, Terriac E, King MC, Jacobelli J, Alberts AS, Stradal T *et al.*: **Perinuclear Arp2/3-driven actin polymerization enables nuclear deformation to facilitate cell migration through complex environments.** *Nat Commun* 2016, **7**:10997.
- This work shows that 3D migration of dendritic cells through narrow constrictions induces the rapid assembly of an Arp2/3 nucleated actin cap around the nucleus and transient lamina rupture, that facilitates nuclear squeezing through constrictions. This mechanism could explain how, inside organisms, migrating cells can cross dense tissues to achieve their function.
21. Denais CM, Gilbert RM, Isermann P, McGregor AL, te Lindert M, Weigelin B, Davidson PM, Friedl P, Wolf K, Lammerding J: **Nuclear envelope rupture and repair during cancer cell migration.** *Science* 2016, **352**:353-358.
- In this study, 3D migration of tumour cells through confining microenvironments affects nuclear integrity by generating transient nuclear envelope ruptures and DNA damage, which is restored by the ESCRT III machinery. This paper is an example of mechanical stress affecting nuclear content.
22. Raab M, Gentili M, de Belly H, Thiam HR, Vargas P, Jimenez AJ, Lautenschlaeger F, Voituriez R, Lennou-Duménil AM, Manel N *et al.*: **ESCRT III repairs nuclear envelope ruptures during cell migration to limit DNA damage and cell death.** *Science* 2016, **352**:359-362.
- Similarly to Denais *et al.* the authors show that 3D migration of dendritic cells through narrow constrictions induces transient nuclear envelope ruptures that are promptly repaired by the ESCRTIII machinery to limit DNA damage.
23. Thomas DG, Yenepalli A, Denais CM, Rape A, Beach JR, Wang Y-L, Schiemann WP, Baskaran H, Lammerding J, Egelhoff TT: **Non-muscle myosin IIB is critical for nuclear translocation during 3D invasion.** *J Cell Biol* 2015, **210**:583-594.
24. Barzilai S, Yadav SK, Morrell S, Roncato F, Klein E, Stoler-Barak L, Golani O, Feigelson SW, Zemel A, Nourshargh S *et al.*: **Leukocytes breach endothelial barriers by insertion of nuclear lobes and disassembly of endothelial actin filaments.** *Cell Rep* 2017, **18**:685-699.
25. Cho S, Irianto J, Discher DE: **Mechanosensing by the nucleus: from pathways to scaling relationships.** *J Cell Biol* 2017, **216**:305-315.

26. Kirby TJ, Lammerding J: **Emerging views of the nucleus as a cellular mechanosensor.** *Nat Cell Biol* 2018, **20**:373-381.
  27. Khatau SB, Hale CM, Stewart-Hutchinson PJ, Patel MS, Stewart CL, Searson PC, Hodzic D, Wirtz D: **A perinuclear actin cap regulates nuclear shape.** *Proc Natl Acad Sci U S A* 2009, **106**:19017-19022.
  28. Hatch EM, Hetzer MW: **Nuclear envelope rupture is induced by actin-based nucleus confinement.** *J Cell Biol* 2016, **215**:27-36.
  29. Makhija E, Jokhun DS, Shivashankar GV: **Nuclear deformability and telomere dynamics are regulated by cell geometric constraints.** *Proc Natl Acad Sci U S A* 2016, **113**:E32-E40.
  30. Keeling MC, Flores LR, Dodhy AH, Murray ER, Gavara N: **Actomyosin and vimentin cytoskeletal networks regulate nuclear shape, mechanics and chromatin organization.** *Sci Rep* 2017, **7**:5219.
  31. Lombardi ML, Jaalouk DE, Shanahan CM, Burke B, Roux KJ, Lammerding J: **The interaction between nesprins and sun proteins at the nuclear envelope is critical for force transmission between the nucleus and cytoskeleton.** *J Biol Chem* 2011, **286**:26743-26753.
  32. Elosegui-Artola A, Andreu I, Beedle AEM, Lezamiz A, Uroz M, Kosmalska AJ, Oria R, Kechagia JZ, Rico-Lastres P, Le Roux A-L **et al.**: **Force triggers YAP nuclear entry by regulating transport across nuclear pores.** *Cell* 2017, **171**:1397-1410.e14.
- In this work Elosegui-Artola *et al.* mimicked the effects of focal adhesion and stress fibers on the nucleus. They showed that direct application of a force on the nucleus flattens it, stretches the nuclear pores and leads to increased import of the transcriptional regulator YAP.
33. Guilluy C, Osborne LD, Van Landeghem L, Sharek L, Superfine R, Garcia-Mata R, Burridge K: **Isolated nuclei adapt to force and reveal a mechanotransduction pathway in the nucleus.** *Nat Cell Biol* 2014, **16**:376-381.
  34. Swift J, Ivanovska IL, Buxboim A, Harada T, Dingal PCDP, Pinter J, Pajeroski JD, Spinler KR, Shin J-W, Tewari M **et al.**: **Nuclear lamin-A scales with tissue stiffness and enhances matrix-directed differentiation.** *Science* 2013, **341**:1240104.
  35. de Leeuw R, Gruenbaum Y, Medalia O: **Nuclear lamins: thin filaments with major functions.** *Trends Cell Biol* 2018, **28**:34-45.
  36. Pajeroski JD, Dahl KN, Zhong FL, Sammak PJ, Discher DE: **Physical plasticity of the nucleus in stem cell differentiation.** *Proc Natl Acad Sci U S A* 2007, **104**:15619-15624.
  37. Hampoelz B, Lecuit T: **Nuclear mechanics in differentiation and development.** *Curr Opin Cell Biol* 2011, **23**:668-675.
  38. Enyedi B, Jelcic M, Niethammer P: **The cell nucleus serves as a mechanotransducer of tissue damage-induced inflammation.** *Cell* 2016, **165**:1160-1170.
- The authors show how in HeLa cells, nuclear swelling, by increasing nuclear membrane tension, induces recruitment of pro-inflammatory phospholipase A2 (PLA2) at the inner nuclear membrane. This mechanosensitive cascade is part of the inflammatory response to wounding in the context of a whole organism like zebrafish.
39. Furukawa K, Sugiyama S, Osouda S, Goto H, Inagaki M, Horigome T, Omata S, McConnell M, Fisher PA, Nishida Y: **Barrier-to-autointegration factor plays crucial roles in cell cycle progression and nuclear organization in Drosophila.** *J Cell Sci* 2003, **116**:3811-3823.
  40. Wilson KL, Foisner R: **Lamin-binding proteins.** *Cold Spring Harb Perspect Biol* 2010, **2**:a000554.
  41. Kumar A, Mazzanti M, Mistrik M, Kosar M, Beznoussenko GV, Mironov AA, Garré M, Parazzoli D, Shivashankar GV, Scita G **et al.**: **ATR mediates a checkpoint at the nuclear envelope in response to mechanical stress.** *Cell* 2014, **158**:633-646.
  42. Geyer PK, Vitalini MW, Wallrath LL: **Nuclear organization: taking a position on gene expression.** *Curr Opin Cell Biol* 2011, **23**:354-359.
  43. Chen H, Chen J, Muir LA, Ronquist S, Meixner W, Ljungman M, Ried T, Smale S, Rajapakse I: **Functional organization of the human 4D Nucleome.** *Proc Natl Acad Sci U S A* 2015, **112**:8002-8007.
  44. Misteli T: **Beyond the sequence: cellular organization of genome function.** *Cell* 2007, **128**:787-800.
  45. Sexton T, Schober H, Fraser P, Gasser SM: **Gene regulation through nuclear organization.** *Nat Struct Mol Biol* 2007, **14**:1049-1055.
  46. Zullo JM, Demarco IA, Piqué-Regi R, Gaffney DJ, Epstein CB, Spooner CJ, Luperchio TR, Bernstein BE, Pritchard JK, Reddy KL **et al.**: **DNA sequence-dependent compartmentalization and silencing of chromatin at the nuclear lamina.** *Cell* 2012, **149**:1474-1487.
  47. Guelen L, Pagie L, Brasset E, Meuleman W, Faza MB, Talhout W, Eussen BH, de Klein A, Wessels L, de Laat W **et al.**: **Domain organization of human chromosomes revealed by mapping of nuclear lamina interactions.** *Nature* 2008, **453**:948-951.
  48. Peric-Hupkes D, Meuleman W, Pagie L, Bruggeman SWM, Solovei I, Brugman W, Gräf S, Flicek P, Kerkhoven RM, van Lohuizen M **et al.**: **Molecular maps of the reorganization of genome-nuclear lamina interactions during differentiation.** *Mol Cell* 2010, **38**:603-613.
  49. Lee DC, Welton KL, Smith ED, Kennedy BK: **A-type nuclear lamins act as transcriptional repressors when targeted to promoters.** *Exp Cell Res* 2009, **315**:996-1007.
  50. Reddy KL, Zullo JM, Bertolino E, Singh H: **Transcriptional repression mediated by repositioning of genes to the nuclear lamina.** *Nature* 2008, **452**:243-247.
  51. Ramdas NM, Shivashankar GV: **Cytoskeletal control of nuclear morphology and chromatin organization.** *J Mol Biol* 2015, **427**:695-706.
  52. Wang Y, Nagarajan M, Uhler C, Shivashankar GV: **Orientation and repositioning of chromosomes correlate with cell geometry-dependent gene expression.** *Mol Biol Cell* 2017, **28**:1997-2009.
  53. Le HQ, Ghatak S, Yeung C-YC, Tellkamp F, Günschmann C, Dieterich C, Yeroslaviz A, Habermann B, Pombo A, Niessen CM **et al.**: **Mechanical regulation of transcription controls Polycomb-mediated gene silencing during lineage commitment.** *Nat Cell Biol* 2016, **18**:864-875.
  54. Tajik A, Zhang Y, Wei F, Sun J, Jia Q, Zhou W, Singh R, Khanna N, Belmont AS, Wang N: **Transcription upregulation via force-induced direct stretching of chromatin.** *Nat Mater* 2016, **15**:1287-1296.
- This paper provides the most direct evidence to date that applying a force with a magnetic bead at the cell surface via integrins induces transcriptional upregulation of an artificial transgene.
55. Almonacid M, Terret M-E, Verlhac M-H: **Control of nucleus positioning in mouse oocytes.** *Semin Cell Dev Biol* 2018, **82**:34-40.
  56. Tang N, Marshall WF: **Centrosome positioning in vertebrate development.** *J Cell Sci* 2012, **125**:4951-4961.
  57. Szollosi D, Calarco P, Donahue RP: **Absence of centrioles in the first and second meiotic spindles of mouse oocytes.** *J Cell Sci* 1972, **11**:521-541.
  58. Dalton CM, Carroll J: **Biased inheritance of mitochondria during asymmetric cell division in the mouse oocyte.** *J Cell Sci* 2013, **126**:2955-2964.
  59. Halet G, Carroll J: **Rac activity is polarized and regulates meiotic spindle stability and anchoring in mammalian oocytes.** *Dev Cell* 2007, **12**:309-317.
  60. FitzHarris G, Marangos P, Carroll J: **Changes in endoplasmic reticulum structure during mouse oocyte maturation are controlled by the cytoskeleton and cytoplasmic dynein.** *Dev Biol* 2007, **305**:133-144.
  61. Almonacid M, Ahmed WW, Bussonnier M, Mailly P, Betz T, Voituriez R, Gov NS, Verlhac M-H: **Active diffusion positions the nucleus in mouse oocytes.** *Nat Cell Biol* 2015, **17**:470-479.
  62. Almonacid M, El-Hayek S, Othmani A, Queguiner I, Coulpier F, Lemoine S, Bastianelli L, Klein C, Piolot T, Mailly P **et al.**: **Active fluctuations modulate gene expression in mouse oocytes.** *bioRxiv* 2018 <http://dx.doi.org/10.1101/347310>.



63. Hampoelz B, Azou-Gros Y, Fabre R, Markova O, Puech P-H, Lecuit T: **Microtubule-induced nuclear envelope fluctuations control chromatin dynamics in *Drosophila* embryos.** *Development* 2011, **138**:3377-3386.
64. Chaigne A, Campillo C, Gov NS, Voituriez R, Azoury J, Umaña-Díaz C, Almonacid M, Queguiner I, Nassoy P, Sykes C *et al.*: **A soft cortex is essential for asymmetric spindle positioning in mouse oocytes.** *Nat Cell Biol* 2013, **15**:958-966.
65. Chaigne A, Campillo C, Gov NS, Voituriez R, Sykes C, Verlhac MH, Terret ME: **A narrow window of cortical tension guides asymmetric spindle positioning in the mouse oocyte.** *Nat Commun* 2015, **6**:6027.
66. Borsos M, Torres-Padilla M-E: **Building up the nucleus: nuclear organization in the establishment of totipotency and pluripotency during mammalian development.** *Genes Dev* 2016, **30**:611-621.
67. Chen B, Gilbert LA, Cimini BA, Schnitzbauer J, Zhang W, Li G-W, Park J, Blackburn EH, Weissman JS, Qi LS *et al.*: **Dynamic imaging of genomic loci in living human cells by an optimized CRISPR/Cas system.** *Cell* 2013, **155**:1479-1491.
68. Nelles DA, Fang MY, O'Connell MR, Xu JL, Markmiller SJ, Doudna JA, Yeo GW: **Programmable RNA tracking in live cells with CRISPR/Cas9.** *Cell* 2016, **165**:488-496.