The Role of the Actin Cytoskeleton in the Mitotic Nuclear Envelope Disassembly and Reassembly

M. Joensuu, G. Piras, M. Puhka, and E. Jokitalo

Institute of Biotechnology, Electron Microscopy Unit, PO Box 56, Fin-00014, University of Helsinki, Finland

merja.s.joensuu@helsinki.fi
Keywords: Mitosis, Endoplasmic reticulum, Nuclear envelope breakdown and reformation, Actin cytoskeleton

Endoplasmic reticulum (ER) is the most versatile cell organelle. It has several functions in the cell, including the synthesis and modification of proteins and lipids and regulation of cytoplasmic Ca^{2+} level. ER is classically divided into three subdomains, the nuclear envelope (NE), rough and smooth ER, that branch to generate a continuous network. Morphologically ER is composed of sheets and tubules.

It is not entirely known how the morphological and functional subdomains of the ER are created and regulated. Several structural elements, such as membrane proteins, cytoskeleton and lipid composition, have been proposed to be liable. It is likely that several different mechanisms contribute to the morphogenesis and regulation of the ER network. We have previously suggested that the morphology of the sheet structures might be stabilized by the ER membrane bound polysomes [1].

During cell division, nuclear envelope break down (NEBD) starts in prophase, after which it no longer can be morphologically distinguished from the ER network as a separate subdomain. Both live cell imaging [1,2] as well as analysis of TEM thin sections and electron tomography [1] have shown that NE membrane proteins spread into the ER network during mitosis, but it is not yet clear whether the distribution of the NE membrane proteins is homogeneous or concentrated on specific domains within the ER network.

We are currently studying the role of the actin cytoskeleton on the fate of the NE membrane movements during cell division using several microscopic methods, such as live cell imaging and TEM. Firstly, we are focusing in the early phases of the mitosis and the role of the actin cytoskeleton in the redistribution of the NE components into the ER network during NEBD. We have previously shown that the ER undergoes a dramatic sheet-to-tubule transformation during mitosis [1]. Thus upon the redistribution of the NE components they have to face an altered morphological environment, as they become a part of the tubular ER network. The precise timing and rapidity of the NEBD and the redistribution of the NE components into the ER network, as well as the structural transformation of the NE, indicate that these fundamental events have to be strictly regulated and controlled in the cells. Our preliminary studies on mammalian cells have indicated that the NEBD is prolonged due to treatment of actin polymerization inhibitors, such as Latrunculin A and Cytochalasin B, indicating that the actin cytoskeleton has a role in the NEBD (Figure 1). Secondly, we are studying the role of the actin cytoskeleton on the reformation of the NE in the late anaphase/telophase. Our results suggest that by knocking down Cofilin1, and thus changing the dynamics of the actin cytoskeleton, the accumulation of the NE components around the chromosomes in the late anaphase/telophase is delayed (Figure 2).

1. M. Puhka et al., J. Cell Biol. **5** (2007) p895.
2. J. Ellenberg et al., J. Cell Biol. **138** (1997) p1193.

3. M. Joensuu and M. Puhka are students of Viikki Graduate School in Molecular Biosciences (VGSB). Research was funded by the Academy of Finland (Project 111 5025).

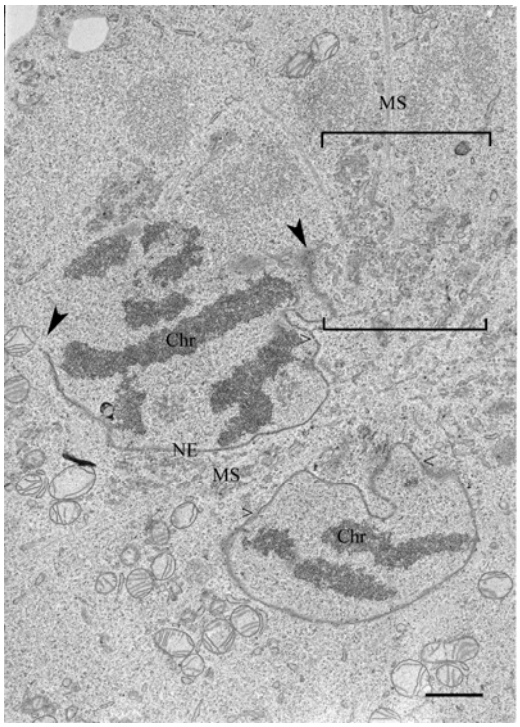

Figure 1. Inhibition of actin depolymerization with Cytochalasin B in cultured mammalian CHO-K1 cells leads to prolongation or prevention of the nuclear envelope breakdown. Actin depolymerized metaphase cells have clear nuclear envelope (NE) structure surrounding fully condensed chromosomes (Chr). Mitotic spindle (MS) and mitotic Golgi fragments (brackets) as well as nuclear pores (open arrows) are indicated. Bar, 1µm.

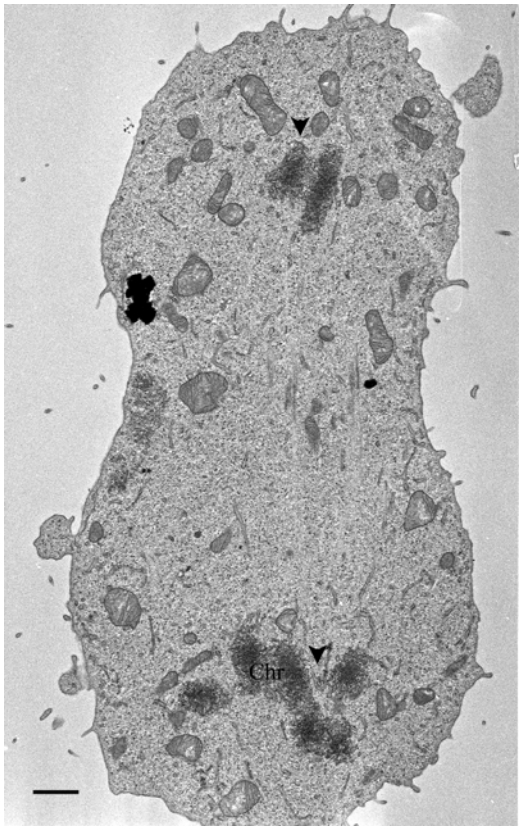

Figure 2. Changing the actin cytoskeleton dynamics in cultured mammalian CHO-K1 cells, by knocking down Cofilin1, the accumulation of the NE components (arrowheads) around the chromosomes in the late anaphase/telophase is delayed. Separated sister chromatids (Chr) are indicated. Bar, 1µm.