Regulatory processes of cell division

Cell division is a process that requires a lot of regulation.

One such regulatory element is Cyclin A2. It is one of the two cyclin A isoforms, the other being cyclin A1 which is largely expressed in male testes with roles in male gamete formation. Cyclin A2's role in female meiosis is the main focus in Zhang et al.'s research article, " Cyclin A2 modulates kinetochore-microtubule attachment in meiosis II." [1] The role of cyclin A2 in female meiosis has largely remained unexplored due to the fact the knock-out (genetic deletion) in animal models of the gene expressing cyclin A2 results in peri-implantation lethality [1] . Peri-implantation refers to the time between a free blastocyst in the uterus to implantation of embryo to the uterine walls [2] .

Cyclin A2's role in mitosis (somatic cell division, as depicted in Figure 1) involves regulating cell cycle progression during S phase by binding CDK2 and G_2 phase by binding CDK1; both of which are CDK kinases and have roles in regulating transcription and mRNA processing [1] . Cyclin A2 is first synthesised on the onset of the S phase of cell division and localised in the nucleus before being destroyed by Anaphase-Promoting Complex (APC/C) and cleared before the cells reach metaphase of mitosis [1] . If not, it will lead to disrupted microtubule-kinetochore interactions which makes it difficult for the cell to progress in anaphase; as the chromosomes will lag in separation [1] .

With the development of using anti-cyclin A2 antibodies, the role of cyclin A2 was finally able to be investigated in female mice oocytes [3] . At different

points of meiosis, oocytes were injected with anti-cyclin A2 antibodies to inhibit the actions of cyclin A2 in a study conducted by Winston et al [3] . Results from this study showed inhibition of G2-M phase transition, and failure of sister chromatids to separate in meiosis II [3] . It seemed that Winston's team's results were promising for Zhang et al. as they were able to develop their study from the team's work and conclude from their own data that cyclin A2 played an essential role in the second stage of meiosis and in the oocyte to embryo transition [1] .

Contrast to figure 1's depiction of mitosis in the M phase of cell division, meiosis has a longer cell division process that is not limited to only prophase, metaphase, anaphase and telophase. As gametes only require half the amount of genetic material as normal somatic cells do, meiosis cell division requires two divisions, rather than the single division that mitosis does. As depicted in figure 2, the first division ensures homologous chromosomes are split into two different cells. The second division is to ensure chromosomes with two sister chromatids are split into chromosomes with just the single chromatid.

One of the first discoveries discussed in the paper noted the necessity of cyclin A2 for normal meiosis II [1] . This was achieved by comparing cyclin A2 depleted cells and normal cells in the meiotic phase of ovulated oocytes. Using immunofluorescence and labelling techniques, the team was able to visualise the oocytes. Errors in spindle formation in cyclin A2 depleted cells were noted to be higher that that of the control group [1] . Cyclin A2 mRNA was injected back into the experimental group and decreased spindle

formation abnormalities were noted. To truly understand the role of cyclin A2 in meiosis II, the team used time-lapsing confocal imaging to visualise and compare the control and experimental group. They found that the experimental group showed lagging chromosomes during anaphase. Furthermore, cyclin A2 was also found to be involved in the transition of an “ immature” MII spindle to “ mature” spindles. The team were able to examine the differences in kinetochore-microtubule attachments in the meiosis II spindle and found that the cyclin A2 absent group showed more “ immature” spindles and slower progression to “ mature” spindles. Thus, the team were able to conclude cyclin A2’s role in spindle and chromosome organisation during meiosis II [1] .

The authors were able to eliminate the possibility of spindle formation error arising in Meiosis I by comparing the timing of germinal vesicle breakdown (GVBD) and polar body formations between experimental and control groups [1] . Germinal Vesicle Breakdown marks the end of the prophase of meiosis I, when oocyte development was previously paused. The team found that there were no differences in the timing of GVBD, polar body formation and entry into meiosis II. To further confirm the hypothesis that no spindle formation error was present in Meiosis I, the team found that no aneuploidy (abnormal chromosome numbers) was present in meiosis-II oocytes in the experimental group.

Whereas the Winston’s team used antibodies to inhibit acutely inhibit cyclin A2 activity, Zhang’s team used conditional knock-out mice. The system they used was the ZP3-Cre-LoxP system [4] . This is a conditional knock out system,

which is used to delete target genes in a specific organ/tissue at a specific development stage [4] . This is used because conventional knockout of the cyclin A2 gene results in lethality [1] . The team sourced their cyclin A2 gene (flanked by LoxP) carrying mice from the University of Newcastle in New South Wales, which were crossed with Zp3-Cre transgenic mice. This was done to generate cyclin A2 deficient oocytes. To ensure experimental oocytes had successfully had cyclin A2 removed, Western blotting was performed, which detects for specific protein. To visualise the oocytes for comparison between control and experimental group, immunofluorescence imaging and photoactivated fluorescence imaging was used.

This study indeed has wider implications to female infertility and genetic diseases present. Genetic disorder caused by chromosomal abnormalities are growing to become more common with the rising trend to delayed childbirth. Advanced maternal age is associated with increased likelihood of genetic abnormalities in female gametes. Although there are some cases of viable aneuploidy conditions, such as Down syndrome and sex chromosomes aneuploidy, most embryos with aneuploidy conditions end in spontaneous abortions. Although no therapeutic approach was explored in this study, the results can definitely aid in furthering the understanding of female infertility and chromosomal aneuploidy disorders, and perhaps therapies can be developed in the future to prevent cases of infertility and genetic disorders. This could likely revolutionise the IVF industry.

References

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