Fertile Research

Egg and sperm cells are highly sensitive during their development. When, for example, there is an error in the way the genetic material is divided between the individual gametes, the resulting embryo will often either be nonviable or suffer from severe birth defects. **Melina Schuh** from the **Max Planck Institute for Biophysical Chemistry** in Göttingen wants to find out why egg maturation is so error-prone. The results of her research could one day help couples who are unable to have children.

TEXT **CATARINA PIETSCHMANN**

At long last, a warm, dry summer evening in historic downtown Göttingen. Out on the terrace of the restaurant where we arranged to meet, Melina Schuh is waiting for us, already studying the menu. She has the night off from the kids, with her husband on childcare duty. She puts her phone where she can see it, "Just in case there are any problems at home," she says with a smile.

Schuh's professional interest is in a little understood process without which none of us would be here: meiosis. Whether fruit flies or humans – without meiosis, sexual reproduction would be impossible. While other cells in our bodies have two sets of chromosomes, egg cells and sperm must have only one. "Otherwise, when the egg and sperm cells fuse, the number of chromosomes would double – instead of two sets of chromosomes, the new cell would have four," explains Schuh. "That's why, during meiosis, one of the two sets of chromosomes is eliminated from the egg cell."

This first meiotic cell division occurs while the egg cell is still in the ovary. Unlike frogs, mice and most other animals, in humans, meiosis sometimes goes wrong. As a result, the desire to start a family remains unfulfilled, with miscarriages or an embryo that contains too many or too few chromosomes.

HALF A SET OF CHROMOSOMES

In meiosis, before cell division takes place, the cell doubles the amount of its genetic material. The meiotic spindle then arranges the chromosomes of each pair in such a way that they lie opposite one another in the egg cell. The protein fibers in the meiotic spindle then each pull one of the chromosomes in each pair to the spindle poles situated at opposite ends of the egg cell. At the end of this process, the two sets of chromosomes have been separated. In the second meiotic division, the same process is used to separate the copies that were produced when the amount of genetic material was doubled. In males, this results in the production of four sperm cells. In females, in contrast, only one egg cell is produced, along with two tiny polar bodies that contain the extra chromosomes.

Errors can occur during meiotic spindle formation, and also during chromosome separation. "If, for example, the egg cell ends up with two copies of chromosome 21, fertilization produces an embryo with trisomy 21, or Down syndrome," explains Schuh. Other chromosome number abnormalities also impact embryonic development. "In most cases, an embryo with an abnormal number of chromosomes will have such severe defects that it won't even be able to implant into the uterus. Often, women might not even be aware that fertilization has taken place."

An embryo with too many X chromosomes, however, is viable. "It's estimated that around one in a thousand women have three X chromosomes instead of the normal two, and they are usually unaware of it." So how do these errors – which become increasingly common as women enter their late 30s

Photo: Frank Vinken

At the moment, Melina Schuh's workplace is still a construction site: In late 2017, her department will move into the new premises designed especially for the study of live egg cells. The new site will allow them to accommodate high-resolution microscopes with which living, developing egg cells can be observed for long periods.

and early 40s – occur? At present, we really have a very limited understanding of the underlying mechanisms. Melina Schuh wants to change this.

In mammals, egg cells are produced early in embryonic development. These very small cells are enclosed in a thin layer of somatic cells, known as follicle cells, and are already present in the ovaries at birth. They remain inactive until puberty, after which two to three cells mature during each menstrual cycle. The follicle cells are connected with the immature egg and feed it via fine channels. The follicle cells divide and form a cocoon around the egg cell. Of the initially two to three follicles – that is, the eggs cells with the surrounding follicle cells – only one reaches full size, in the middle of the menstrual cycle. The others die off.

At this point, the pituitary gland secretes a hormone that initiates ovulation. Meiosis commences, and the follicle cell layer on the outside begins to loosen. Once chromosome segregation is complete, the egg cell slides out of its cocoon, exits the ovary and moves into the fallopian tube, ready for fertilization by a sperm.

"When a 40-year-old woman wants to get pregnant, both her egg cells and the chromosomes inside them are also 40 years old," says Melina Schuh. Chromosome pairs are held together by ring-shaped protein complexes, which act like a kind of glue. Normally, enzymes ensure that the protein complexes that hold the chromosome pairs together aren't severed until meiosis has commenced. "We have discovered, however, that chromosome pairs aren't as well bound together starting as early as age 25. Some chromosome pairs literally fall apart." Under the microscope, we observe that this results in chromosomes that are incompletely paired, chromosomes that turn during separation, or premature separation of chromosome pairs. "This means that the spindle fibers can't grasp and separate the chromosomes properly." As a result, depending on age, between 10 and 50 percent of egg cells are no longer viable.

GOOD EGGS AND BAD EGGS

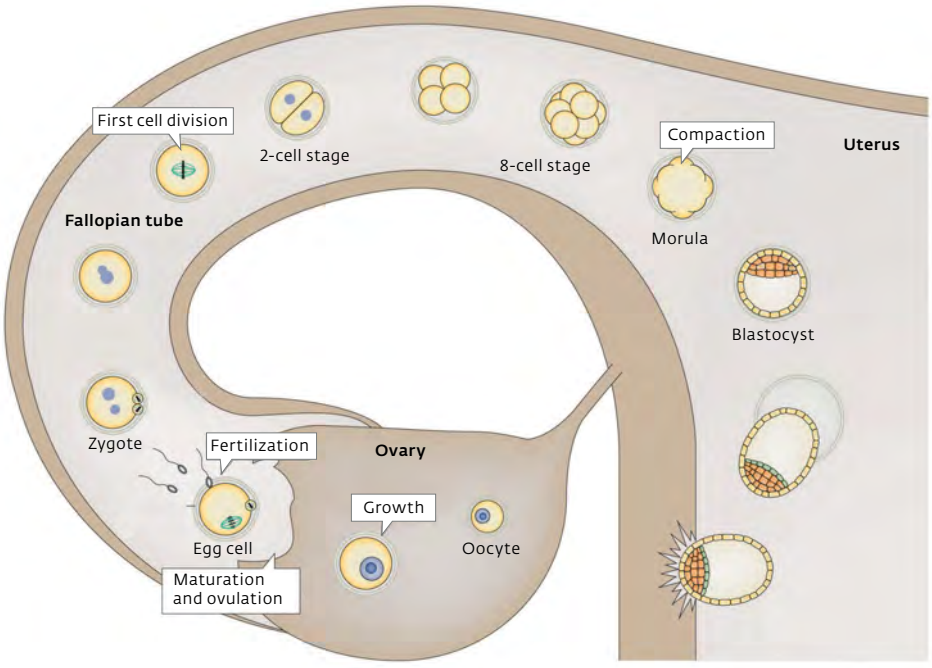
The drop in quality of immature egg cells is one of the reasons why the risk of chromosome anomalies and miscarriages increases with age. "But the good news is that, even if some egg cells are no longer good, other eggs may still be fine," notes Schuh. So, if nothing has happened after the first few months, don't give up! A good egg might come along with the very next ovulation.

Before she came to Göttingen, Melina Schuh worked at the MRC Laboratory of Molecular Biology (LMB) in Cambridge, England. There, she mostly worked on mouse egg cells, but she also conducted some research on human eggs. Even after her move to Göttingen, her laboratory maintains close links with Cambridge. She has a small laboratory at the Bourn Hall Fertility Clinic, where she researches how human egg cells develop. Bourn Hall Fertility Clinic is no ordinary clinic: it was the world's first in-vitro fertilization (IVF) clinic and was founded by IVF pioneers Sir Robert Edwards and Patrick Steptoe. In 1978, their pioneering work led to the birth of the very first "test tube baby," Louise Brown, paving the way for millions more IVF babies.

"When you collect egg cells for in-vitro fertilization, they are generally at different stages of development. If a cell hasn't yet undergone meiotic division, it can't be used for IVF. In that case, if the patient consents, we can use these extra, unfertilized cells – which would otherwise be discarded – for research."

Schuh comes from Bad Pyrmont, where she grew up with three younger siblings. As a child, she did a lot of sports, including swimming, athletics, volleyball and badminton. She also had

Photo: Frank Vinken



Left Katarina Harasimov (left) and Melina Schuh use a special microscope to inject RNA molecules into egg cells. This enables the researchers to visualize specific structures within the cells.

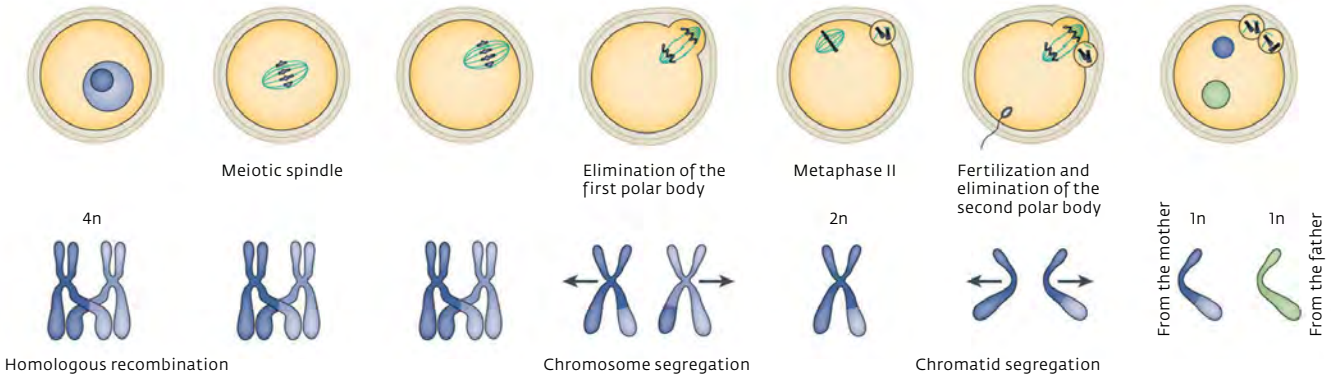
Right During human embryonic development, immature egg cells (oocytes) are stored in the ovaries. These egg cells are activated during puberty. In the middle of the menstrual cycle, one egg cell matures by undergoing the first meiotic division. It is then released into the fallopian tubes, ready to be fertilized. If fertilization takes place, it divides until it forms a compact ball of 32 cells known as a morula. After further cell division, the morula forms a fluid-filled cavity. At this stage, the embryo, now called a blastocyst, sheds its protein shell and implants into the uterus.

diverse musical interests: she sang in a choir and played several instruments, including piano and flute.

She majored in German and biology in high school before going to Bayreuth to study biochemistry. "Every time I walked out of a lecture, I felt like I understood a little more about the world around me." For her dissertation, she studied cell division in fruit fly embryos, completed her degree with distinction and then started her PhD at the European Molecular Biology Laboratory (EMBL) in Heidelberg. Jan Ellenberg, a specialist in cell division and microscopy, offered her the opportunity to work with starfish egg cells. She spent the first summer of her doctorate at the renowned Marine Biological Laboratory in Woods Hole, Massachusetts, which has been conducting research on reproduction in marine animals for more than a century.

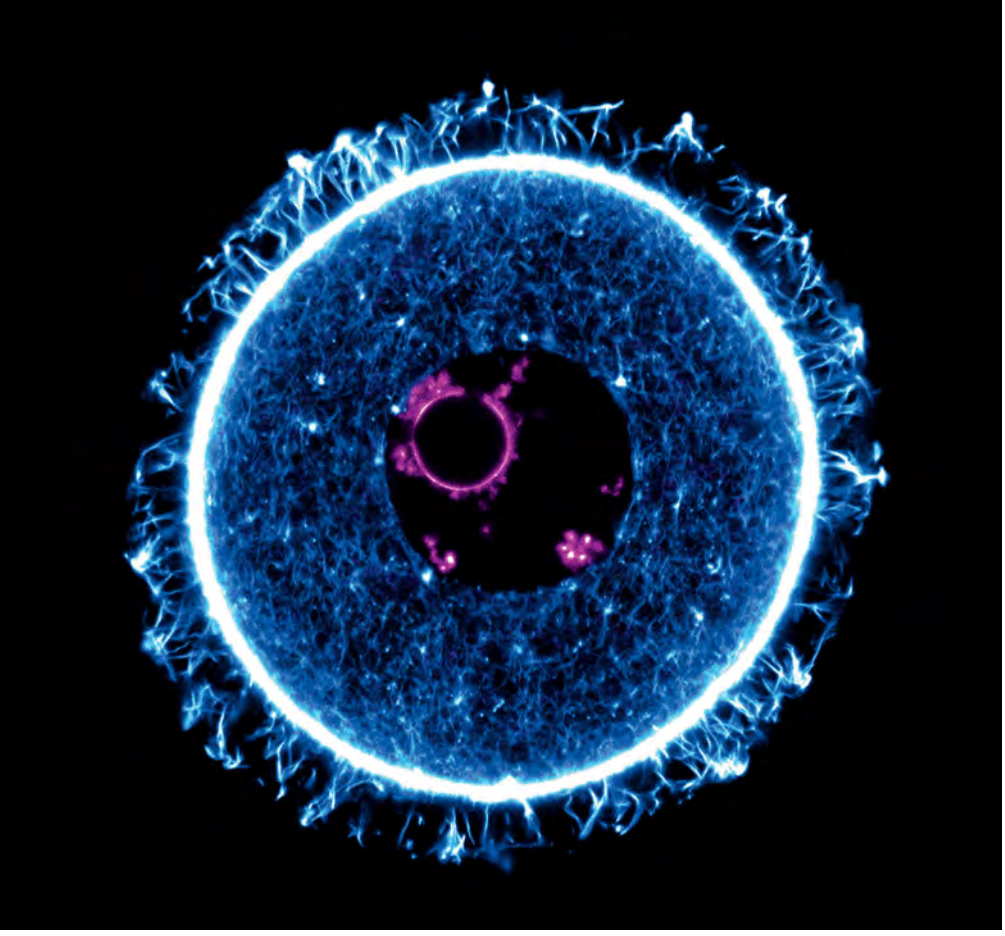
"Starfish egg cells are relatively large, and they can be harvested in fairly large quantities, making them great for use in biochemical experiments. Starfish eggs are also transparent, so they are easy to observe under the microscope." But what Melina was really interested in were the processes happening in human egg cells, so in Heidelberg, she started to develop techniques for high-resolution microscopy of mouse egg cells, which are much more similar to human egg cells.

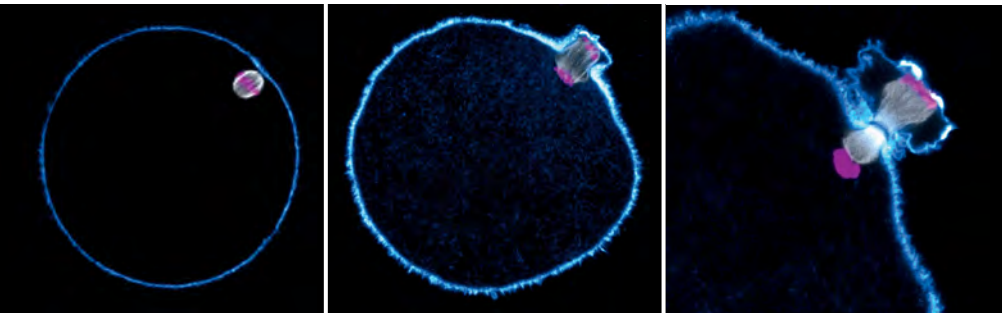
It was while writing her doctorate that she received an inquiry from Cambridge. She was encouraged to apply for a group leader position at the LMB. "I had just a week to put together a research plan," Melina Schuh re-



The phases of egg cell maturation: After the nuclear envelope has broken down, the newly formed meiotic spindle migrates to the surface of the cell and segregates the (previously doubled) chromosome pairs. After forming a second spindle, the cell remains in metaphase II until fertilization. If a sperm cell fuses with the egg cell, the spindle segregates the two copies of each chromosome. This second step thus halves the number of chromosomes. The extra chromosomes are eliminated from the cell into polar bodies. The fertilized egg cell (zygote) now contains two pronuclei, each containing a single set of chromosomes from either the mother (blue circle) or father (green).

Graphic: Nature Reviews, Molecular Cell Biology Vol.14, Sept. 2013





Above Mouse egg cells in prophase. Viewed under a fluorescence microscope, the network of actin fibers glows blue and the chromosomes in the interior of the cell nucleus (black circle) glow magenta. In this phase of meiosis, the chromosomes condense and the spindle forms.

Below Human egg cells during the first meiotic division. During the metaphase, the spindle (gray: microtubules; magenta: chromosomes) is anchored to the cell surface (left; blue: actin fibers). The chromosomes are separated from each other (center). One half remains in the egg cell while the other is eliminated from it. The first polar body is formed (right).

members. But the timing was perfect: "My husband had just successfully applied to the MBA program at the London Business School, so we decided he would go to London and I would go to Cambridge." In the beginning, they did a lot of commuting, then they bought and renovated a house in Cambridge. Their son was born just as the house was finished, followed two years later by a daughter.

"Cambridge is a small university city, perfect for bringing up children. There are lots of green spaces between the colleges, great playgrounds, cows right in the middle of the city – it's very idyllic."

Distances are short and most journeys can be made by bicycle – with the kids in a bike trailer. In England, childcare and working hours are designed to meet the needs of two working parents. The only time things got a little tricky was if she had to go to a conference and her husband, a management consultant, was away on business. When that happened, Melina's parents came over from Germany to spend some time with their grandchildren.

Nevertheless, when she received an offer from the Max Planck Society to become Director at the Max Planck Institute for Biophysical Chemistry in Göttingen, Melina didn't have to think twice. "We have the perfect conditions for our research here, and it's close to my hometown!"

Does she miss anything about England? "Friends of course, and sometimes the British politeness. Shops open on Sundays. And healthy convenience foods!" she adds with a laugh. "All in all, I could have stayed there forever." But in Cambridge she had only a small research group. "Eventually, our ideas outgrew our capabilities. It's great that I can now pursue longer-term and higher-risk projects at the Max Planck Society."

Research, work trips, family, even meeting journalists like this – it all has to be organized. Since early 2016, the 37-year-old biochemist has been head of the Meiosis Department at the Max Planck Institute in Göttingen. She now has a secretary and lab manager to take care of most of the organization and planning – and for that she is very grateful: "When you have young kids, there are never enough hours in the day. I'd much rather spend my time at the Institute doing actual research, rather than administration."

Seven years as a researcher in Cambridge have also left their mark on her research style. "In Cambridge I experienced a largely non-hierarchical, friendly and collaborative way of working. I try to put a lot of what I learned at the EMBL and subsequently in Cambridge into practice here in my department." She now has a group of 15 PhD students and postdocs, "a good number," she says, for people to be able to bounce ideas off each other and develop a vision. "I want to give my staff as much freedom as possible and support them in tackling ambitious research projects."

Photos: Zuzana Holubcova and Melina Schuh, MRC Laboratory of Molecular Biology

As very few human egg cells are available for research, the researchers are in need of other model systems. Mouse egg cells, however, in addition to being smaller than human egg cells, also have a meiotic spindle with a different structure. In her search for suitable alternatives that more closely resemble the processes happening in human eggs, Melina Schuh stumbled upon pigs. Pig egg cells are easy to procure from slaughterhouses and they are easily isolated.

NEW SPINDLE PROTEIN DISCOVERED

Her team recently succeeded in showing that actin, a protein that, among other things, plays a crucial role in the shape and migration of cells, is present in the spindle in mice, sheep, pigs and humans. Without this protein, chromosome segregation in mammalian egg cells runs into problems. "If there's not enough actin, the chromosomes don't align correctly in the center of the cell prior to cell division. In addition, chromosome segregation is often slower, and the egg cell ends up with too many or too few chromosomes," explains Schuh. Actin is therefore thought to be required for spindle fiber assembly.

Why meiosis is less error-prone in other species – such as mice – remains a mystery. "You can argue, from an evolutionary point of view, that having lots of offspring is important for the survival of mice as a species." Mice are able to produce a litter of five to ten pups every few weeks. "If, say, one quarter of the embryos failed to develop, it would be an enormous waste of resources." Humans have always had relatively few offspring. "In humans, even if a quarter of egg cells are defective, this may not have much of a negative effect on reproduction. Most defective embryos are unable to implant into the uterus and the mother can become pregnant during her next menstrual cycle. Perhaps egg cells in humans just don't need to be as reliable as in other species," suggests Schuh.

Understanding all the details of meiosis, finding ways of distinguishing between egg cells with the right and wrong number of chromosomes before they are fertilized, and perhaps even helping them develop properly is a big goal. Melina Schuh's research could also help generate insights to improve fertility treatments. Her research thus also has implications beyond pure basic research. By the time Schuh first became pregnant, she already knew plenty about the subject and was well aware of the potential risks. "Before my first child was born, I could hardly believe that everything could turn out okay – there are so many developmental stages that have to go right before a child is born." Today, Melina and her husband have three healthy children.

Without doubt, Melina Schuh's life is very busy and there are hardly enough hours in a day. That there are not enough women in leadership positions in science is a recurring complaint. "There are programs for the advancement of women, quotas for women and many other initiatives, but the sticking point is still the compatibility of a research career with family life," stresses Schuh.

Particularly in research, it is very difficult for women and men to stay at home for long after having a child. Science today advances so quickly that a year of parental leave can mean that projects have already become outdated. There are very few childcare facilities with enough staff for children under the age of one. "It's never easy leaving your children in the care of others, so it's very important to know that they're happy and they're going to be well looked after. In my view, high-quality childcare options are important for combining research work with family life." The fact that the nursery on the Max Planck campus is open to children of ages 6 months and older – something that is normal in England – is due in part to her committed efforts.

A good 30 years of research lie ahead of Melina. Was it helpful to be made Director at such a young age? "Definitely. It means you can set yourself really big goals. I'm very much looking forward to seeing what the future has in store!"

Research and family? For Melina Schuh at least, it works well. In principle, it's just like meiosis: it's all a question of organization. ◂

GLOSSARY

Set of chromosomes: With the exception of the gametes, the cells of most vertebrates generally have two sets of chromosomes – that is, they contain two copies of each chromosome. Human embryos that have the wrong number of chromosomes usually die before implantation in the uterus. Only rarely are such embryos viable, for example embryos with three copies of chromosome 21, which results in Down syndrome.

Spindle: During cell division, this ensures that the chromosomes are arranged in pairs in the equatorial plane and are pulled away from each other in opposite directions. The spindle is constructed from protein fibers known as microtubules. These consist of an array of tubulin proteins and attach to a special protein complex on the chromosome, called kinetochore. Recent research has shown that, in addition to microtubule and kinetochore proteins, correct chromosome segregation by the spindle also requires actin. This protein also forms long fibers and enables muscle cells to contract, for example.