Frontiers in Bioscience Landmark

Front. Biosci. (Landmark Ed) **2023**; 28(12): 350
https://doi.org/10.31083/j.fbl2812350

Review

The Role of Nucleoporins in Cardiac Tissue Development and Disease

Xiaocong Chen[1,†], Rui Shi[1,†], Yu Luo[2,*], Liang Xu[3,4,*]

[1]Department of Obstetrics and Gynecology, East Hospital, Tongji University School of Medicine, 200120 Shanghai, China
[2]Department of Cardiology, Zigong Fourth People's Hospital, 643099 Zigong, Sichuan, China
[3]Key Laboratory of Arrhythmias of the Ministry of Education of China, Research Center for Translational Medicine, Shanghai East Hospital, Tongji University School of Medicine, 200120 Shanghai, China
[4]Department of Cardiology, Shanghai East Hospital, Tongji University School of Medicine, 200120 Shanghai, China
*Correspondence: wangyily1839@126.com (Yu Luo); xuliang_east@126.com (Liang Xu)
[†]These authors contributed equally.

Academic Editor: Natascia Tiso
Submitted: 7 June 2023 Revised: 22 August 2023 Accepted: 6 September 2023 Published: 27 December 2023

Abstract

Nuclear pore complexes (NPCs) are intricate intracellular structures composed of approximately 30 nuclear pore proteins (NUPs) that regulate the transport of materials between the nucleus and cytoplasm in eukaryotic cells. The heart is a crucial organ for sustaining the vital functions of the body, pumping blood rich in nutrients and energy to all organs and tissues. Recent studies have shown that NPCs play pivotal roles not only in normal cardiac physiological processes such as myocardial cell proliferation and differentiation but also in various pathological processes such as ischemic and hypoxic myocardial injury. Due to their mass and complicated nature, the structures of NPCs have been challenging to identify by the scientific community. With the development of cryo-electron microscopy and advanced sampling techniques, researchers have made significant progress in understanding the structures of NPCs. This review aims to summarize the latest research on the structural aspects of NPCs and their roles in cardiac physiology and pathology, increase the understanding of the intricate mechanisms of NPC actions, provide valuable insights into the pathogenesis of heart diseases and describe the development of potential novel therapeutic strategies.

Keywords: nuclear pore complex; heart; development; diseases; mechanism

1. Introduction

Nucleoporins (NUPs) constitute a class of proteins that form the nuclear pore complex (NPC), which is critical for the transport of molecules between the nucleus and cytoplasm in eukaryotic cells [1]. NUPs form the structure of an NPC and mediate the selective transport of molecules such as RNAs and proteins [2–4]. They play distinct roles in an NPC; some function as receptors for transport factors, others form the permeability barrier and another group of NPCs are involved in NPC assembly [5,6]. NPCs are also involved in maintaining the structural integrity of an NPC and in regulating the flow of macromolecules mediated by NPCs. An NPC is a large assembly of protein molecules and plays a crucial role in the transport of molecules between the nucleus and the cytoplasm [7]. As the "gatekeeper" of the nucleus, NPCs vary in composition and structure across organisms, tissues, physiological states, and pathological conditions [7–10]. Moreover, NPCs perform multiple functions, including nucleoplasmic translocation, chromatin remodeling, regulation of gene expression, and DNA repair, through various mechanisms [11–16].

2. Exploration of the NPC Structure

The nucleus is surrounded by a double nuclear membrane that divides the cell into two parts, the nucleus and the cytoplasm; therefore, the nuclear membrane is one of the features that distinguishes eukaryotes from prokaryotes [17]. In eukaryotic cells, transcription and translation occur at different sites. RNA transcription occurs in the nucleus, while protein translation takes place in the cytoplasm [18,19]. The NPC is an important bridge for nucleoplasmic transport, consisting of large proteins that assemble bidirectionally (Fig. 1). They are embedded in the nuclear membrane and consist of four circular scaffolds: the cytoplasmic ring (CR), the inner ring (IR), the nuclear ring (NR), and the luminal ring (LR) [20]. Cytoplasmic fibrils are attached to the cytoplasmic ring, while the nuclear basket is attached to the nucleoplasmic ring [2,21–24].

The structure of the nuclear membrane on the oocyte nucleus was first observed by electron microscopy in 1950, and nuclear pores were then described [25]. In 1959, Waston used terms such as pore complex to describe a complex, which appears as a relatively independent cylindrical structure on the nuclear membrane [26]. The eightfold symmetric cylindrical structure of an assembled NPC was confirmed in 1967 [27]. With advancements in biotechnology, knowledge of NPCs has advanced, and the determination of its molecular mass, production of structural models, preparation of antibodies, and cloning of phenylalanine-glycine (FG)-NUP have all been accomplished in the past century [28–31].

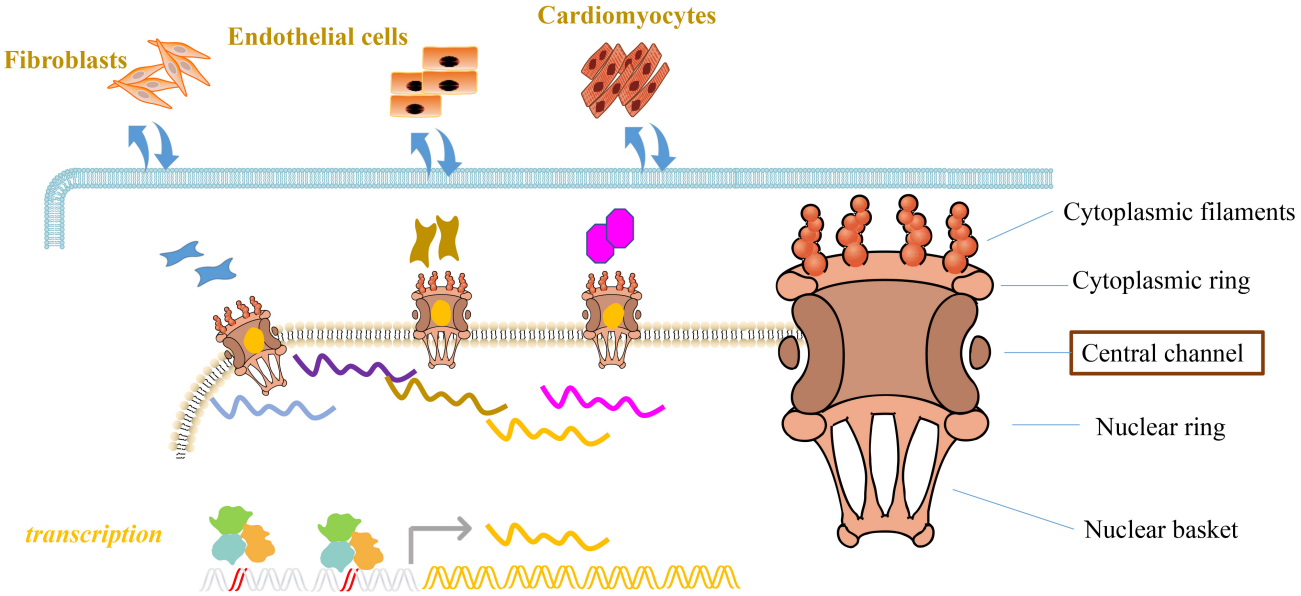

Fig. 1. The structure of nuclear pore complexes (NPCs) and the function of nucleoporins in heart tissues. NPCs are anchored to the nuclear membrane and consist of four annular scaffolds: the cytoplasmic ring, the inner ring, the nuclear ring, and the luminal ring. As the gatekeepers of the nucleus, NPCs exhibit asymmetric modifications that are essential for nucleoplasmic transport. In heart tissues, NPCs perform multiple functions through various mechanisms, including nucleoplasmic translocation, chromatin remodeling, and gene expression regulation. In the nucleus, genes are transcribed into mRNA, which then enters the cytoplasm through nuclear pores, which are transcribed into proteins. Moreover, proteins in the cytoplasm can enter the nucleus through nuclear pores and participate in gene expression regulation. These processes are crucial for the normal growth and development of organisms.

In 1993, an NPC in yeast was isolated, and a cryo-electron microscope map was drawn, which revealed the NPC structure, which was an epoch-making discovery [32, 33]. In the early 21st century, Michael adopted a comprehensive approach to identify and locate each NPC in yeast [34]. The next two decades were followed by the realization that the NPC is a "behemoth" composed of multiple modules. In other words, multiple different nucleoporins constitute the subcellular structural complex of a nucleoporin [3]. Multimers containing NUP62 (yeast NSP1) isolated from *Xenopus laevis* and yeast constitute proof of the NPC multimeric structure [35,36].

With the development of visualization-related technologies, including immune electron microscopy and cryo-electron tomography (cryo-ET), the three-dimensional location and *in vitro* reconstruction of NPCs have been realized, which has led to more intuitive understanding of their spatial structure [23,31,33,34,37]. The detailed molecular structures of NPCs are gradually being determined, including those from the Baker's yeast *Saccharomyces cerevisiae*, the slime mold *Dictyocele dictyocele*, *Xenopus laevis* oocytes, and the unicellular algae *Chlamydomonas reinhinella* to those in cultured human cells [38–40].

The NPC is one of the largest macromolecular complexes in a cell, and it consists of an eightfold rotationally symmetric core with asymmetric distribution on the nucleoplasmic surface [3,41,42]. The size, number, and conformation of the NPC vary across organisms, tissues, and physiological states, making it a complex structure to study [43,44]. An NPC is constructed from multiple copies of more than 30–50 different NUPs, with different molecular mass, ranging from 50 kDa to 358 kDa [2,34,45–48]. Most of these NUPs are conserved in eukaryotes, indicating their importance in the function of NPCs. The composition of an NPC also varies within the same organism, depending on the function of the cell, the degree of cell differentiation, and the cell cycle of the cell in which it is located [49].

Scientists have been studying the structure and function of NPCs for decades using techniques such as low-resolution imaging and high-resolution composition analysis. However, reproducing a complete high-resolution structure of an NPC remains a primary challenge for NPC researchers [40]. Despite this difficulty, our knowledge of the structure of NPCs continues to evolve, providing us with a better understanding of the assembly of different protein copies of cellular NPCs.

3. Progress in Understanding the NPC Structure

In recent years, significant progress has been made in the study of the structure of the NPC [50]. Multiple studies have revealed the near-atomic resolution structure of the human NPC obtained by cryo-electron microscopy, as well as images of single particles of the vertebrate NPC from the African clawed toad obtained via cryo-electron microscopy

[9,20,51–53]. These findings are groundbreaking in terms of both the understanding of the symmetric core structure and asymmetric distribution of the NPC.

One study reported the complete structural set capturing all linker–scaffold interactions of the symmetric core, as determined by docking the complete linker–scaffold structure to a cryo-ET reconstruction of a human NPC resolved at ~12 Å and an *in situ* cryo-ET reconstruction of a brewer's yeast NPC resolved at ~25 Å [52]. From these finding at the near-atomic level, the molecular structure and evolutionary conservation of the linker–scaffold of the human NPC has been established.

Another study reported a 70-MKDa model of the human NPC scaffold based on artificial intelligence predictions. By fitting individual NUP structural models to high-resolution cryo-electron chromatograms of human NPC in contracted and expanded conformational states, the NPC structure was resolved through dynamic states of flux. This finding indicated conformational changes occur during both expansion and contraction, and this dynamic model has provided the basis for identifying the precise anchoring points of intrinsically disordered NUPs [53].

The study of the human NPC cytoplasmic surface has been a topic of interest, particularly NPC asymmetric modifications, for scientists. Researchers such as Bley at Caltech have made significant progress in this area, establishing a near-atomic composite structure through a combination of techniques, including *in vitro* complex reconstruction, crystal structure determination, quantitative docking, and *in vivo* validation [51]. One important finding is the role of the cytoplasmic filament (CF) NUP in targeting exported mRNA by modifying mRNP in preparation for subsequent translation [3,54]. This process is crucial for protein function. The assembly of the CF NUP complex (CFNC) module has been shown to be conserved in humans and *C. thermophilum*. It consists of a central heterotrimeric coiled-coil hub that tethers two independent mRNP-remodeling complexes together [51]. This is an extension and expansion of previous studies on CFNC in *S. cerevisiae* [40,55,56].

Another key component on the NUP cytoplasmic surface is NUP358, which is a postnatally acquired animal-specific CF component and the largest protein predicted to be in NPCs [20,57,58]. Biochemical analysis and determination of its crystal structure have revealed that NUP358 is composed of 16 distinct structural domains. These domains include an N-terminal S-shaped α-helical solenoid, a coiled-coil oligomeric element, numerous Ran-interacting structural domains, an E3 ligase structural domain, and a C-terminal alanine isomerase structural domain. The basic FG repeat sequence is involved in selective cargo passaging mediated through a disordered C-terminal region [20,51].

Nup358 is one of the key components of the CR. The cytoplasmic loop of the African clawed toad NPC was reconstructed using single-particle cryo-electron microscopy, and the near-complete NPC cytoplasmic loop structure in African clawed toad oocytes was determined using AlphaFold [9,20]. Cryo-electron microscopy-based structural maps were obtained, showing that each CR subunit included five copies of Nup358, two copies of Nup93, two copies of Nup205, and two copies of the Y-complex. The final model of the African clawed toad CR was generated via AlphaFold prediction [9].

Another team also reported five copies of Nup358, four of which were sandwiched around the internal and external regions of a Y-complex to stabilize the CR, with a fifth Nup358 copy located in the center of the sandwiched cluster. The binding pattern was identified, and the homo-oligomeric coiled-coil structure of Nup358 was predicted by AlphaFold. This result suggests the possibility that Nup358 is recruited to NPCs and lowers the threshold for Nup358 condensation during NPC biogenesis [20].

4. NPC and Heart Development

The development of the heart is a complex process that is not fully understood. The heart is the first organ to form during embryonic development, and cardiomyocytes originate in the mesoderm [59]. During myocardial differentiation, a series of transcription factors are activated, but the molecular regulatory mechanisms involved in this process are still not clear [60].

Recent research has shown that NUPs expression and distribution are significantly altered during myocardial differentiation [61,62]. The annulate lamellae, which are thought to be a collection of extraneous NUPs, are almost all absent in stem cells, whereas they are significantly more abundant in mature cardiomyocytes. This suggests that fewer extraneous NUPs are in stem cells, allowing the transport through the nuclear membrane into the nucleus to be maximized [63].

Cardiomyocytes highly proliferate during the mammalian embryonic period and gradually cease to proliferate in neonates. Differentiated mature cardiomyocytes show little proliferative capacity and in contrast promote cardiac growth through polyploidization and increased cell volume, eventually entering a state of terminal differentiation [64]. Nuclear pores are the only pathways for the entry and exit of macromolecules into and out of the nucleus, and they regulate gene expression by regulating the entry of signal transduction proteins into the nucleus [65]. Recent studies have shown that as cardiomyocytes mature, the number of nuclear pores in the nuclear membrane decreases, paralleling the reduced transmission of cytoplasmic signals to the nucleus [61]. This alters the transcription of a range of genes, as it affects proliferative signaling, consistent with previous reports of neurons during the progressive deterioration of nuclear pore structure and function during aging [61,66].

As a channel for the transfer of information between the nucleoplasm and the nucleus, changes in the number of nuclear pores are closely related to nuclear input [67].

To explore the reasons underlying the decrease in the number of nuclear pores during cardiac cell maturation, scientists studied nuclear pores in mice. The results showed that mammalian cardiomyocytes with a marked reduction in the number of nuclear pores during maturation reduced the nuclear input of signal transduction molecules, decreased gene expression, and ameliorated adverse myocardial remodeling caused by signals, such as stress signals [61,68]. Stem cell-derived cardiomyocytes ensure the translocation of nuclear transport regulators such as cell cycle regulators and cardiac cells-derived transcription factors, suggesting that NPCs may influence the proliferative capacity of cardiomyocytes by participating in the regulation of the cell cycle. In addition, the nuclear transport requirements necessary for cardiomyocyte proliferation may also be met by structural adaptations of NPCs, thereby increasing internucleoplasmic communication [62]. Taken together, adaptive changes in NPCs may be responses to changing cellular needs during cardiac tissue development.

5. NPC and Cardiovascular Disease

NPCs are crucial components of the nuclear membrane in eukaryotic cells. NUPs comprise two major classes: structural scaffold NUPs and peripheral component NUPs. These NUPs are critical for regulating molecular transport to and from the nuclear membrane, and they play vital roles in many important cellular activities [16]. However, NUPs have been associated with various diseases, including immune disorders, neurological disorders, cancers, and cardiovascular diseases [69].

Among these diseases, cardiovascular disease is currently a hot topic of research. NUPs have been found to play important roles in the pathophysiology of cardiovascular diseases (Fig. 1). In fact, the first link between NUPs and cardiovascular disease was reported in a study of a family with atrial fibrillation (AF) with member who succumbed to sudden death in early childhood. The study identified a mutation in NUP155, R391H, which affected the nuclear localization of NUP155 and reduced the permeability of the nuclear membrane [70,71]. Subsequent animal and cell experimental studies confirmed these findings, identifying NUP155 as a driver of AF. The possible mechanism involved a lamin A/C mutation that impaired lamin A/C interaction with NUP155, resulting in impaired export of Hsp70 mRNA and nuclear import of Hsp70 protein. This damage led to AF and early incidents of sudden cardiac death [72]. Another study, conducted by genetically analyzing samples from subjects with heterotaxy, a congenital heart disease caused by abnormalities in left and right body patterns, revealed Nup188 with previously undocumented genetic Copy number variations (CNVs) [73]. To elucidate the importance of nuclear pore proteins in congenital heart disease, Del Viso *et al*. [74] used the African clawed toad as a model animal and revealed that deletion of the inner ring nuclear pore proteins, including Nup188, was critical to the absence of cilia in embryonic development. This finding may explain the connection of cilium deletion and ectopic Nup188 duplication in patients.

Recent research has elucidated the relationship between NUPs and cardiovascular diseases, specifically metabolic alterations in cardiomyocytes under ischemic and hypoxic conditions. However, the mechanisms underlying these relationships are still unclear. One NUP, namely, Nup35, has been shown to regulate cardiomyocyte NHE1 expression and modulate pH homeostasis under normal and hypoxic-ischemic conditions by controlling NHE1 mRNA nucleoplasmic transport. Proper expression of NUPs is crucial to maintain normal metabolic functions of cardiomyocytes [75]. Another study revealed that Nup93 was significantly downregulated in hypoxic cardiomyocytes, which was associated with abnormal NUP expression and ultimately led to cardiomyocyte injury and death. The expression of Nup93 was negatively correlated with that of Atrial natriuretic peptide (ANP) and Brain natriuretic peptide (BNP), molecular markers of cardiomyocyte function. Furthermore, knockdown of Nup93 regulated the transcription of various mRNAs in cardiac myocytes, most notably Yes-associated protein 1 (YAP1), which resulted in abnormalities in oxidative phosphorylation and ribosome biogenesis in cardiac myocytes [76]. These findings highlight the importance of proper NUP expression in maintaining healthy cardiomyocyte function and suggest potential therapeutic targets for cardiovascular disease.

Recent research has indicated that NUP plays a crucial role in cell-specific gene regulation, particularly in cardiac electrophysiology and electrocardiography-measurable activity. Potassium ion channels, known as Kcna4, are complex voltage-gated ion channels that are essential for regulating myocardial membrane potential by enabling the transport of potassium ions across the excitable cell membrane [77]. Nup50, which is localized to the nucleus of cardiomyocytes, has been shown to bind directly to the Kcna4 FG-repeat domain, thus enhancing Kcna4 expression at the transcriptional and translational levels [78]. Another important ion channel, SCN5A, mediates the voltage-dependent sodium ion permeability of excitable membranes [79]. Nup107 is involved in regulating Scn5a mRNA output through the control of nucleoplasmic transport of Scn5a mRNA. Furthermore, Nup107 has been observed to rapidly regulate cardiomyocyte Nav1.5 channels posttranscriptionally in cardiomyocytes and heart tissue injured under hypoxic and ischemic conditions [80]. These findings suggest that the Nup107 protein is associated with ischemic heart damage.

Taken together, these studies suggest that abnormalities in Nups can lead to cardiovascular disease, particularly metabolism-related ischemic/hypoxic cardiomyocyte-damaging diseases. Further research into the role of NUPs is necessary to better understand the pathogenesis of cardiovascular disease and identify potential therapeutic targets.

6. Conclusions

NPCs play crucial roles in the transport of molecules between the nucleus and cytoplasm of a cell. The characteristics of NPCs vary across different organisms and disease conditions. Over the past decade, advancements in biochemical reconstructions, X-ray crystallography, mass spectrometry, mutagenesis, and cell biology have allowed scientists to reconstruct a human NPC at near-atomic resolution. Artificial intelligence has also been used to accurately model the components of NPCs. These advancements have provided a better understanding of the structure and function of NPCs. They have also shed light on how defects in NPCs lead to various diseases, especially cardiac diseases. The plasticity of NPCs ensures that they play essential roles in cardiac developmental processes such as myocardial differentiation and proliferation. Cardiovascular diseases are polygenic complex diseases, and NUPs are considered a potential causes of various heart diseases. Further research in this area may lead to the development of new therapies for these conditions.

Abbreviations

NPC, Nuclear pore complex; NUP, nuclear pore protein; CR, cytoplasmic ring; IR, the inner ring; NR, the nuclear ring; LR, luminal ring; FG, phenylalanine-glycine; cryo-ET, cryo-electron tomography; CF, cytoplasmic filament; CFNC, CF NUP complex; AF, atrial fibrillation; ANP, atrial natriuretic peptide; BNP, brain natriuretic peptide; YAP1, Yes-associated protein 1; CNVs, copy number variations.

Author Contributions

LX and YL conceptualized the work. XC and RS screened and analyzed the relevant literature. XC wrote the original draft, and RS made the figure. RS and YL interpreted the data for the work, critically reviewed the work for important intellectual content, and edited the manuscript. LX investigated all aspects of the work and supervised the whole process. All authors contributed editorial changes to the manuscript. All the authors have read and approved the final manuscript. All authors have participated sufficiently in the work to take responsibility for appropriate portions of the content and agreed to be accountable for all aspects of the work by ensuring its accuracy or integrity to the public.

Ethics Approval and Consent to Participate

Not applicable.

Acknowledgment

Not applicable.

Funding

This work was supported by the grant from the National Natural Science Foundation of China for the General Program (82270397 to Liang Xu), and Top-level Clinical Discipline Project of Shanghai Pudong District (PWYgf 2021-01).

Conflict of Interest

The authors declare no conflict of interest.

References

[1] Ptak C, Aitchison JD, Wozniak RW. The multifunctional nuclear pore complex: a platform for controlling gene expression. Current Opinion in Cell Biology. 2014; 28: 46–53.

[2] Schwartz TU. The Structure Inventory of the Nuclear Pore Complex. Journal of Molecular Biology. 2016; 428: 1986–2000.

[3] Lin DH, Hoelz A. The Structure of the Nuclear Pore Complex (An Update). Annual Review of Biochemistry. 2019; 88: 725–783.

[4] Terry LJ, Wente SR. Nuclear mRNA export requires specific FG nucleoporins for translocation through the nuclear pore complex. The Journal of Cell Biology. 2007; 178: 1121–1132.

[5] Fernandez-Martinez J, Kim SJ, Shi Y, Upla P, Pellarin R, Gagnon M, *et al*. Structure and Function of the Nuclear Pore Complex Cytoplasmic mRNA Export Platform. Cell. 2016; 167: 1215–1228.e25.

[6] Kozai T, Fernandez-Martinez J, van Eeuwen T, Gallardo P, Kapinos LE, Mazur A, *et al*. Dynamic molecular mechanism of the nuclear pore complex permeability barrier. bioRxiv. 2023. (preprint)

[7] Ashkenazy-Titelman A, Atrash MK, Boocholez A, Kinor N, Shav-Tal Y. RNA export through the nuclear pore complex is directional. Nature Communications. 2022; 13: 5881.

[8] Wang J, Pei G, Wang Y, Wu D, Liu X, Li G, *et al*. Phase separation of the nuclear pore complex facilitates selective nuclear transport to regulate plant defense against pathogen and pest invasion. Molecular Plant. 2023; 16: 1016–1030.

[9] Zhu X, Huang G, Zeng C, Zhan X, Liang K, Xu Q, *et al*. Structure of the cytoplasmic ring of the *Xenopus laevis* nuclear pore complex. Science. 2022; 376: eabl8280.

[10] Varberg JM, Unruh JR, Bestul AJ, Khan AA, Jaspersen SL. Quantitative analysis of nuclear pore complex organization in *Schizosaccharomyces pombe*. Life Science Alliance. 2022; 5: e202201423.

[11] Gay S, Foiani M. Nuclear envelope and chromatin, lock and key of genome integrity. International Review of Cell and Molecular Biology. 2015; 317: 267–330.

[12] Pascual-Garcia P, Capelson M. Nuclear pores as versatile platforms for gene regulation. Current Opinion in Genetics & Development. 2014; 25: 110–117.

[13] Schellhaus AK, De Magistris P, Antonin W. Nuclear Reformation at the End of Mitosis. Journal of Molecular Biology. 2016; 428: 1962–1985.

[14] Imamoto N, Funakoshi T. Nuclear pore dynamics during the cell cycle. Current Opinion in Cell Biology. 2012; 24: 453–459.

[15] Van de Vosse DW, Wan Y, Wozniak RW, Aitchison JD. Role of the nuclear envelope in genome organization and gene expression. Wiley Interdisciplinary Reviews. Systems Biology and Medicine. 2011; 3: 147–166.

[16] Beck M, Hurt E. The nuclear pore complex: understanding its function through structural insight. Nature Reviews. Molecular Cell Biology. 2017; 18: 73–89.

[17] Tai L, Yin G, Sun F, Zhu Y. Cryo-electron Microscopy Reveals the Structure of the Nuclear Pore Complex. Journal of Molecular Biology. 2023; 435: 168051.

[18] Köhler A, Hurt E. Exporting RNA from the nucleus to the cytoplasm. Nature Reviews. Molecular Cell Biology. 2007; 8: 761–

773.
[19] Kornblihtt AR, Schor IE, Alló M, Dujardin G, Petrillo E, Muñoz MJ. Alternative splicing: a pivotal step between eukaryotic transcription and translation. Nature Reviews. Molecular Cell Biology. 2013; 14: 153–165.
[20] Fontana P, Dong Y, Pi X, Tong AB, Hecksel CW, Wang L, *et al*. Structure of cytoplasmic ring of nuclear pore complex by integrative cryo-EM and AlphaFold. Science. 2022; 376: eabm9326.
[21] Fahrenkrog B, Aebi U. The nuclear pore complex: nucleocytoplasmic transport and beyond. Nature Reviews. Molecular Cell Biology. 2003; 4: 757–766.
[22] Dickmanns A, Kehlenbach RH, Fahrenkrog B. Nuclear Pore Complexes and Nucleocytoplasmic Transport: From Structure to Function to Disease. International Review of Cell and Molecular Biology. 2015; 320: 171–233.
[23] von Appen A, Beck M. Structure Determination of the Nuclear Pore Complex with Three-Dimensional Cryo electron Microscopy. Journal of Molecular Biology. 2016; 428: 2001–2010.
[24] Vial A, Costa L, Dosset P, Rosso P, Boutières G, Faklaris O, *et al*. Structure and mechanics of the human nuclear pore complex basket using correlative AFM-fluorescence superresolution microscopy. Nanoscale. 2023; 15: 5756–5770.
[25] CALLAN HG, TOMLIN SG. Experimental studies on amphibian oocyte nuclei. I. Investigation of the structure of the nuclear membrane by means of the electron microscope. Proceedings of the Royal Society of London. Series B, Biological Sciences. 1950; 137: 367–378.
[26] WATSON ML. Further observations on the nuclear envelope of the animal cell. The Journal of Biophysical and Biochemical Cytology. 1959; 6: 147–156.
[27] Gall JG. Octagonal nuclear pores. The Journal of Cell Biology. 1967; 32: 391–399.
[28] Hurt EC. A novel nucleoskeletal-like protein located at the nuclear periphery is required for the life cycle of Saccharomyces cerevisiae. The EMBO Journal. 1988; 7: 4323–4334.
[29] Reichelt R, Holzenburg A, Buhle EL, Jr, Jarnik M, Engel A, Aebi U. Correlation between structure and mass distribution of the nuclear pore complex and of distinct pore complex components. The Journal of Cell Biology. 1990; 110: 883–894.
[30] Hinshaw JE, Carragher BO, Milligan RA. Architecture and design of the nuclear pore complex. Cell. 1992; 69: 1133–1141.
[31] Panté N, Bastos R, McMorrow I, Burke B, Aebi U. Interactions and three-dimensional localization of a group of nuclear pore complex proteins. The Journal of Cell Biology. 1994; 126: 603–617.
[32] Rout MP, Blobel G. Isolation of the yeast nuclear pore complex. The Journal of Cell Biology. 1993; 123: 771–783.
[33] Akey CW, Radermacher M. Architecture of the Xenopus nuclear pore complex revealed by three-dimensional cryo-electron microscopy. The Journal of Cell Biology. 1993; 122: 1–19.
[34] Rout MP, Aitchison JD, Suprapto A, Hjertaas K, Zhao Y, Chait BT. The yeast nuclear pore complex: composition, architecture, and transport mechanism. The Journal of Cell Biology. 2000; 148: 635–651.
[35] Finlay DR, Meier E, Bradley P, Horecka J, Forbes DJ. A complex of nuclear pore proteins required for pore function. The Journal of Cell Biology. 1991; 114: 169–183.
[36] Dabauvalle MC, Loos K, Scheer U. Identification of a soluble precursor complex essential for nuclear pore assembly in vitro. Chromosoma. 1990; 100: 56–66.
[37] Beck M, Förster F, Ecke M, Plitzko JM, Melchior F, Gerisch G, *et al*. Nuclear pore complex structure and dynamics revealed by cryoelectron tomography. Science. 2004; 306: 1387–1390.
[38] Alber F, Dokudovskaya S, Veenhoff LM, Zhang W, Kipper J, Devos D, *et al*. The molecular architecture of the nuclear pore complex. Nature. 2007; 450: 695–701.
[39] Amlacher S, Sarges P, Flemming D, van Noort V, Kunze R, Devos DP, *et al*. Insight into structure and assembly of the nuclear pore complex by utilizing the genome of a eukaryotic thermophile. Cell. 2011; 146: 277–289.
[40] Kim SJ, Fernandez-Martinez J, Nudelman I, Shi Y, Zhang W, Raveh B, *et al*. Integrative structure and functional anatomy of a nuclear pore complex. Nature. 2018; 555: 475–482.
[41] Hampoelz B, Andres-Pons A, Kastritis P, Beck M. Structure and Assembly of the Nuclear Pore Complex. Annual Review of Biophysics. 2019; 48: 515–536.
[42] Hinshaw JE, Milligan RA. Nuclear pore complexes exceeding eightfold rotational symmetry. Journal of Structural Biology. 2003; 141: 259–268.
[43] Grossman E, Medalia O, Zwerger M. Functional architecture of the nuclear pore complex. Annual Review of Biophysics. 2012; 41: 557–584.
[44] Zimmerli CE, Allegretti M, Rantos V, Goetz SK, Obarska-Kosinska A, Zagoriy I, *et al*. Nuclear pores dilate and constrict in cellulo. Science. 2021; 374: eabd9776.
[45] Cronshaw JM, Krutchinsky AN, Zhang W, Chait BT, Matunis MJ. Proteomic analysis of the mammalian nuclear pore complex. The Journal of Cell Biology. 2002; 158: 915–927.
[46] Ori A, Banterle N, Iskar M, Andrés-Pons A, Escher C, Khanh Bui H, *et al*. Cell type-specific nuclear pores: a case in point for context-dependent stoichiometry of molecular machines. Molecular Systems Biology. 2013; 9: 648.
[47] Hoelz A, Glavy JS, Beck M. Toward the atomic structure of the nuclear pore complex: when top down meets bottom up. Nature Structural & Molecular Biology. 2016; 23: 624–630.
[48] Doye V, Hurt E. From nucleoporins to nuclear pore complexes. Current Opinion in Cell Biology. 1997; 9: 401–411.
[49] Hurt E, Beck M. Towards understanding nuclear pore complex architecture and dynamics in the age of integrative structural analysis. Current Opinion in Cell Biology. 2015; 34: 31–38.
[50] Jiang D. Building the nuclear pore complex. Science. 2022; 376: 1172–1173.
[51] Bley CJ, Nie S, Mobbs GW, Petrovic S, Gres AT, Liu X, *et al*. Architecture of the cytoplasmic face of the nuclear pore. Science. 2022; 376: eabm9129.
[52] Petrovic S, Samanta D, Perriches T, Bley CJ, Thierbach K, Brown B, *et al*. Architecture of the linker-scaffold in the nuclear pore. Science. 2022; 376: eabm9798.
[53] Mosalaganti S, Obarska-Kosinska A, Siggel M, Taniguchi R, Turoňová B, Zimmerli CE, *et al*. AI-based structure prediction empowers integrative structural analysis of human nuclear pores. Science. 2022; 376: eabm9506.
[54] Stewart M. Polyadenylation and nuclear export of mRNAs. The Journal of Biological Chemistry. 2019; 294: 2977–2987.
[55] Gaik M, Flemming D, von Appen A, Kastritis P, Mücke N, Fischer J, *et al*. Structural basis for assembly and function of the Nup82 complex in the nuclear pore scaffold. The Journal of Cell Biology. 2015; 208: 283–297.
[56] Allegretti M, Zimmerli CE, Rantos V, Wilfling F, Ronchi P, Fung HKH, *et al*. In-cell architecture of the nuclear pore and snapshots of its turnover. Nature. 2020; 586: 796–800.
[57] Braun DA, Sadowski CE, Kohl S, Lovric S, Astrinidis SA, Pabst WL, *et al*. Mutations in nuclear pore genes NUP93, NUP205 and XPO5 cause steroid-resistant nephrotic syndrome. Nature Genetics. 2016; 48: 457–465.
[58] Muir AM, Cohen JL, Sheppard SE, Guttipatti P, Lo TY, Weed N, *et al*. Bi-allelic Loss-of-Function Variants in NUP188 Cause a Recognizable Syndrome Characterized by Neurologic, Ocular, and Cardiac Abnormalities. American Journal of Human Genetics. 2020; 106: 623–631.
[59] Kelly RG. The heart field transcriptional landscape at single-cell

resolution. Developmental Cell. 2023; 58: 257–266.
[60] Paige SL, Plonowska K, Xu A, Wu SM. Molecular regulation of cardiomyocyte differentiation. Circulation Research. 2015; 116: 341–353.
[61] Han L, Mich-Basso JD, Li Y, Ammanamanchi N, Xu J, Bargaje AP, *et al*. Changes in nuclear pore numbers control nuclear import and stress response of mouse hearts. Developmental Cell. 2022; 57: 2397–2411.e9.
[62] Perez-Terzic C, Behfar A, Méry A, van Deursen JMA, Terzic A, Pucéat M. Structural adaptation of the nuclear pore complex in stem cell-derived cardiomyocytes. Circulation Research. 2003; 92: 444–452.
[63] Wu X, Kasper LH, Mantcheva RT, Mantchev GT, Springett MJ, van Deursen JM. Disruption of the FG nucleoporin NUP98 causes selective changes in nuclear pore complex stoichiometry and function. Proceedings of the National Academy of Sciences of the United States of America. 2001; 98: 3191–3196.
[64] Kirillova A, Han L, Liu H, Kühn B. Polyploid cardiomyocytes: implications for heart regeneration. Development. 2021; 148: dev199401.
[65] D'Angelo MA, Hetzer MW. Structure, dynamics and function of nuclear pore complexes. Trends in Cell Biology. 2008; 18: 456–466.
[66] D'Angelo MA, Raices M, Panowski SH, Hetzer MW. Age-dependent deterioration of nuclear pore complexes causes a loss of nuclear integrity in postmitotic cells. Cell. 2009; 136: 284–295.
[67] Knockenhauer KE, Schwartz TU. The Nuclear Pore Complex as a Flexible and Dynamic Gate. Cell. 2016; 164: 1162–1171.
[68] Han L, Choudhury S, Mich-Basso JD, Ammanamanchi N, Ganapathy B, Suresh S, *et al*. Lamin B2 Levels Regulate Polyploidization of Cardiomyocyte Nuclei and Myocardial Regeneration. Developmental Cell. 2020; 53: 42–59.e11.
[69] Burdine RD, Preston CC, Leonard RJ, Bradley TA, Faustino RS. Nucleoporins in cardiovascular disease. Journal of Molecular and Cellular Cardiology. 2020; 141: 43–52.
[70] Zhang X, Chen S, Yoo S, Chakrabarti S, Zhang T, Ke T, *et al*. Mutation in nuclear pore component NUP155 leads to atrial fibrillation and early sudden cardiac death. Cell. 2008; 135: 1017–1027.
[71] Oberti C, Wang L, Li L, Dong J, Rao S, Du W, *et al*. Genome-wide linkage scan identifies a novel genetic locus on chromosome 5p13 for neonatal atrial fibrillation associated with sudden death and variable cardiomyopathy. Circulation. 2004; 110: 3753–3759.
[72] Han M, Zhao M, Cheng C, Huang Y, Han S, Li W, *et al*. Lamin A mutation impairs interaction with nucleoporin NUP155 and disrupts nucleocytoplasmic transport in atrial fibrillation. Human Mutation. 2019; 40: 310–325.
[73] Fakhro KA, Choi M, Ware SM, Belmont JW, Towbin JA, Lifton RP, *et al*. Rare copy number variations in congenital heart disease patients identify unique genes in left-right patterning. Proceedings of the National Academy of Sciences of the United States of America. 2011; 108: 2915–2920.
[74] Del Viso F, Huang F, Myers J, Chalfant M, Zhang Y, Reza N, *et al*. Congenital Heart Disease Genetics Uncovers Context-Dependent Organization and Function of Nucleoporins at Cilia. Developmental Cell. 2016; 38: 478–492.
[75] Xu L, Pan L, Li J, Huang B, Feng J, Li C, *et al*. Nucleoporin 35 regulates cardiomyocyte pH homeostasis by controlling Na+-H+ exchanger-1 expression. Journal of Molecular Cell Biology. 2015; 7: 476–485.
[76] Pan L, Song XW, Song JC, Shi CY, Wang ZK, Huang SQ, *et al*. Downregulation of NUP93 aggravates hypoxia-induced death of cardiomyocytes in vitro through abnormal regulation of gene transcription. Acta Pharmacologica Sinica. 2023; 44: 969–983.
[77] Kang GJ, Xie A, Kim E, Dudley SC, Jr. miR-448 regulates potassium voltage-gated channel subfamily A member 4 (KCNA4) in ischemia and heart failure. Heart Rhythm. 2023; 20: 730–736.
[78] Gao X, Yu S, Guan Y, Shen Y, Xu L. Nucleoporin 50 mediates Kcna4 transcription to regulate cardiac electrical activity. Journal of Cell Science. 2021; 134: jcs256818.
[79] Han D, Tan H, Sun C, Li G. Dysfunctional Nav1.5 channels due to SCN5A mutations. Experimental Biology and Medicine. 2018; 243: 852–863.
[80] Guan Y, Gao X, Tang Q, Huang L, Gao S, Yu S, *et al*. Nucleoporin 107 facilitates the nuclear export of Scn5a mRNA to regulate cardiac bioelectricity. Journal of Cellular and Molecular Medicine. 2019; 23: 1448–1457.