Talk

Strategies to improve the robustness of acentrosomal spindle formation in female meiosis

Nazaret Fernández Castillo, Alberto Pineda-Santaella, Ángela Sánchez Gómez, Ana María Brokate, Alfonso Fernández Álvarez

Área de Genética. Departamento de Biología Molecular e Ingeniería Bioquímica

Centro Andaluz de Biología del Desarrollo (CABD), Carretera de Utrera Km 1 41013 Sevilla

Tutor académico: Brokate, Ana M. y Fernández Álvarez, Alfonso

Keywords: meiosis; fission yeast; acentrosomal spindle

ABSTRACT

In meiosis, centrosomes are so important because they organize microtubules and nucleation of the spindle for a correct chromosomal segregation in eukaryotics cells. Human female oocytes lack centrosomes, so microtubules must self-assembly, which can cause mistakes in the process and diseases to the embryo. To study the molecular mechanisms supporting acentrosomal spindle, we are using the fission yeast Saccharomyces pombe as model scenario. In this organism, spindle pole bodies (SPBs), the functional equivalents of centrosomes, are sitting on the nuclear envelope (NE), which is dissasembled in each cell cycle by activating proteins like Sad1 and Bqt1, that mediate chromosome-NE contacts (Pineda-Santaella & Fernández-Álvarez, 2019). Based on these findings, our aims are making the acentrosomal spindle more robust and minimizing chromosomal segregation errors. In order to get then, we want to analyze the effects of overexpression of Cls1p, a cytoplasmic linker associated protein (CLASP) that stabilizes specific groups of MTs in S. pombe and has two homologous proteins in humans, CLASP1 and CLASP2. They contribute to the formation and maintenance of the spindle midzone by promoting MT rescue events (Al-Bassam et al., 2010). On the other hand, there are another important proteins in this process, like Klp6, a kind of kinesin-8, whose homologous proteins in humans are Kif18A, Kif18B, and Kif19. An in vivo study suggets that Klp6 binds to the tubulin triggering the birth of new MTs and promoting nucleation and catastrophe at the growing MT tip (Erent et al., 2012). Deletion or knockdown of Klp6 leads to longer spindles and defects in its assembly and position in many cases (Gergely et al., 2017), but we suggest that a longer acentrosomal spindle could also be stronger and more stable. So, we also pretend to observe the impact of deletion of klp6 on the spindle behavior and chromosome movements. To perform that experiments, we have obtained two different mutants for klp6 and cls1 in a bqt1Δ sad1.2 background by crossing some strains with these characteristics and we are studying what happens in the cell nucleus by fluorescent microscopy, using a DeltaVision microscope. As a result, we expect that chromosomal segregation in mutants for cls1 and klp6 will be more efficient with respect to the mutant control, which has only bqt1Δ sad1.2, and, ultimately, improve the meiotic process in this context.

REFERENCES

Al-Bassam, J., Kim, H., Brouhard, G., van Oijen, A., Harrison, S. C., & Chang, F. (2010). CLASP promotes microtubule rescue by recruiting tubulin dimers to the microtubule. Developmental cell, 19(2), 245-258.

Erent, M., Drummond, D. R., & Cross, R. A. (2012). S. pombe kinesins-8 promote both nucleation and catastrophe of microtubules. PloS one, 7(2).

Gergely, Z. R., Crapo, A., Hough, L. E., McIntosh, J. R., & Betterton, M. D. (2017). Kinesin-8 effects on mitotic microtubule dynamics contribute to spindle function in fission yeast. Molecular biology of the cell, 27(22), 3490-3514.

Pineda-Santaella, A., & Fernández-Álvarez, A. (2019). Spindle assembly without spindle pole body insertion into the nuclear envelope in fission yeast meiosis. Chromosoma, 128(3), 267-277.