Form 3

Abstract of Thesis

Name （Bilir Sukriye）	
Title	A study for the roles of Nup133, Nup153 and membrane fenestrae in post-mitotic nuclear pore complex formation （有糸分裂後核膜孔形成におけるNup133、Nup153、および開窓膜構造の役割の研究）

Abstract of Thesis

Nuclear pore complexes (NPC) are gates residing in the nuclear envelope (NE) and responsible for nucleo-cytoplasmic exchange. They are huge protein complexes including 8-fold of ~30 proteins which are called as nucleoporins. On the start of open-mitosis NE breaks down and all components of the NPC dispersed to the cytoplasm. At the end of mitosis these components are collected quickly to form the full NPC. This post-mitotic NPC reassembly is previously described to happen in an ordered stepwise process by starting with a seed nucleoporin structure. It has been previously reported that Y-complex components and Nup153 may be playing a role to initiate NPC formation according to observations made on mammalian cells by live fluorescence imaging. While this quick formation of NPC is also related to the accompanying membranal structure, the responsible mechanisms relating the membranal structures with possible seed nucleoporins for initiating the assembly of functional NPC remain unknown. To obtain direct evidence for this unknown problem, I used a novel experimental method including an artificial bead conjugated with a molecule of interest as an effector molecule to assemble the NPC. The beads are introduced into living cells and observed if the NPC, NE or their protein components are assembled at the effector molecules on the surface of the beads on post-mitosis. As a result of these experiments, it has been revealed that Nup133 (a component of Y-complex) and Nup153 are not enough alone to create a full NPC molecule. As an addition to this, it has been seen that both nucleoporins have the ability to collect NE-like double membrane and NPC-like structures under electron microscope with a different affinity for double layered membranes; Nup133 can collect double layered membranes more efficiently than Nup153. Also, membranes in relation with Nup133 has higher number of fenestrations than Nup153. In relation to these results, fenestrated endoplasmic reticulum (ER) structures were discovered as the major membranal structure during cell division which are also the pre-cursors of NE. Because of the early accumulation of Nup133 around chromosomes and high affinity of it for fenestrated membranes, I introduce here that it can collect and interact directly with these fenestrations residing on NE precursors while Nup153 can collect the other nucleoporins and they help together the quick assembly of NPC at the end of mitosis. As a result, it is introduced that instead of a single seed nucleoporin, multiple nucleoporins are working together with membranes and also possibly with chromosomes in different roles to initiate and finish NPC formation post-mitotically.

様式 7

論文審査の結果の要旨及び担当者

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論文審査の結果の要旨

申請者 Şükriye BILIR は、ヒト培養細胞を用いて、細胞分裂終期におこる核膜孔の再構築の仕組みを研究し、核膜前駆体となる小胞体膜に存在する小さな穴の構造が、核膜孔再構築に利用されることを、蛍光顕微鏡による生細胞観察と蛍光電子相関顕微鏡法（Live CLEM法）を用いて明らかにした。さらに、目的分子の細胞内での機能を調べる方法として、人工ビーズに目的分子を結合させたものを細胞内に導入する方法を用いた。この方法は、ビーズ周辺で起こる細胞内反応を調べることにより、目的分子の機能を知ることができる新規な方法である。この方法を用いて、核膜孔複合体構成タンパク質であるNup133とNup153の機能を検討し、これらの分子が核膜孔形成にそれぞれ異なる機能を持つことを明らかにした。生きた細胞内に核膜孔構造を含む核膜を人工的に形成させるのに成功したのは初めてであり、細胞核の研究に大きく貢献した。

よって、博士の学位を授与するに値すると認める。