Chapter 8
Control of Oocyte Growth and Development by Intercellular Communication Within the Follicular Niche

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Abstract In the mammalian ovary, each oocyte grows and develops within its own structural and developmental niche—the follicle. Together with the female germ cell in the follicle are somatic granulosa cells, specialized companion cells that surround the oocyte and provide support to it, and an outer layer of thecal cells that serve crucial roles including steroid synthesis. These follicular compartments function as a single physiological unit whose purpose is to produce a healthy egg, which upon ovulation can be fertilized and give rise to a healthy embryo, thus enabling the female germ cell to fulfill its reproductive potential. Beginning from the initial stage of follicle formation and until terminal differentiation at ovulation, oocyte and follicle growth depend absolutely on cooperation between the different cellular compartments. This cooperation synchronizes the initiation of oocyte growth with follicle activation. During growth, it enables metabolic support for the follicle-enclosed oocyte and allows the follicle to fulfill its steroidogenic potential. Near the end of the growth period, intra-follicular interactions prevent the precocious meiotic resumption of the oocyte and ensure its nuclear differenti-

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ation. Finally, cooperation enables the events of ovulation, including meiotic maturation of the oocyte and expansion of the cumulus granulosa cells. In this chapter, we discuss the cellular interactions that enable the growing follicle to produce a healthy oocyte, focusing on the communication between the germ cell and the surrounding granulosa cells.

8.1 Introduction

Each human female is born with a population of oocytes estimated to range from several hundred thousand to several million (Wallace and Kelsey 2010). Although this may seem like a large number, it pales in comparison to the millions of sperm produced daily by a young adult male (Oatley and Brinster 2012). Moreover, although some controversy remains, the weight of available evidence indicates that, unlike males, female mammals do not possess a population of germ-line stem cells that give rise to new oocytes postnatally (Eggan et al. 2006; Lei and Spradling 2013; Zhang et al. 2014a; Zhang et al. 2015). Oocytes are thus relatively rare, and each presents a precious reproductive opportunity. It is not surprising that each has been allocated its own structural and developmental niche—a follicle. Each follicle contains a single oocyte together with somatic granulosa cells, which are specialized companion cells that surround the oocyte and provide support to it as well as producing steroid hormones, and an outer layer of thecal cells that serve crucial roles in steroid synthesis and in ovulation. The follicular compartment functions as a single physiological unit whose purpose is to produce a healthy egg that upon ovulation can be fertilized and will give rise to a new organism, thus enabling the female germ cell to fulfill its reproductive potential. Here, based mainly on findings using the murine model, we will discuss the cellular interactions that enable the follicle to achieve this goal, focusing on the communication between the germ and somatic cells,

8.2 Overview of Oogenesis and Folliculogenesis

During embryonic development in the female, oogonia proliferate to generate a population of germ cells. These synchronously enter meiosis and progress to diplotene of prophase I where they become arrested (Borum 1961). By the time of birth, somatic pre-granulosa cells within the ovary have surrounded oocytes to generate primordial follicles, each consisting of one oocyte (~15 μm diameter in the mouse) enclosed by a single layer of squamous granulosa cells (Findlay et al. 2015; Grive and Freiman 2015; Jorgensen 2013; Pepling 2012). However, for reasons not yet fully understood, a significant proportion of the oocyte population is lost by apoptosis during late embryogenesis (Ene et al. 2013; Hutt 2015; Malki et al. 2014),

reducing the number of primordial follicles that form or remain at birth. Primordial follicles thus constitute the ovarian reserve pool, from which follicles are continually recruited during reproductive life into the growth phase to produce fertilizable eggs. Recruitment progressively depletes the ovarian reserve and when, as a consequence of aging, disease, or environmental toxins, the supply of primordial follicles drops below a functional threshold, the female is no longer fertile.

When a primordial follicle enters the growth phase, the enclosed oocyte begins to increase in size. The granulosa cells undergo a change in shape and proliferate mitotically so that they continue to fully enclose the surface of the expanding oocyte. Such follicles, consisting of a newly growing oocyte surrounded by a single layer of granulosa cells, are termed primary. During this stage, the oocyte begins to secrete a family of glycoproteins that assemble into an extracellular coat, termed the *zona pellucida*, which physically separates the oocyte from the bodies of the adjacent granulosa cells. As the granulosa cells continue to proliferate, they generate multiple layers around the growing oocyte. In addition, thecal cells become deposited around the outside of the basement membrane surrounding the follicle. These follicles, now termed secondary, also begin to acquire their own vasculature (Suzuki et al. 1998). Eventually, a fluid-filled cavity termed the antrum appears within the follicle, defining it as antral. Once the antrum expands, it separates the granulosa cells into two populations—the mural granulosa cells that line the inside of the follicular wall and the cumulus granulosa cells that surround the oocyte (Fig. 8.1). Near the time of antral formation, the oocyte, having increased >100-fold in volume and accumulated enormous reserves of transcripts, proteins, and organelles needed to sustain early embryo development, stops increasing in size. The oocyte nonetheless continues to undergo molecular differentiation and it is only at the late antral stage that it acquires the ability—known as developmental competence—to develop as an embryo following ovulation and fertilization.

The final stage of oocyte development is meiotic maturation. Maturation is physiologically triggered by the preovulatory surge of luteinizing hormone (LH) that also triggers cumulus expansion and ovulation, thus coordinating the completion of oocyte development with its deposition into the reproductive tract to be fertilized (Eppig 1979; Espey 1980; Meinecke and Meinecke-Tillmann 1979). During maturation, the cell cycle, which has been arrested at late prophase I throughout growth, resumes and in most mammals, the oocyte completes meiosis I before becoming arrested at metaphase of meiosis II (Fig. 8.1). In addition to these nuclear events, many cytoplasmic changes also occur during maturation. These include changes in the ultrastructure of the endoplasmic reticulum (Mehlmann et al. 1995; Payne and Schatten 2003), migration of the cortical granules to just underneath the plasma membrane (Liu et al. 2003; Nicosia et al. 1977), and accumulation of the mitochondria around the meiotic spindle (Dalton and Carroll 2013; Van Blerkom et al. 2003). Many previously silent mRNAs become translationally activated, whereas others become silenced and in some cases

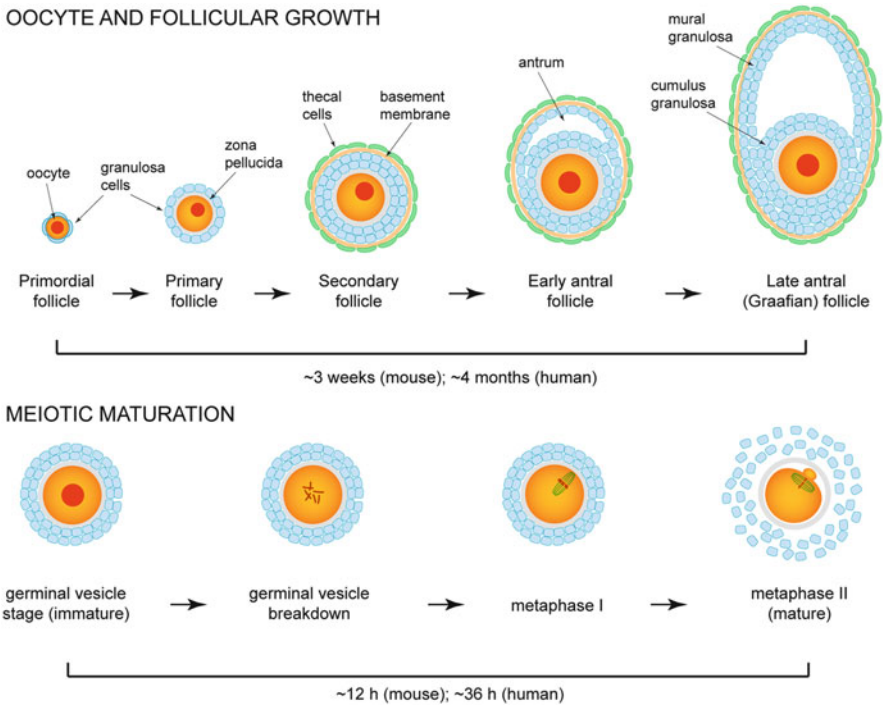

Fig. 8.1 Postnatal oocyte development. *Upper*: Oocyte and follicular growth. Oocytes in primordial follicles are surrounded by a small number of squamous granulosa cells. Incompletely understood signals cause primordial follicles to transition to the primary stage, characterized by a growing oocyte enclosed by cuboidal, mitotically active granulosa cells. When multiple granulosa cell layers have been generated and a basement membrane and thecal cell layer have been recruited, the follicles are termed secondary. The appearance of a small cavity, termed the antrum, within the granulosa cell layer defines antral follicles; oocytes at this stage are (nearly) fully grown. As the antrum enlarges, the mural and cumulus granulosa cell populations become clearly separated, and the fully grown Graafian follicle is ready to ovulate. *Lower*: Meiotic maturation. Upon the initiation of maturation, the nucleus (germinal vesicle) breaks down and the chromosomes condense and migrate to the periphery of the cell where they become assembled on the first meiotic spindle. Half of the products of meiosis I are discarded in the first polar body, and the oocyte chromosomes are assembled on the second meiotic spindle in preparation for meiosis II, which will occur after fertilization. The maturing oocyte also undergoes many cytoplasmic changes, not illustrated. During maturation, in a process termed cumulus expansion, the cumulus cells secrete proteins that generate an extracellular matrix that separates them from each other and from the oocyte and facilitates fertilization

degraded (Clarke 2012; Kang and Han 2011). During maturation, physical contact between the oocyte and granulosa cells, which has persisted since primordial follicle formation, is irreversibly lost. By the end of maturation, the mature germ cell, now termed an egg, is ovulated and can be fertilized.

8.3 Intra-follicular Cell Contact and Bidirectional Communication

The ovarian follicle is among the few physiological structures in mammals whose development can be sustained for a relatively long period of time in vitro and for which the quality of the cells that grow or differentiate in vitro can be rigorously tested—in this case, by the ability of the oocyte to give rise to an embryo. Thus, the ovarian follicle is an ideal model system to study mechanisms that regulate the formation and function of complex tissues. The relatively simple structure and cellular composition of the ovarian follicle, together with the development of culture conditions that enable oocyte and follicular growth to be reproduced in vitro, have provided researchers with an unparalleled opportunity to discover how the different cell types within the follicle cooperate to produce a healthy egg. We will focus on postnatal growth and development of the oocyte and follicle. We refer readers to recent reviews for studies of pre- and perinatal stages (Findlay et al. 2015; Grive and Freiman 2015; Jorgensen 2013; Pepling 2012).

Experimental studies have revealed a dynamic bidirectional communication network that links the germ-line and somatic cells of the follicular niche and is governed by both direct cell–cell contact and paracrine signaling. This communication is absolutely indispensable for the production of a healthy oocyte. When granulosa–oocyte complexes at an early stage of growth are removed from the ovarian follicle and placed in culture, the oocyte can complete normal development, as witnessed by its ability to give rise to offspring following fertilization (Eppig and Schroeder 1989; Hasegawa et al. 2006; Yamamoto et al. 1999). This remarkable result demonstrates that, with the possible exception of the earliest stages of growth, which have not been tested, the granulosa cells alone are sufficient to provide all of the external support needed by the oocyte to acquire developmental competence. If the granulosa cells are removed, however, the oocyte fails to grow, even when the two cell types are cocultured. Thus, physical contact with the granulosa is essential for oocyte growth.

Because the *zona pellucida* becomes assembled around the oocyte at an early stage of growth, thereby physically separating it from the bodies of the granulosa cells, an adaptation is required to allow the two cellular compartments to remain in physical contact. This is achieved by structures termed transzonal projections (TZPs). These narrow cytoplasmic fingers extend from the granulosa cells to the oocyte plasma membrane. TZPs link the surrounding follicle cells to the oocyte in most, and possibly all, mammalian species (Anderson and Albertini 1976; Hertig and Adams 1967), and similar structures have been observed in numerous nonmammalian species (Kessel et al. 1985; Schroeder 1981). Most TZPs contain an actin backbone, but a small fraction contains microtubules instead (Albertini and Rider 1994; De Smedt and Szollosi 1991). At the tips of the TZPs, where they contact the oocyte plasma membrane, are located gap junctions (which may also link TZPs to each other) (Anderson and Albertini 1976; Albertini and Rider 1994). These intercellular channels, composed of proteins known as connexins, permit the

passage of molecules up to about 1 kDa in size (Kidder and Mhawi 2002). Connexin37 is the principal gap junction component in oocytes, and deletion of its encoding gene, *Gja4*, completely blocks gap junctional communication between the two cell types. As a result, oocytes of *Gja4*$^{-/-}$ mice fail to complete growth and do not become competent to develop as embryos (Simon et al. 1997). Thus, oocyte development depends crucially on gap junctional communication with the surrounding follicular granulosa cells.

On the other hand, oocyte-derived paracrine factors (ODPFs) regulate a wide range of granulosa cell activities (Emori and Sugiura 2014). For instance, in the absence of the growth-differentiation factor 9 (GDF9), a member of the transforming growth factor β (TGF β) family provided by the oocyte, the granulosa cells exhibit numerous abnormalities including a failure to proliferate to generate multilayered secondary follicles (Carabatsos et al. 1998; Dong et al. 1996). They also are unable to support normal oocyte development. These observations collectively underscore the essential nature of the bidirectional interactions between the somatic and germ-line compartments during folliculogenesis. By coordinating oocyte and follicular development from the time of growth initiation until the final stages of development, this communication network ensures that ovulation releases a developmentally competent egg. In the following, we discuss how these bidirectional interactions govern the various stages of follicle development.

8.4 Initiation of Oocyte Growth

At the time of primordial follicle formation, the association of the small oocytes with the somatic granulosa cells protects them from undergoing cell death (Pepling and Spradling 2001; Pesce et al. 1997; Rajah et al. 1992). Thus, interactions between the germ-line and somatic compartments of the follicle are indispensable even at the earliest stages of folliculogenesis, and indeed presage an ongoing interaction that is essential to yield a fertilizable egg. The first indication that a primordial follicle has been activated to begin growth is a morphological transition of the granulosa cells from a squamous to cuboidal morphology (Braw-Tal 2002; Hirshfield 1991). The external signals (if they exist) that initiate this transition remain unknown; however, it is associated with the increase in mitotic activity of the cells that is essential to enable them to fully enclose the growing oocyte (Da Silva-Buttkus et al. 2008). This morphological transition is apparently not indispensable for the initiation of oocyte growth though, because the oocytes of mice lacking forkhead box protein L2 (*Foxl2*) begin to grow even though the granulosa cells remain squamous (Schmidt et al. 2004; Uda et al. 2004). These oocytes grow only modestly before becoming arrested, however, suggesting that maintaining oocyte growth depends on this transition and/or continued proliferation of the granulosa cells.

The early morphological change observed in the granulosa cells of newly growing follicles suggests that follicular activation may initiate in this cellular

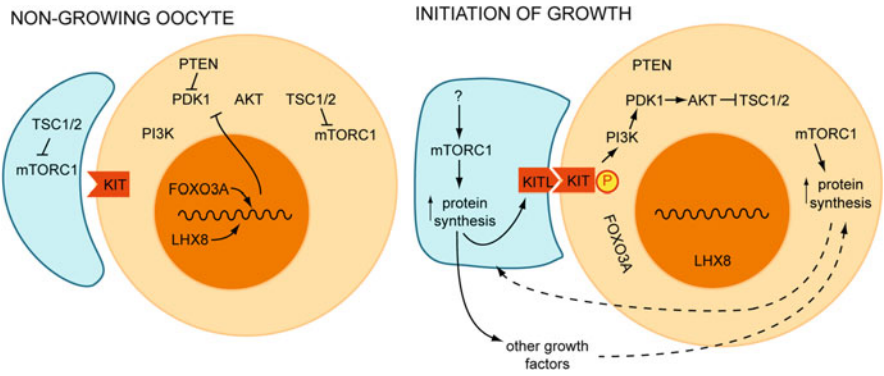

Fig. 8.2 Oocyte–granulosa communication and the initiation of growth. In a primordial follicle, TSC1/2 are active in granulosa cells, inhibiting mTORC1 activity and therefore limiting protein synthesis. In the oocyte, the transcription factors, FOXO3A and LHX8, prevent the oocytes from growing, possibly by inhibiting PI3-kinase activity. Signals that increase mTORC1 activity and therefore protein synthesis in granulosa cells are associated with a squamous-to-cuboidal morphological transition, mitotic proliferation, and increased production of KITL. This activates KIT on the oocyte plasma membrane, leading to increased protein synthesis. FOXO3A becomes excluded from the nucleus and activity of other transcription factors including LHX8 may be altered. Other growth factors from the granulosa cells may also be able to initiate oocyte growth, and triggering oocyte growth can also stimulate the morphological transition and mitotic proliferation of the granulosa cell (*dotted lines*)

compartment, and recent work supports this model. Mechanistic target of rapamycin complex-1 (mTORC1) regulates growth of many cell types, typically by increasing the rate of protein synthesis (Brunn et al. 1997). When Raptor (*Rptor*), which encodes an essential component of mTORC1, was selectively deleted in granulosa cells, the primordial follicles did not initiate growth (Zhang et al. 2014b). This was not due to cell death, as the follicles remained intact within the ovary for several weeks, although they subsequently degenerated. Conversely, when tuberous sclerosis 1 (*Tsc1*), which encodes an inhibitor of mTORC1 signaling, was deleted, the granulosa cells of most primordial follicles became cuboidal and the enclosed oocytes grew robustly (Zhang et al. 2014b). These results suggest that an increase in protein synthesis within the granulosa cells could be the signal that initiates oocyte growth (Fig. 8.2).

Several lines of evidence implicate KIT ligand (KITL) as a key growth-promoting signal for the oocyte. KITL is expressed by developing granulosa cells and binds to its receptor KIT on the oocyte, causing its phosphorylation (Manova et al. 1993) and activation of downstream signaling via the phosphatidylinositide (PI) 3-kinase pathway (Liu et al. 2006; Manova et al. 1993; Reddy et al. 2005, 2008). Early studies showed that adding KITL to cultures of ovarian fragments obtained from early postnatal animals induced follicle growth whereas an anti-KIT antibody known to block its function, or chemical inhibitors of KIT, reduced the number of growing follicles (Packer et al. 1994; Parrott and Skinner 1999; Yoshida et al. 1997). Similarly, fibroblasts expressing membrane-associated KITL induce

growth of oocytes in the absence of granulosa cells (Thomas et al. 2008). Consistent with these results, deletion of *Tsc1* in granulosa cells of primordial follicles (which activates oocyte growth—see above) is associated with a substantial increase in KITL expression. Conversely, deletion within the oocyte of genes encoding negative regulators of PI3-kinase signaling, including phosphatase and tensin homolog (*Pten*), *Tsc1*, and *Tsc2*, is sufficient to cause the oocytes of most primordial follicles to begin growth (Adhikari et al. 2009, 2010).

Despite this considerable evidence favoring a key role for KIT signaling in the initiation of oocyte growth, the picture is not entirely clear. Mice carrying an Y719F mutation in KIT are unable to signal through the PI3-kinase pathway (John et al. 2009). Although females are subfertile, a small number of oocytes grow and give rise to apparently normal offspring (John et al. 2008; Kissel et al. 2000; Zhang et al. 2014b). This result seems to clearly indicate that oocyte growth does not absolutely require KIT activity or, by implication, KITL produced by the granulosa cells. Numerous other growth factors have been implicated in the activation of primordial follicles, including leukemia inhibiting factor (LIF), nerve growth factor (NGF), basic fibroblast growth factor (bFGF), keratinocyte growth factor (KGF), platelet-derived growth factor (PDGF), bone morphogenetic proteins (BMP), and connective tissue growth factor (CTGF) (Dissen et al. 2001; Kezele et al. 2005; Lee et al. 2001; Nilsson and Skinner 2001; Nilsson et al. 2006; Schindler et al. 2010). One possibility is that activation of KIT signaling by KITL plays an important role under normal conditions but that, when this pathway is disabled, other signals initiate oocyte growth. It is also worth noting that the relative importance of different growth factors may vary among species (Gilchrist et al. 2008).

Although the evidence described above favors granulosa cell-derived signals as initiators of primordial follicle activation and oocyte growth, several intriguing observations suggest that this is not an exclusive mechanism. Within the oocyte, the transcriptional regulator, forkhead box O 3A (FOXO3A), plays a central role in regulating the initiation of growth. FOXO3A is predominantly nuclear in oocytes within primordial follicles, but becomes translocated to the cytoplasm in growing oocytes (Li et al. 2010). As FOXO3A is thought to function primarily as a transcriptional repressor, this result suggests that it represses genes whose products promote oocyte growth (Brunet et al. 1999). In females lacking *Foxo3a*, the oocytes within virtually all primordial follicles begin to grow shortly after birth (Castrillon et al. 2003; Hosaka et al. 2004). In these follicles, the granulosa cells surrounding these oocytes become cuboidal and proliferate. Similarly, when the suppressors of the PI3-kinase pathway are genetically deleted in the oocyte, as described above, not only do the oocytes begin to grow, but the granulosa cells again become cuboidal and proliferate (Castrillon et al. 2003; Hosaka et al. 2004; John et al. 2008). Thus, it seems that activation of primordial follicles can be triggered by signals originating either in the granulosa cells or in the oocyte and that each cell type is capable of responding appropriately to signals received from the other so that oocyte and follicular growth are initiated in a coordinated manner (Fig. 8.2).

Other transcriptional regulators also play key roles in regulating the initiation of oocyte growth. LIM homeobox gene 8 (*Lhx8*) encodes a transcription factor that is

abundantly expressed in oocytes in primordial follicles, as well as at later stages. When *Lhx8* was globally deleted, oocytes within primordial follicles failed to grow, suggesting an important role in oocyte growth (Choi et al. 2008a; Pangas et al. 2006). Recently, however, transgenic mice expressing Cre recombinase under the control of the promoter of *Gdf9* were used to selectively inactivate *Lhx8* in oocytes within primordial follicles (Ren et al. 2015). Many oocytes began to grow shortly after birth, accompanied by an increase in phosphorylated protein kinase B (AKT), indicating that PI3-kinase signaling was active and that FOXO3A translocated from the nucleus to the cytoplasm. Strikingly, however, the granulosa cells failed to undergo the squamous-cuboidal transition and the oocytes did not grow beyond ~30 μm in diameter (Ren et al. 2015). These results not only suggest that that LHX8 normally acts to maintain oocytes in a quiescent nongrowing condition, but may hint that signaling pathways that can relay the growth signal from the oocyte to the granulosa are dependent on LHX8. The spermatogenesis- and oogenesis-specific basic helix-loop-helix 1/2 (SOHLH1/2) proteins may play a similar role. These are abundant in oocytes of primordial follicles, but less detectable in growing oocytes. When either *Sohlh1* or *Sohlh2* is deleted, there is a rapid loss of oocytes and follicles. Some oocytes begin to grow, but the granulosa cells remain squamous (Choi et al. 2008b; Pangas et al. 2006), likely explaining why oocyte growth fails to progress. Taken together, the studies demonstrate that the entry of primordial follicles into the growth pool is an intricately coordinated process that can be regulated by multiple factors in both the oocyte and the granulosa cells.

8.5 Maintenance of Oocyte Growth and Follicle Development

8.5.1 Metabolic Support

The enormous increase in size of the growing oocyte makes it among the largest cells in the body. Growth is associated with an accumulation of a large stock of mRNAs and proteins, which direct late oocyte development and early embryonic development after fertilization, and of organelles such as mitochondria. This activity imposes a substantial metabolic burden on the oocyte (Collado-Fernandez et al. 2012). The oocyte does not shoulder this alone but outsources some essential tasks to the granulosa cells that surround it, allowing it to minimize its own energy expenditure (Fig. 8.3).

Follicular growth is associated with an increased consumption of glucose and oxygen, both in vivo (Harris et al. 2009) and in vitro (Boland et al. 1994; Harris et al. 2007). Many studies have shown that mammalian oocytes, including human (Tsutsumi et al. 1990) and mouse (Brinster 1971; Eppig 1976; Fagbohun and Downs 1992; Tsutsumi et al. 1992; Zuelke and Brackett 1992), cannot efficiently

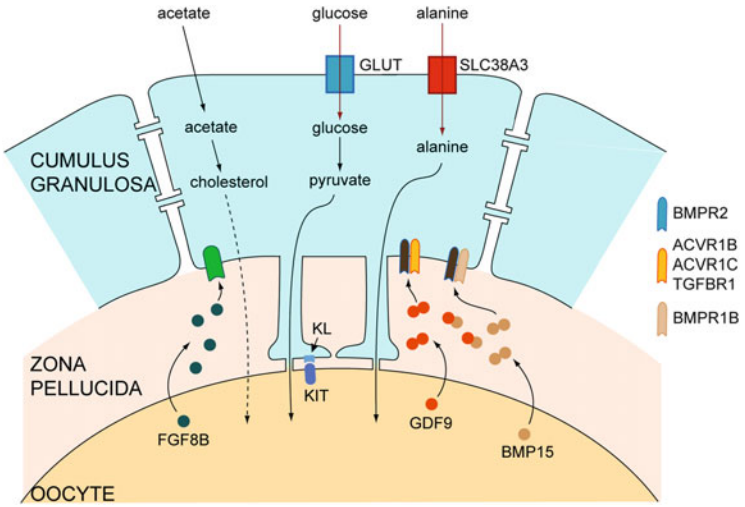

Fig. 8.3 Oocyte–granulosa cell communication during growth. The oocyte is unable to efficiently produce cholesterol or pyruvate or to take up certain amino acids such as alanine. The granulosa cells produce these essential factors and transfer them to the oocyte, via gap junctions in the case of pyruvate and amino acids. Conversely, the oocyte secretes several growth factors that are required for granulosa cell proliferation and/or differentiation. GDF9 and BMP15 may be presented to the granulosa cells as homo- or heterodimers

metabolize glucose to generate pyruvate. Indeed, oocytes are relatively deficient in expression of mRNAs encoding glycolytic enzymes including *Idoa*, *Eno1*, *Ldha*, *Pfkp*, *Pkm2*, and *Tpi1*, whereas these genes are highly expressed by surrounding granulosa cells (Sugiura et al. 2005; Sugiura et al. 2007). Granulosa cells metabolize glucose and then transfer pyruvate to the oocyte (Brinster 1971; Tsutsumi et al. 1990). Nevertheless, one study traced the uptake of glucose by granulosa cells via glucose transporters and its transfer into the oocyte through gap junctions (Wang et al. 2012). Glucose transferred to the oocyte may be metabolized by non-glycolytic pathways such as the pentose phosphate or the hexosamine biosynthesis pathways (Collado-Fernandez et al. 2012). Hence, the oocyte relies on the somatic compartment to obtain its source of ATP. As reduced ATP content in the oocyte has been linked to defects of the meiotic spindle (Zhang et al. 2006), as well as diminished developmental competence (Igarashi et al. 2005; Van Blerkom et al. 1995), this cooperation between the oocyte and granulosa cells is essential for normal embryo development and fertility.

The protein synthetic activity of the oocyte evidently depends on a large supply of amino acids. But the oocyte is only poorly able to take up certain amino acids including alanine, glycine, and proline in vitro. Uptake significantly improves in the presence of surrounding granulosa cells, which transfer the amino acids through gap junctions (Colonna and Mangia 1983; Haghighat and Van Winkle 1990). Consistent with these observations, certain transporters including solute carrier (SLC) 38A3, whose substrates include histidine and alanine (Eppig et al. 2005),

and SLC7A6, a cationic amino acid transporter (Corbett et al. 2014), are present only in granulosa cells. In addition, although other amino acid transport systems are active in denuded oocytes, they are functionally improved by the presence of surrounding granulosa cells (Colonna and Mangia 1983; Haghighat and Van Winkle 1990; Pelland et al. 2009). By supplying the oocyte with the amino acids for which it lacks transporters, granulosa cells thus greatly increase the efficiency of oocyte protein synthesis.

The oocyte also depends on the granulosa cells to obtain cholesterol. Many enzymes that act in the cholesterol biosynthetic pathway are expressed at relatively low levels in oocytes. Consistently, acetate is inefficiently converted to cholesterol in granulosa-free oocytes (Su et al. 2008). Oocytes also lack the ability to import cholesterol as they do not express receptors for HDL cholesterol and LDL cholesterol (Sato et al. 2003; Trigatti et al. 1999). Oocytes are thus deficient in both cholesterol synthesis and uptake of extracellular cholesterol. Instead, the surrounding granulosa cells, which express the necessary enzymes, synthesize cholesterol and transfer it to the oocyte (Su et al. 2008). Accumulation of cholesterol in oocytes is essential for normal embryo development (Comiskey and Warner 2007), as early preimplantation embryos are also unable to synthesize cholesterol (Pratt 1982). Thus, the metabolic cooperation between the granulosa cells and the oocyte, which ensures the deposition of cholesterol into the oocyte, is critical for the oocyte to acquire embryonic developmental competence.

The molecular deficiencies of the oocyte thus force it to be strictly dependent in several metabolic respects on the granulosa cells. However, the oocyte does not merely receive these metabolites from the granulosa cells. Rather, it instructs the granulosa cells to synthesize and/or supply the nutrients it needs to meet its metabolic requirements (Fig. 8.3). A clue to this was the observation that expression of several enzymes of the glycolytic pathway (Emori et al. 2013; Sugiura et al. 2005; Sugiura et al. 2007) and that of several amino acid transporters (Emori et al. 2013; Eppig et al. 2005) are all enriched in the cumulus cells adjacent to the oocyte as compared to mural granulosa on the inner wall of the follicle. It was then discovered that their expression, as well as that of several enzymes in the cholesterol biosynthetic pathway, is enhanced by oocytes or ODPFs, including GDF9 and BMP15 (Emori et al. 2013; Eppig et al. 2005; Nakamura et al. 2015; Sugiura et al. 2005, 2007; Su et al. 2008). These observations highlight the dynamic and bidirectional nature of the signaling between the oocyte and granulosa cells that allow the growing oocyte to meet its high metabolic demands.

8.5.2 Estradiol Synthesis

Steroidogenesis occurs in the adrenal gland, ovary, testis, and placenta. In the ovary, this process has long been considered as an example of intercellular cooperation, as exemplified by the "two-cell, two-gonadotropin" model describing cooperation between the granulosa and theca cells (Fortune and Armstrong

1977). Steroidogenesis involves the uptake and metabolism of cholesterol by steroidogenic enzymes and is driven by follicle-stimulating hormone (FSH) and LH (Conley et al. 1994; Parker and Schimmer 1995; Richards 1994; Richards et al. 2002; Tian et al. 1995). Because theca cells do not express the FSH receptor (Camp et al. 1991; Zeleznik et al. 1974), the theca–granulosa cooperation is necessary for estrogen synthesis.

Steroidogenesis is initiated by the import of cholesterol into mitochondria by steroidogenic acute regulatory protein (StAR) (Arakane et al. 1996; Clark et al. 1994; Stocco 2001). Cholesterol is then cleaved by the mitochondrial enzyme, cytochrome P450 (CYP) side-chain cleavage (CYP11A) producing pregnenolone. CYP17, 3-β-hydroxysteroid dehydrogenase (HSD), and 17β-HSD metabolize pregnenolone to produce androgens. These are then converted by aromatase (CYP19) to estradiol (Miller 1988; Peter and Dubuis 2000; Simpson et al. 1994). LH, whose receptor is on theca as well as granulosa cells (Camp et al. 1991; Erickson et al. 1979; Piquette et al. 1991; Segaloff et al. 1990; Zeleznik et al. 1974), drives androgen synthesis (Fortune and Armstrong 1977; Richards et al. 1987; Smyth et al. 1994), while FSH stimulates aromatase activity in granulosa cells (Dorrington et al. 1975; Erickson et al. 1979; Hickey et al. 1988; Fitzpatrick and Richards 1991; Whitelaw et al. 1992), enabling the production of estrogen from the theca-derived androgens. In combination with other follicular factors such as insulin-like growth factor 1 (IGF-1), LH (Devoto et al. 1999; McGee et al. 1996; Schoppee et al. 2002; Sekar et al. 2000; Simone et al. 1993; Willis et al. 1996; Zhang et al. 2000) and FSH (Adashi 1994; Balasubramanian et al. 1997; Glister et al. 2001; Hsu and Hammond 1987; Khalid et al. 2000; Monniaux and Pisselet 1992; Pescador et al. 1997; Silva and Price 2002; Spicer et al. 2002; Yoshimura 1998) increases expression of these steroidogenic enzymes in the theca and the granulosa cells, thus promoting androgen and estrogen synthesis, respectively, both in vivo and in vitro.

Hence, the combined actions of theca cells and granulosa cells enable steroidogenesis in the developing ovarian follicle. Recent findings suggest, however, that ovarian steroidogenesis is regulated by three, rather than two, follicular cell types. The removal of an oocyte from a granulosa–oocyte complex in vitro by oocytectomy leads to reduction in estradiol levels, suggesting that the oocyte promotes its production (Vanderhyden et al. 1993). Oocytes have been shown to enhance estradiol production in nonmammalian species as well (Sretarugsa and Wallace 1997; Yoshimura et al. 1994). In contrast, other studies have shown that oocyte signaling decreases the expression of steroidogenic enzymes (Diaz et al. 2007; Pangas et al. 2006; Wigglesworth et al. 2015) and suppresses FSH-stimulated estradiol synthesis (Glister et al. 2001; Vitt et al. 2000). These seemingly contradictory results may indicate that oocyte signaling promotes estradiol synthesis while also inhibiting the premature luteinization of the granulosa cells (Coskun et al. 1995; el-Fouly et al. 1970; Nekola and Nalbandov 1971) through independent pathways (Vanderhyden et al. 1993; Vanderhyden and Tonary 1995). The mechanism of how the oocyte achieves such a balanced effect remains to be elucidated.

The follicle itself is a target of the estradiol, as well as of the androgens, that it produces (Berisha et al. 2002; LaVoie et al. 2002; Rosenfeld et al. 2001; Walters et al. 2012). Estradiol drives folliculogenesis (Couse et al. 2005; Emmen et al. 2005; Krege et al. 1998) and prevents follicular atresia (Britt et al. 2000; Dupont et al. 2000; Richards 1980; Robker and Richards 1998; Rosenfeld et al. 2001). It promotes the survival of granulosa cells as well as the oocyte by inhibiting the expression of pro-apoptotic genes (Billig et al. 1993; Lund et al. 1999; Quirk et al. 2006; Toda et al. 2001). It also increases expression of IGF-1, thus promoting expression of FSH and LH targets within the follicle (Hsu and Hammond 1987; Khalid et al. 2000). Moreover, estradiol has also been shown to enhance the expression of natriuretic peptide receptor B (NPR2; see below), thus enabling the maintenance of meiotic arrest (Zhang et al. 2011). Hence, the cooperation between the various follicular compartments, resulting in the production of estradiol, is crucial for survival and development of the follicle as a whole.

8.6 Terminal Stages of Oocyte Growth and Development

8.6.1 Nuclear Differentiation of the Oocyte

Growing oocytes are highly active transcriptionally (Bachvarova 1985), likely reflecting that they must manufacture not only the RNA needed to sustain growth itself but also the RNA needed during the transcriptionally inactive periods of meiotic maturation and early embryonic development. In the mouse, the oocyte RNA content increases 300-fold during growth (Schultz et al. 1979; Sternlicht and Schultz 1981). Upon reaching full size, however, the oocyte dramatically reduces its transcriptional activity to an undetectable level (Bouniol-Baly et al. 1999; Christians et al. 1999; Worrad et al. 1994). Coincident with transcriptional arrest, the chromatin, which was previously dispersed throughout the nucleus in a configuration termed non-surrounded nucleolus (NSN), becomes partially condensed and prominently arrayed around the nucleolus in a configuration termed surrounded nucleolus (SN) (Mattson and Albertini 1990; Wickramasinghe et al. 1991). This conformational change, termed the NSN-SN transition, occurs in oocytes of many mammalian species (Fuhrer et al. 1989; Hinrichs et al. 1993; Parfenov et al. 1989). Although the events of chromatin remodeling and transcriptional silencing are temporally correlated, they do not strictly depend on each other (Abe et al. 2010; Andreu-Vieyra et al. 2010; De La Fuente 2006). For example, the dissociation of RNA polymerase II from DNA, an indication of transcriptional silencing, is not dependent on the NSN-SN transition (Abe et al. 2010).

The NSN-SN is associated with a number of events of late oocyte development, including a change in the nuclear concentration of the transcription factors, specificity protein 1 (SP1) and TATA box-binding protein (TBP), and a rearrangement of cytoplasmic components including the appearance of microtubule-organizing

centers (Lodde et al. 2008; Mattson and Albertini 1990; Wickramasinghe et al. 1991). Hence, it is considered to be a reliable and consistent marker of oocyte developmental progression and generally strongly correlates with the ability of the oocyte to develop as an embryo following fertilization (Wickramasinghe et al. 1991; Zuccotti et al. 2002). Intriguingly, the granulosa cells play an important but not fully defined role in mediating the NSN-SN transition (De La Fuente 2006; Luciano et al. 2011). When the FSH analogue, equine chorionic gonadotropin, was injected into females and the cumulus cell–oocyte complexes recovered 2 days later from antral follicles, an increased number had undergone the NSN-SN transition and had ceased transcriptional activity as compared to oocytes obtained from non-injected females. This effect was not observed in oocytes that were not closely associated with cumulus cells at the time of recovery. Similarly, incubation of bovine cumulus–oocyte complexes in the presence of FSH promoted the NSN-SN transition together with increased gap junctional communication. As the FSH receptors are located on the granulosa cells, these observations imply that the granulosa send signals to the oocyte that promote its nuclear differentiation.

8.6.2 Maintenance of Meiotic Arrest

Throughout its growth, the oocyte remains arrested at late prophase I of the meiotic cell cycle. Upon the initiation of meiotic maturation, it completes the first meiotic division and then becomes arrested at metaphase of meiosis II. Physiologically, maturation is triggered by LH (Dekel et al. 1979; Eppig 1980), thereby coordinating it with ovulation. However, experiments in which oocytes of antral follicles are isolated and placed in culture showed that they acquire the ability to mature to metaphase II about midway through growth, which is well before the follicle acquires the ability to ovulate in response to LH (Sorensen and Wassarman 1976). This result established the concept that the follicular environment actively prevents oocytes from undergoing maturation until this inhibition is relieved by LH. The granulosa cells play a central role in mediating this inhibition.

Maturation is triggered by a protein complex, known as metaphase-promoting factor (MPF), that consists of a catalytic subunit, cyclin-dependent kinase 1 (CDK1), and a regulatory subunit, cyclin B1 (Brunet and Maro 2005). The activity of this complex is regulated by phosphorylation of specific sites on CDK1. The WEE1/MYT1 kinases phosphorylate two sites, which inhibits CDK1 activity, whereas the CDC25 phosphatases dephosphorylate these sites, thereby activating CDK1 (Lew and Kornbluth 1996; Lincoln et al. 2002). The relative activities of WEE/MYT1 and CDC25 thus control CDK1 activity. These two regulators are themselves controlled by phosphorylation. Phosphorylation by cyclic (c) AMP-dependent kinase A activates WEE1 and inhibits CDC25 (Han et al. 2005; Oh et al. 2010; Pirino et al. 2009; Zhang et al. 2008). Hence, high protein kinase A activity maintains CDK1 in an inactive state.

As would be anticipated based on the foregoing, cAMP levels have long been known to be high in follicle-enclosed oocytes and to drop rapidly when oocytes are removed from the follicle and placed in culture (Schultz et al. 1983). This suggests that the follicular environment prevents oocyte maturation by maintaining a high level of cAMP within the oocyte (Cho et al. 1974; Dekel and Beers 1978). Because the growth of antral follicles is promoted by FSH, whose receptors are located on the granulosa cells, and gonadotropins can increase intracellular cAMP, an attractive hypothesis was that the granulosa cells transferred cAMP to the oocyte via the communicating gap junctions. Subsequent work showed, however, that the oocyte is able to synthesize cAMP (Olsiewski and Beers 1983; Urner et al. 1983) through the activity of a G protein related receptor, GPR3 (Mehlmann et al. 2002, 2004). Importantly, when *Gpr3* was deleted from oocytes, they underwent maturation within the follicle in the absence of LH signaling (Mehlmann et al. 2004), providing convincing evidence that the oocyte-generated cAMP is needed to maintain high protein kinase A activity and inhibit maturation.

Although the granulosa cells may not be a primary source of cAMP for the oocyte, they nonetheless play a critical role in maintaining its meiotic arrest. cAMP in the oocyte is metabolized by phosphodiesterase (PDE)-3A (Bornslaeger et al. 1984; Dekel and Beers 1978, 1980; Jensen et al. 2002; Richard et al. 2001; Shitsukawa et al. 2001; Tsafriri et al. 1996; Vivarelli et al. 1983). Within the follicle, the granulosa cells transfer cyclic GMP, an inhibitor of PDE3A, to the oocyte, again via the gap junctions (Norris et al. 2009; Richard and Baltz 2014; Zhang et al. 2010). This helps to maintain a high steady-state level of cAMP within the oocyte. Intriguingly, the production of cGMP illustrates the cooperativity between the mural and cumulus granulosa cells. The mural cells secrete C-type natriuretic peptide C (CNP, encoded by *Nppc*), which binds to NPR2 located on the plasma membrane of the cumulus cells. NPR2 is associated with guanylate cyclase and thus upon its activation increases the production of cGMP (Fig. 8.4) (Franciosi et al. 2014; Kawamura et al. 2011; Zhang et al. 2010, 2011). The essential role of this cGMP is demonstrated by mice bearing mutations in either *Npr2* or *Nppc*. Their oocytes are unable to maintain meiotic arrest and thus undergo maturation within the ovarian follicle independently of LH signaling. Thus, by supplying an inhibitor of PDE3A activity, the granulosa cells maintain a high level of cAMP in the oocyte, which activates protein kinase A and keeps CDK1 in an inactive state.

Once again, the oocyte plays a key role in facilitating this granulosa cell function. GDF9 and BMP15 promote the expression of *Nppc* and *Npr2* (Lee et al. 2013; Wigglesworth et al. 2015; Zhang et al. 2010, 2011) and also enhance the expression of *Impdh*, an enzyme required for the production of cGMP (Wigglesworth et al. 2013). The oocyte thus plays a key role in maintaining its meiotic arrest. This cooperation between the two compartments, by preventing LH-independent meiotic maturation within the follicle, ensures that the production of a fertilizable egg is temporally coordinated with its ovulation.

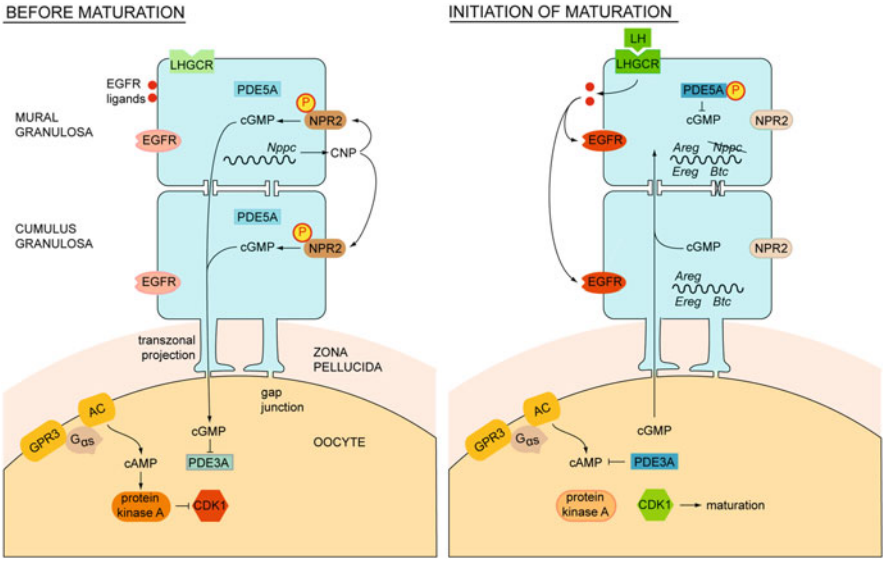

Fig. 8.4 Oocyte–granulosa communication and the regulation of meiotic maturation. Prior to maturation, CNP secreted by the granulosa cells activates NPR2 receptors that are coupled to guanylate cyclase, thereby activating cGMP synthesis. cGMP is transferred to the oocyte via gap junctions, where it inhibits PDE3A. This allows the level of cAMP in the oocyte, generated by the activity of Gs-coupled GPR3, to remain high. Protein kinase A is therefore active and CDK1 is inactive. Upon binding of LH to its receptor, LHGCR, on the mural granulosa cells, EGFR ligands (amphiregulin, beta-cellulin, and epiregulin) are released by the mural granulosa cells and activate EGFR on both mural and cumulus granulosa cells. Signaling through LHGCR and EGFR causes cGMP levels in the granulosa to fall via phosphorylation and activation of PDE5A, dephosphorylation of NPR2, and reduced *Nppc* transcription and CNP production. Transcription of *Areg*, *Btc*, and *Ereg* are also increased. The specific contributions of LHGCR and EGFR signaling to each of these events remain to be fully defined. The net result is a loss of cGMP from the oocyte, thus relieving the inhibition of PDE3A. Shortly after the initiation of maturation, gap junctional communication between granulosa cells and oocytes is disrupted

8.7 Ovulation-Associated Events

8.7.1 Meiotic Maturation

Meiotic maturation, the final stage of oocyte development, is triggered by the LH surge (Lei et al. 2001; Neal and Baker 1975). The LH receptors are expressed only on the mural granulosa cells within the follicle (as well as the thecal cells on the outside of the follicle). Therefore, this signal must be relayed to the oocyte. The discovery of the central role of granulosa cell-derived cGMP in maintaining meiotic arrest enabled the mechanism by which the LH signal is relayed to be deciphered. Less than 5 min after adding LH to antral follicles in vitro, the level of cGMP in the mural granulosa cells falls (Shuhaibar et al. 2015). This is followed by a decrease in cGMP in the cumulus granulosa cells and in the oocyte. Importantly, the rapid

decrease in cumulus cell and oocyte cGMP is prevented by chemical inhibitors of gap junction permeability. cGMP decreases later in the cumulus cells and oocytes even when gap junctions are not functional, suggesting that multiple mechanisms may contribute to the decrease (Shuhaibar et al. 2015).

Several processes that could contribute to the fall in cGMP have been identified. Upon LH stimulation, NPR2 becomes rapidly dephosphorylated and inactivated, thus inhibiting the synthesis of new cGMP (Egbert et al. 2014; Robinson et al. 2012). In addition, PDE5A, which degrades cGMP, becomes phosphorylated and activated (Egbert et al. 2014). This likely increases the rate of degradation of existing cGMP. Following these early events, the amount of *Nppc* mRNA falls, which could lead to a reduction in the amount of CNP (Kawamura et al. 2011; Lee et al. 2013; Liu et al. 2014; Robinson et al. 2012). Finally, gap junctional communication becomes impaired both between granulosa cells and between the granulosa cells and oocyte, thus preventing the transfer of cGMP into the germ cell. It has been proposed that LH triggers a direct loss of cGMP in the mural granulosa cells that is followed by a flow of cGMP from the cumulus cells and oocyte to the mural cells to equalize the concentration of cGMP through the follicle (Shuhaibar et al. 2015). Thus, gap junctional communication is required for the initial reduction in cGMP within the cumulus cells and oocyte. Later, however, the mechanisms described above may directly reduce cGMP in cumulus cells, leading to a further fall in the oocyte cGMP.

The mechanism by which LH activates the multiple pathways that reduce cGMP remains to be fully elucidated. Upon binding of LH to its receptor, the mural granulosa cells release three members of the epidermal growth factor (EGF) family—amphiregulin (AREG), beta-cellulin (BTC), and epiregulin (EREG) (Conti et al. 2012; Park et al. 2004). As both the mural and cumulus granulosa cells express EGF receptors, this suggests that LH could act by transactivating EGF receptor (EGFR) signaling (Fig. 8.4). This provides a pathway by which the LH signal received by the mural granulosa cells could be relayed to the cumulus granulosa and ultimately the oocyte, and numerous observations support a central role for EGFR signaling in maturation. First, EGF is able to induce maturation of follicle-enclosed oocytes and expansion of the cumulus layer in the absence of LH (Ashkenazi et al. 2005; Downs and Chen 2008; Park et al. 2004). Second, the EGF-like ligands released by the granulosa cells in response to LH, as well as EGF, trigger the decrease in follicular cGMP independently of LH (Hsieh et al. 2011; Norris et al. 2010). Third, in mutants carrying the hypomorphic $Egfr^{wa2/wa2}$ allele or lacking *Areg* or *Ereg*, the oocytes fail to mature in response to LH (Hsieh et al. 2007, 2011).

Yet, other observations imply that LH may act independently of EGFR signaling. The extremely rapid drop in cGMP in the mural granulosa cells, which is detectable within a minute of adding LH in vitro (Shuhaibar et al. 2015), suggests that the gonadotropin acts directly on the granulosa cells. Consistent with this, LH induces a rapid drop in cGMP in the oocytes of $Egfr^{wa2/wa2}$ mice, although some EGFR activity remains in this mutant (Hsieh et al. 2011). In addition, the decrease in *Nppc* mRNA during maturation does not appear to require EGFR activity (Liu

et al. 2014). It may be that LH acts through EGFR-dependent and -independent pathways to achieve a robust and stable decrease in cGMP throughout the preovulatory follicle, thus enabling activation of PDE3A in the oocyte and the consequent resumption of meiosis.

In addition to their role in the initiation of maturation, the granulosa cells also influence the maturation process itself. It is well established that cumulus cell-free oocytes can complete nuclear maturation to metaphase II in vitro, but develop poorly as embryos as compared to oocytes that mature within cumulus–oocyte complexes (Chang et al. 2005; De La Fuente et al. 1999). This suggests that cytoplasmic events of maturation may depend on or occur more efficiently in the presence of the cumulus cells. As noted earlier, a subset of mRNAs that had previously been synthesized and stored become translationally activated during maturation (Chen et al. 2011; Paynton and Bachvarova 1994; Su et al. 2007). Among these is *Tpx2*, encoding a protein required for proper assembly and function of the meiotic spindle (Chen et al. 2013). Although TPX2 increases when cumulus-free oocytes are matured in vitro, this increase is greater in cumulus-enclosed oocytes than in cumulus-free oocytes and is further enhanced in the presence of AREG. Translational activation is associated with an AREG-dependent increase in the activity of mTORC (mechanistic target of rapamycin complex) in the oocyte. Further evidence of the importance of signals from the cumulus cells comes from oocytes of mice deficient in either *Egfr* or *Areg* (Chen et al. 2013). These show reduced translational activation during maturation and impaired progression to the two-cell stage following fertilization and give rise to relatively small litters. These results indicate that the EGFR-mediated increase in translational activity during meiotic maturation is developmentally important and are consistent with previous reports that oocytes matured in vitro in the presence of EGF (De La Fuente et al. 1999; Richani et al. 2014) manifested increased developmental competence. Because gap junctional communication between the oocyte and granulosa cells is lost relatively early during the maturation process, it will be important to identify when and by what pathway the granulosa cells exert their beneficial effects on maturing oocytes.

8.7.2 Cumulus Expansion

As a result of LH-driven cumulus expansion, the egg is ovulated embedded in a mucified mass of cumulus granulosa cells. Cumulus expansion facilitates transport of the cumulus–oocyte complex through the oviduct (Chen et al. 1993) and is also essential for fertilization, likely by permitting the sperm to pass through the cumulus layers to reach the oocyte (Eisenbach 1999; Salustri et al. 2004; Van Soom et al. 2002). Cumulus expansion is characterized by the physical dispersal of the cumulus granulosa cells as an extracellular matrix becomes deposited around the individual cells (Fig. 8.5). This matrix consists mostly of the glycosaminoglycan, hyaluronan (HA), as well as HA-binding proteins (Chen et al. 1996; Fulop

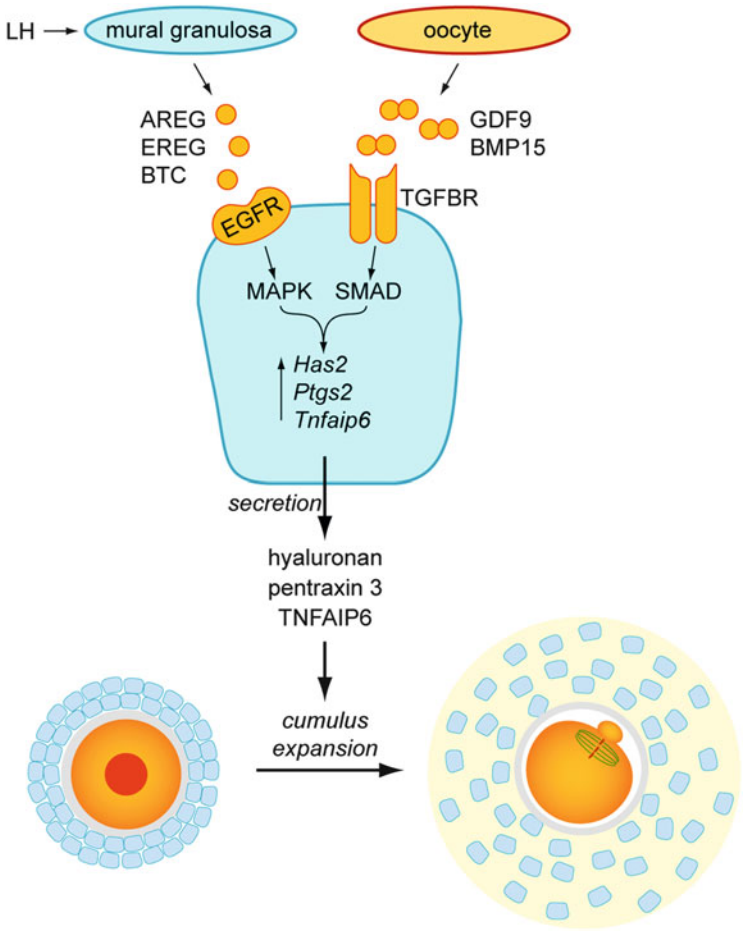

Fig. 8.5 Oocyte–granulosa communication and cumulus expansion. The combined influences of EGFR activation by its ligands and SMAD signaling activated by oocyte-derived factors such as GDF9 and BMP15 trigger the increased transcription in the cumulus cells of genes whose products are secreted and assembled into a matrix. This process, termed cumulus expansion, is required for efficient fertilization

et al. 1997a; Hess et al. 1998; Sato et al. 2001; Yoshioka et al. 2000) and proteoglycans (McArthur et al. 2000) that maintain the stability and structure of the matrix. The integrity of the mucified matrix also requires proteins such as pentraxin 3 (PTX3) and tumor necrosis factor-inducible gene 6 (TNFAIP6) (Baranova et al. 2014; Fulop et al. 2003; Ievoli et al. 2011; Salustri et al. 2004; Sanggaard et al. 2008; Scarchilli et al. 2007) which mediate the interactions between the components of the matrix. These factors, along with other proteins that are needed for cumulus expansion including *Ptgs2* (prostaglandin-endoperoxide synthase 2) (Davis et al. 1999; Ochsner et al. 2003b; Sirois et al. 1992), hyaluronan synthase

2 (*Has2*) (Sugiura et al. 2009), and *Tnfaip6* (Fulop et al. 2003; Ochsner et al. 2003a; Sugiura et al. 2009), are produced by the cumulus cells in response to LH surge (Fig. 8.5) (Eppig 1981; Fulop et al. 1997b; Salustri et al. 1999).

Although cumulus expansion occurs in response to the LH stimulus, which is transduced at least in part through the EGFR pathway, it also requires a signaling input from the oocyte. If the oocyte is microsurgically removed from a cumulus–oocyte complex, the remaining oocytectomized cumulus cell shell cannot undergo expansion in response to EGFR ligands. It can, however, expand if it is cocultured with oocytes or with oocyte-conditioned medium (Buccione et al. 1990; Salustri et al. 1990; Vanderhyden et al. 1990). Many studies using the mouse have demonstrated that signaling by ODPFs is required for upregulation of expansion-related mRNAs and for cumulus expansion (Diaz et al. 2006; Dragovic et al. 2005, 2007; Joyce et al. 2001; Mazerbourg et al. 2004; Moore et al. 2003; Sasseville et al. 2010; Su et al. 2003, 2010; Vanderhyden et al. 2003). Other studies have suggested that the ODPFs are needed for the activation of mitogen-activated protein kinase (MAPK) (Su et al. 2003) and for maintaining the activity of EGFR (Su et al. 2010) in the cumulus cells. The activity of the ODPFs appears to be potentiated by heparan sulfate proteoglycans (HSPG) acting as co-receptors (Watson et al. 2012). The oocyte thus enables cumulus expansion both indirectly through promoting EGFR activity and directly by increasing the expression of expansion-related transcripts.

Although the ability of the oocyte to induce cumulus expansion in vitro is also a feature of other species such as pig and rat (Prochazka et al. 1998; Vanderhyden 1993), in the cow, sheep, and pig, oocyte signaling is not required to induce cumulus expansion (Nagyova et al. 1999; Prochazka et al. 1991; Ralph et al. 1995; Varnosfaderani et al. 2013). Interestingly, oocytes of these species are capable of inducing the expansion of mouse cumulus granulosa cells (Vanderhyden 1993). Whether signaling from the oocyte is required during cumulus expansion in humans remains to be determined. In summary, in the mouse at least, the somatic and germ-line compartments of the follicle cooperate to induce the cumulus cells to expand and mucify in response to the LH signal, thus fulfilling an essential prerequisite for fertilization.

8.8 Conclusion

As in many tissues and organs, growth and differentiation of the ovarian follicle requires a precisely regulated program of intercellular communication and cooperation. We have described several important events during ovarian follicle growth that exemplify this cooperation, specifically between the oocyte and the granulosa cells. Although the gap junction-mediated support from the granulosa to the oocyte is best understood, it is plausible that the granulosa also support the growing oocyte through gap junction-independent pathways. For example, recent work suggests that bovine granulosa cells can transfer mRNAs and long noncoding RNAs to the

oocyte (Macaulay et al. 2014). Yet uncharacterized secreted or membrane-associated molecules of the granulosa may also be important. In addition to the events we have discussed, other aspects of folliculogenesis also depend on intercellular cooperation, including the transition from a primary to a secondary follicle (Braw-Tal 2002; Dong et al. 1996; Galloway et al. 2000; Parrott and Skinner 1999; Yoshida et al. 1997), formation of the TZPs (Carabatsos et al. 1998), thecal cell recruitment (Dong et al. 1996; Liu et al. 2015; Spicer et al. 2008), differentiation of the granulosa into mural and cumulus cell compartments, and granulosa cell luteinization (Eppig et al. 1997; Glister et al. 2003; Li et al. 2000; Nekola and Nalbandov 1971; Vanderhyden 1993). All of these processes serve, directly or indirectly, the growth and development of the oocyte and thereby enable the ovulation of a mature and developmentally competent egg ready to give rise to a new organism.

Competing Interests The authors declare no competing interests.

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References

Abe K, Inoue A, Suzuki MG, Aoki F (2010) Global gene silencing is caused by the dissociation of RNA polymerase II from DNA in mouse oocytes. J Reprod Dev 56:502–507

Adashi EY (1994) Endocrinology of the ovary. Hum Reprod 9:815–827

Adhikari D, Flohr G, Gorre N et al (2009) Disruption of Tsc2 in oocytes leads to overactivation of the entire pool of primordial follicles. Mol Hum Reprod 15:765–770

Adhikari D, Zheng W, Shen Y et al (2010) Tsc/mTORC1 signaling in oocytes governs the quiescence and activation of primordial follicles. Hum Mol Genet 19:397–410

Albertini DF, Rider V (1994) Patterns of intercellular connectivity in the mammalian cumulus-oocyte complex. Microsc Res Tech 27:125–133

Anderson E, Albertini DF (1976) Gap junctions between the oocyte and companion follicle cells in the mammalian ovary. J Cell Biol 71:680–686

Andreu-Vieyra CV, Chen R, Agno JE et al (2010) MLL2 is required in oocytes for bulk histone 3 lysine 4 trimethylation and transcriptional silencing. PLoS Biol 8:e1000453

Arakane F, Sugawara T, Nishino H et al (1996) Steroidogenic acute regulatory protein (StAR) retains activity in the absence of its mitochondrial import sequence: implications for the mechanism of StAR action. Proc Natl Acad Sci U S A 93:13731–13736

Ashkenazi H, Cao X, Motola S et al (2005) Epidermal growth factor family members: endogenous mediators of the ovulatory response. Endocrinology 146:77–84

Bachvarova R (1985) Gene expression during oogenesis and oocyte development in mammals. Dev Biol (N Y 1985) 1:453–524

Balasubramanian K, Lavoie HA, Garmey JC et al (1997) Regulation of porcine granulosa cell steroidogenic acute regulatory protein (StAR) by insulin-like growth factor I: synergism with follicle-stimulating hormone or protein kinase A agonist. Endocrinology 138:433–439

Baranova NS, Inforzato A, Briggs DC et al (2014) Incorporation of pentraxin 3 into hyaluronan matrices is tightly regulated and promotes matrix cross-linking. J Biol Chem 289:30481–30498

Berisha B, Pfaffl MW, Schams D (2002) Expression of estrogen and progesterone receptors in the bovine ovary during estrous cycle and pregnancy. Endocrine 17:207–214
Billig H, Furuta I, Hsueh AJ (1993) Estrogens inhibit and androgens enhance ovarian granulosa cell apoptosis. Endocrinology 133:2204–2212
Boland NI, Humpherson PG, Leese HJ, Gosden RG (1994) The effect of glucose metabolism on murine follicle development and steroidogenesis in vitro. Hum Reprod 9:617–623
Bornslaeger EA, Wilde MW, Schultz RM (1984) Regulation of mouse oocyte maturation: involvement of cyclic AMP phosphodiesterase and calmodulin. Dev Biol 105:488–499
Borum K (1961) Oogenesis in the mouse. A study of the meiotic prophase. Exp Cell Res 24:495–507
Bouniol-Baly C, Hamraoui L, Guibert J et al (1999) Differential transcriptional activity associated with chromatin configuration in fully grown mouse germinal vesicle oocytes. Biol Reprod 60:580–587
Braw-Tal R (2002) The initiation of follicle growth: the oocyte or the somatic cells? Mol Cell Endocrinol 187:11–18
Brinster RL (1971) Oxidation of pyruvate and glucose by oocytes of the mouse and rhesus monkey. J Reprod Fertil 24:187–191
Britt KL, Drummond AE, Cox VA et al (2000) An age-related ovarian phenotype in mice with targeted disruption of the Cyp 19 (aromatase) gene. Endocrinology 141:2614–2623
Brunet S, Maro B (2005) Cytoskeleton and cell cycle control during meiotic maturation of the mouse oocyte: integrating time and space. Reproduction 130:801–811
Brunet A, Bonni A, Zigmond MJ et al (1999) Akt promotes cell survival by phosphorylating and inhibiting a Forkhead transcription factor. Cell 96:857–868
Brunn GJ, Hudson CC, Sekulic A et al (1997) Phosphorylation of the translational repressor PHAS-I by the mammalian target of rapamycin. Science 277:99–101
Buccione R, Vanderhyden BC, Caron PJ, Eppig JJ (1990) FSH-induced expansion of the mouse cumulus oophorus in vitro is dependent upon a specific factor(s) secreted by the oocyte. Dev Biol 138:16–25
Camp TA, Rahal JO, Mayo KE (1991) Cellular localization and hormonal regulation of follicle-stimulating hormone and luteinizing hormone receptor messenger RNAs in the rat ovary. Mol Endocrinol 5:1405–1417
Carabatsos MJ, Elvin J, Matzuk MM, Albertini DF (1998) Characterization of oocyte and follicle development in growth differentiation factor-9-deficient mice. Dev Biol 204:373–384
Castrillon DH, Miao L, Kollipara R et al (2003) Suppression of ovarian follicle activation in mice by the transcription factor Foxo3a. Science 301:215–218
Chang HC, Liu H, Zhang J et al (2005) Developmental incompetency of denuded mouse oocytes undergoing maturation in vitro is ooplasmic in nature and is associated with aberrant Oct-4 expression. Hum Reprod 20:1958–1968
Chen L, Russell PT, Larsen WJ (1993) Functional significance of cumulus expansion in the mouse: roles for the preovulatory synthesis of hyaluronic acid within the cumulus mass. Mol Reprod Dev 34:87–93
Chen L, Zhang H, Powers RW et al (1996) Covalent linkage between proteins of the inter-alpha-inhibitor family and hyaluronic acid is mediated by a factor produced by granulosa cells. J Biol Chem 271:19409–19414
Chen J, Melton C, Suh N et al (2011) Genome-wide analysis of translation reveals a critical role for deleted in azoospermia-like (Dazl) at the oocyte-to-zygote transition. Genes Dev 25:755–766
Chen J, Torcia S, Xie F et al (2013) Somatic cells regulate maternal mRNA translation and developmental competence of mouse oocytes. Nat Cell Biol 15:1415–1423
Cho WK, Stern S, Biggers JD (1974) Inhibitory effect of dibutyryl cAMP on mouse oocyte maturation in vitro. J Exp Zool 187:383–386
Choi Y, Ballow DJ, Xin Y, Rajkovic A (2008a) Lim homeobox gene, lhx8, is essential for mouse oocyte differentiation and survival. Biol Reprod 79:442–449

Choi Y, Yuan D, Rajkovic A (2008b) Germ cell-specific transcriptional regulator sohlh2 is essential for early mouse folliculogenesis and oocyte-specific gene expression. Biol Reprod 79:1176–1182

Christians E, Boiani M, Garagna S et al (1999) Gene expression and chromatin organization during mouse oocyte growth. Dev Biol 207:76–85

Clark BJ, Wells J, King SR, Stocco DM (1994) The purification, cloning, and expression of a novel luteinizing hormone-induced mitochondrial protein in MA-10 mouse Leydig tumor cells. Characterization of the steroidogenic acute regulatory protein (StAR). J Biol Chem 269:28314–28322

Clarke HJ (2012) Post-transcriptional control of gene expression during mouse oogenesis. Results Probl Cell Differ 55:1–21

Collado-Fernandez E, Picton HM, Dumollard R (2012) Metabolism throughout follicle and oocyte development in mammals. Int J Dev Biol 56:799–808

Colonna R, Mangia F (1983) Mechanisms of amino acid uptake in cumulus-enclosed mouse oocytes. Biol Reprod 28:797–803

Comiskey M, Warner CM (2007) Spatio-temporal localization of membrane lipid rafts in mouse oocytes and cleaving preimplantation embryos. Dev Biol 303:727–739

Conley AJ, Howard HJ, Slanger WD, Ford JJ (1994) Steroidogenesis in the preovulatory porcine follicle. Biol Reprod 51:655–661

Conti M, Hsieh M, Zamah AM, Oh JS (2012) Novel signaling mechanisms in the ovary during oocyte maturation and ovulation. Mol Cell Endocrinol 356:65–73

Corbett HE, Dube CD, Slow S et al (2014) Uptake of betaine into mouse cumulus-oocyte complexes via the SLC7A6 isoform of y+L transporter. Biol Reprod 90:81

Coskun S, Uzumcu M, Lin YC et al (1995) Regulation of cumulus cell steroidogenesis by the porcine oocyte and preliminary characterization of oocyte-produced factor(s). Biol Reprod 53:670–675

Couse JF, Yates MM, Deroo BJ, Korach KS (2005) Estrogen receptor-beta is critical to granulosa cell differentiation and the ovulatory response to gonadotropins. Endocrinology 146:3247–3262

Dalton CM, Carroll J (2013) Biased inheritance of mitochondria during asymmetric cell division in the mouse oocyte. J Cell Sci 126:2955–2964

Da Silva-Buttkus P, Jayasooriya GS, Mora JM et al (2008) Effect of cell shape and packing density on granulosa cell proliferation and formation of multiple layers during early follicle development in the ovary. J Cell Sci 121:3890–3900

Davis BJ, Lennard DE, Lee CA et al (1999) Anovulation in cyclooxygenase-2-deficient mice is restored by prostaglandin E2 and interleukin-1beta. Endocrinology 140:2685–2695

De La Fuente R (2006) Chromatin modifications in the germinal vesicle (GV) of mammalian oocytes. Dev Biol 292:1–12

De La Fuente R, O'Brien MJ, Eppig JJ (1999) Epidermal growth factor enhances preimplantation developmental competence of maturing mouse oocytes. Hum Reprod 14:3060–3068

De Smedt V, Szollosi D (1991) Cytochalasin D treatment induces meiotic resumption in follicular sheep oocytes. Mol Reprod Dev 29:163–171

Dekel N, Beers WH (1978) Rat oocyte maturation in vitro: relief of cyclic AMP inhibition by gonadotropins. Proc Natl Acad Sci U S A 75:4369–4373

Dekel N, Beers WH (1980) Development of the rat oocyte in vitro: inhibition and induction of maturation in the presence or absence of the cumulus oophorus. Dev Biol 75:247–254

Dekel N, Hillensjo T, Kraicer PF (1979) Maturational effects of gonadotropins on the cumulus-oocyte complex of the rat. Biol Reprod 20:191–197

Devoto L, Christenson LK, McAllister JM et al (1999) Insulin and insulin-like growth factor-I and -II modulate human granulosa-lutein cell steroidogenesis: enhancement of steroidogenic acute regulatory protein (StAR) expression. Mol Hum Reprod 5:1003–1010

Diaz FJ, O'Brien MJ, Wigglesworth K, Eppig JJ (2006) The preantral granulosa cell to cumulus cell transition in the mouse ovary: development of competence to undergo expansion. Dev Biol 299:91–104

Diaz FJ, Wigglesworth K, Eppig JJ (2007) Oocytes determine cumulus cell lineage in mouse ovarian follicles. J Cell Sci 120:1330–1340

Dissen GA, Romero C, Hirshfield AN, Ojeda SR (2001) Nerve growth factor is required for early follicular development in the mammalian ovary. Endocrinology 142:2078–2086

Dong J, Albertini DF, Nishimori K et al (1996) Growth differentiation factor-9 is required during early ovarian folliculogenesis. Nature 383:531–535

Dorrington JH, Moon YS, Armstrong DT (1975) Estradiol-17beta biosynthesis in cultured granulosa cells from hypophysectomized immature rats; stimulation by follicle-stimulating hormone. Endocrinology 97:1328–1331

Downs SM, Chen J (2008) EGF-like peptides mediate FSH-induced maturation of cumulus cell-enclosed mouse oocytes. Mol Reprod Dev 75:105–114

Dragovic RA, Ritter LJ, Schulz SJ et al (2005) Role of oocyte-secreted growth differentiation factor 9 in the regulation of mouse cumulus expansion. Endocrinology 146:2798–2806

Dragovic RA, Ritter LJ, Schulz SJ et al (2007) Oocyte-secreted factor activation of SMAD 2/3 signaling enables initiation of mouse cumulus cell expansion. Biol Reprod 76:848–857

Dupont S, Krust A, Gansmuller A et al (2000) Effect of single and compound knockouts of estrogen receptors alpha (ERalpha) and beta (ERbeta) on mouse reproductive phenotypes. Development 127:4277–4291

Egbert JR, Shuhaibar LC, Edmund AB et al (2014) Dephosphorylation and inactivation of NPR2 guanylyl cyclase in granulosa cells contributes to the LH-induced decrease in cGMP that causes resumption of meiosis in rat oocytes. Development 141:3594–3604

Eggan K, Jurga S, Gosden R et al (2006) Ovulated oocytes in adult mice derive from non-circulating germ cells. Nature 441:1109–1114

Eisenbach M (1999) Mammalian sperm chemotaxis and its association with capacitation. Dev Genet 25:87–94

el-Fouly MA, Cook B, Nekola M, Nalbandov AV (1970) Role of the ovum in follicular luteinization. Endocrinology 87:286–293

Emmen JM, Couse JF, Elmore SA et al (2005) In vitro growth and ovulation of follicles from ovaries of estrogen receptor (ER){alpha} and ER{beta} null mice indicate a role for ER{beta} in follicular maturation. Endocrinology 146:2817–2826

Emori C, Sugiura K (2014) Role of oocyte-derived paracrine factors in follicular development. Anim Sci J 85:627–633

Emori C, Wigglesworth K, Fujii W et al (2013) Cooperative effects of 17beta-estradiol and oocyte-derived paracrine factors on the transcriptome of mouse cumulus cells. Endocrinology 154:4859–4872

Ene AC, Park S, Edelmann W, Taketo T (2013) Caspase 9 is constitutively activated in mouse oocytes and plays a key role in oocyte elimination during meiotic prophase progression. Dev Biol 377:213–223

Eppig JJ (1976) Analysis of mouse oogenesis in vitro. Oocyte isolation and the utilization of exogenous energy sources by growing oocytes. J Exp Zool 198:375–382

Eppig JJ (1979) Gonadotropin stimulation of the expansion of cumulus oophori isolated from mice: general conditions for expansion in vitro. J Exp Zool 208:111–120

Eppig JJ (1980) Regulation of cumulus oophorus expansion by gonadotropins in vivo and in vitro. Biol Reprod 23:545–552

Eppig JJ (1981) Prostaglandin E2 stimulates cumulus expansion and hyaluronic acid synthesis by cumuli oophori isolated from mice. Biol Reprod 25:191–195

Eppig JJ, Schroeder AC (1989) Capacity of mouse oocytes from preantral follicles to undergo embryogenesis and development to live young after growth, maturation, and fertilization in vitro. Biol Reprod 41:268–276

Eppig JJ, Wigglesworth K, Pendola F, Hirao Y (1997) Murine oocytes suppress expression of luteinizing hormone receptor messenger ribonucleic acid by granulosa cells. Biol Reprod 56:976–984

Eppig JJ, Pendola FL, Wigglesworth K, Pendola JK (2005) Mouse oocytes regulate metabolic cooperativity between granulosa cells and oocytes: amino acid transport. Biol Reprod 73:351–357

Erickson GF, Wang C, Hsueh AJ (1979) FSH induction of functional LH receptors in granulosa cells cultured in a chemically defined medium. Nature 279:336–338

Espey LL (1980) Ovulation as an inflammatory reaction—a hypothesis. Biol Reprod 22:73–106

Fagbohun CF, Downs SM (1992) Requirement for glucose in ligand-stimulated meiotic maturation of cumulus cell-enclosed mouse oocytes. J Reprod Fertil 96:681–697

Findlay JK, Hutt KJ, Hickey M, Anderson RA (2015) How is the number of primordial follicles in the ovarian reserve established? Biol Reprod 93:111

Fitzpatrick SL, Richards JS (1991) Regulation of cytochrome P450 aromatase messenger ribonucleic acid and activity by steroids and gonadotropins in rat granulosa cells. Endocrinology 129:1452–1462

Fortune JE, Armstrong DT (1977) Androgen production by theca and granulosa isolated from proestrous rat follicles. Endocrinology 100:1341–1347

Franciosi F, Coticchio G, Lodde V et al (2014) Natriuretic peptide precursor C delays meiotic resumption and sustains gap junction-mediated communication in bovine cumulus-enclosed oocytes. Biol Reprod 91:61

Fuhrer F, Mayr B, Schellander K et al (1989) Maturation competence and chromatin behaviour in growing and fully grown cattle oocytes. Zentralbl Veterinarmed A 36:285–291

Fulop C, Kamath RV, Li Y et al (1997a) Coding sequence, exon-intron structure and chromosomal localization of murine TNF-stimulated gene 6 that is specifically expressed by expanding cumulus cell-oocyte complexes. Gene 202:95–102

Fulop C, Salustri A, Hascall VC (1997b) Coding sequence of a hyaluronan synthase homologue expressed during expansion of the mouse cumulus-oocyte complex. Arch Biochem Biophys 337:261–266

Fulop C, Szanto S, Mukhopadhyay D et al (2003) Impaired cumulus mucification and female sterility in tumor necrosis factor-induced protein-6 deficient mice. Development 130:2253–2261

Galloway SM, McNatty KP, Cambridge LM et al (2000) Mutations in an oocyte-derived growth factor gene (BMP15) cause increased ovulation rate and infertility in a dosage-sensitive manner. Nat Genet 25:279–283

Gilchrist RB, Lane M, Thompson JG (2008) Oocyte-secreted factors: regulators of cumulus cell function and oocyte quality. Hum Reprod Update 14:159–177

Glister C, Tannetta DS, Groome NP, Knight PG (2001) Interactions between follicle-stimulating hormone and growth factors in modulating secretion of steroids and inhibin-related peptides by nonluteinized bovine granulosa cells. Biol Reprod 65:1020–1028

Glister C, Groome NP, Knight PG (2003) Oocyte-mediated suppression of follicle-stimulating hormone- and insulin-like growth factor-induced secretion of steroids and inhibin-related proteins by bovine granulosa cells in vitro: possible role of transforming growth factor alpha. Biol Reprod 68:758–765

Grive KJ, Freiman RN (2015) The developmental origins of the mammalian ovarian reserve. Development 142:2554–2563

Haghighat N, Van Winkle LJ (1990) Developmental change in follicular cell-enhanced amino acid uptake into mouse oocytes that depends on intact gap junctions and transport system Gly. J Exp Zool 253:71–82

Han SJ, Chen R, Paronetto MP, Conti M (2005) Wee1B is an oocyte-specific kinase involved in the control of meiotic arrest in the mouse. Curr Biol 15:1670–1676

Harris SE, Adriaens I, Leese HJ, Gosden RG, Picton HM (2007) Carbohydrate metabolism by murine ovarian follicles and oocytes grown in vitro. Reproduction 134:415–424

Harris SE, Leese HJ, Gosden RG, Picton HM (2009) Pyruvate and oxygen consumption throughout the growth and development of murine oocytes. Mol Reprod Dev 76:231–238

Hasegawa A, Mochida N, Ogasawara T, Koyama K (2006) Pup birth from mouse oocytes in preantral follicles derived from vitrified and warmed ovaries followed by in vitro growth, in vitro maturation, and in vitro fertilization. Fertil Steril 86:1182–1192

Hertig AT, Adams EC (1967) Studies on the human oocyte and its follicle. I. Ultrastructural and histochemical observations on the primordial follicle stage. J Cell Biol 34:647–675

Hess KA, Chen L, Larsen WJ (1998) The ovarian blood follicle barrier is both charge- and size-selective in mice. Biol Reprod 58:705–711

Hickey GJ, Chen SA, Besman MJ et al (1988) Hormonal regulation, tissue distribution, and content of aromatase cytochrome P450 messenger ribonucleic acid and enzyme in rat ovarian follicles and corpora lutea: relationship to estradiol biosynthesis. Endocrinology 122:1426–1436

Hinrichs K, Schmidt AL, Friedman PP et al (1993) In vitro maturation of horse oocytes: characterization of chromatin configuration using fluorescence microscopy. Biol Reprod 48:363–370

Hirshfield AN (1991) Development of follicles in the mammalian ovary. Int Rev Cytol 124:43–101

Hosaka T, Biggs WH 3rd, Tieu D et al (2004) Disruption of forkhead transcription factor (FOXO) family members in mice reveals their functional diversification. Proc Natl Acad Sci U S A 101:2975–2980

Hsieh M, Lee D, Panigone S et al (2007) Luteinizing hormone-dependent activation of the epidermal growth factor network is essential for ovulation. Mol Cell Biol 27:1914–1924

Hsieh M, Thao K, Conti M (2011) Genetic dissection of epidermal growth factor receptor signaling during luteinizing hormone-induced oocyte maturation. PLoS One 6:e21574

Hsu CJ, Hammond JM (1987) Gonadotropins and estradiol stimulate immunoreactive insulin-like growth factor-I production by porcine granulosa cells in vitro. Endocrinology 120:198–207

Hutt KJ (2015) The role of BH3-only proteins in apoptosis within the ovary. Reproduction 149: R81–R89

Ievoli E, Lindstedt R, Inforzato A et al (2011) Implication of the oligomeric state of the N-terminal PTX3 domain in cumulus matrix assembly. Matrix Biol 30:330–337

Igarashi H, Takahashi T, Takahashi E et al (2005) Aged mouse oocytes fail to readjust intracellular adenosine triphosphates at fertilization. Biol Reprod 72:1256–1261

Jensen JT, Schwinof KM, Zelinski-Wooten MB et al (2002) Phosphodiesterase 3 inhibitors selectively block the spontaneous resumption of meiosis by macaque oocytes in vitro. Hum Reprod 17:2079–2084

John GB, Gallardo TD, Shirley LJ, Castrillon DH (2008) Foxo3 is a PI3K-dependent molecular switch controlling the initiation of oocyte growth. Dev Biol 321:197–204

John GB, Shidler MJ, Besmer P, Castrillon DH (2009) Kit signaling via PI3K promotes ovarian follicle maturation but is dispensable for primordial follicle activation. Dev Biol 331:292–299

Jorgensen JS (2013) Defining the neighborhoods that escort the oocyte through its early life events and into a functional follicle. Mol Reprod Dev 80:960–976

Joyce IM, Pendola FL, O'Brien M, Eppig JJ (2001) Regulation of prostaglandin-endoperoxide synthase 2 messenger ribonucleic acid expression in mouse granulosa cells during ovulation. Endocrinology 142:3187–3197

Kang MK, Han SJ (2011) Post-transcriptional and post-translational regulation during mouse oocyte maturation. BMB Rep 44:147–157

Kawamura K, Cheng Y, Kawamura N et al (2011) Pre-ovulatory LH/hCG surge decreases C-type natriuretic peptide secretion by ovarian granulosa cells to promote meiotic resumption of pre-ovulatory oocytes. Hum Reprod 26:3094–3101

Kessel RG, Tung HN, Roberts R, Beams HW (1985) The presence and distribution of gap junctions in the oocyte-follicle cell complex of the zebrafish, Brachydanio rerio. J Submicrosc Cytol 17:239–253

Kezele P, Nilsson EE, Skinner MK (2005) Keratinocyte growth factor acts as a mesenchymal factor that promotes ovarian primordial to primary follicle transition. Biol Reprod 73:967–973
Khalid M, Haresign W, Luck MR (2000) Secretion of IGF-1 by ovine granulosa cells: effects of growth hormone and follicle stimulating hormone. Anim Reprod Sci 58:261–272
Kidder GM, Mhawi AA (2002) Gap junctions and ovarian folliculogenesis. Reproduction 123:613–620
Kissel H, Timokhina I, Hardy MP et al (2000) Point mutation in kit receptor tyrosine kinase reveals essential roles for kit signaling in spermatogenesis and oogenesis without affecting other kit responses. EMBO J 19:1312–1326
Krege JH, Hodgin JB, Couse JF et al (1998) Generation and reproductive phenotypes of mice lacking estrogen receptor beta. Proc Natl Acad Sci U S A 95:15677–15682
LaVoie HA, DeSimone DC, Gillio-Meina C, Hui YY (2002) Cloning and characterization of porcine ovarian estrogen receptor beta isoforms. Biol Reprod 66:616–623
Lee WS, Otsuka F, Moore RK, Shimasaki S (2001) Effect of bone morphogenetic protein-7 on folliculogenesis and ovulation in the rat. Biol Reprod 65:994–999
Lee KB, Zhang M, Sugiura K et al (2013) Hormonal coordination of natriuretic peptide type C and natriuretic peptide receptor 3 expression in mouse granulosa cells. Biol Reprod 88:42
Lei L, Spradling AC (2013) Female mice lack adult germ-line stem cells but sustain oogenesis using stable primordial follicles. Proc Natl Acad Sci U S A 110:8585–8590
Lei ZM, Mishra S, Zou W et al (2001) Targeted disruption of luteinizing hormone/human chorionic gonadotropin receptor gene. Mol Endocrinol 15:184–200
Lew DJ, Kornbluth S (1996) Regulatory roles of cyclin dependent kinase phosphorylation in cell cycle control. Curr Opin Cell Biol 8:795–804
Li R, Norman RJ, Armstrong DT, Gilchrist RB (2000) Oocyte-secreted factor(s) determine functional differences between bovine mural granulosa cells and cumulus cells. Biol Reprod 63:839–845
Li J, Kawamura K, Cheng Y et al (2010) Activation of dormant ovarian follicles to generate mature eggs. Proc Natl Acad Sci U S A 107:10280–10284
Lincoln AJ, Wickramasinghe D, Stein P et al (2002) Cdc25b phosphatase is required for resumption of meiosis during oocyte maturation. Nat Genet 30:446–449
Liu M, Sims D, Calarco P, Talbot P (2003) Biochemical heterogeneity, migration, and pre-fertilization release of mouse oocyte cortical granules. Reprod Biol Endocrinol 1:77
Liu K, Rajareddy S, Liu L, Jagarlamudi K et al (2006) Control of mammalian oocyte growth and early follicular development by the oocyte PI3 kinase pathway: new roles for an old timer. Dev Biol 299:1–11
Liu X, Xie F, Zamah AM et al (2014) Multiple pathways mediate luteinizing hormone regulation of cGMP signaling in the mouse ovarian follicle. Biol Reprod 91:9
Liu C, Peng J, Matzuk MM, Yao HH (2015) Lineage specification of ovarian theca cells requires multicellular interactions via oocyte and granulosa cells. Nat Commun 6:6934
Lodde V, Modina S, Maddox-Hyttel P et al (2008) Oocyte morphology and transcriptional silencing in relation to chromatin remodeling during the final phases of bovine oocyte growth. Mol Reprod Dev 75:915–924
Luciano AM, Franciosi F, Modina SC, Lodde V (2011) Gap junction-mediated communications regulate chromatin remodeling during bovine oocyte growth and differentiation through cAMP-dependent mechanism(s). Biol Reprod 85:1252–1259
Lund SA, Murdoch J, Van Kirk EA, Murdoch WJ (1999) Mitogenic and antioxidant mechanisms of estradiol action in preovulatory ovine follicles: relevance to luteal function. Biol Reprod 61:388–392
Macaulay AD, Gilbert I, Caballero J et al (2014) The gametic synapse: RNA transfer to the bovine oocyte. Biol Reprod 91:90
Malki S, van der Heijden GW, O'Donnell KA et al (2014) A role for retrotransposon LINE-1 in fetal oocyte attrition in mice. Dev Cell 29:521–533

Manova K, Huang EJ, Angeles M et al (1993) The expression pattern of the c-kit ligand in gonads of mice supports a role for the c-kit receptor in oocyte growth and in proliferation of spermatogonia. Dev Biol 157:85–99
Mattson BA, Albertini DF (1990) Oogenesis: chromatin and microtubule dynamics during meiotic prophase. Mol Reprod Dev 25:374–383
Mazerbourg S, Klein C, Roh J et al (2004) Growth differentiation factor-9 signaling is mediated by the type I receptor, activin receptor-like kinase 5. Mol Endocrinol 18:653–665
McArthur ME, Irving-Rodgers HF, Byers S, Rodgers RJ (2000) Identification and immunolocalization of decorin, versican, perlecan, nidogen, and chondroitin sulfate proteoglycans in bovine small-antral ovarian follicles. Biol Reprod 63:913–924
McGee EA, Sawetawan C, Bird I et al (1996) The effect of insulin and insulin-like growth factors on the expression of steroidogenic enzymes in a human ovarian thecal-like tumor cell model. Fertil Steril 65:87–93
Mehlmann LM, Terasaki M, Jaffe LA, Kline D (1995) Reorganization of the endoplasmic reticulum during meiotic maturation of the mouse oocyte. Dev Biol 170:607–615
Mehlmann LM, Jones TL, Jaffe LA (2002) Meiotic arrest in the mouse follicle maintained by a Gs protein in the oocyte. Science 297:1343–1345
Mehlmann LM, Saeki Y, Tanaka S et al (2004) The Gs-linked receptor GPR3 maintains meiotic arrest in mammalian oocytes. Science 306:1947–1950
Meinecke B, Meinecke-Tillmann S (1979) Effects of gonadotropins on oocyte maturation and progesterone production by porcine ovarian follicles cultured in vitro. Theriogenology 11:351–365
Miller WL (1988) Molecular biology of steroid hormone synthesis. Endocr Rev 9:295–318
Monniaux D, Pisselet C (1992) Control of proliferation and differentiation of ovine granulosa cells by insulin-like growth factor-I and follicle-stimulating hormone in vitro. Biol Reprod 46:109–119
Moore RK, Otsuka F, Shimasaki S (2003) Molecular basis of bone morphogenetic protein-15 signaling in granulosa cells. J Biol Chem 278:304–310
Nagyova E, Prochazka R, Vanderhyden BC (1999) Oocytectomy does not influence synthesis of hyaluronic acid by pig cumulus cells: retention of hyaluronic acid after insulin-like growth factor-I treatment in serum-free medium. Biol Reprod 61:569–574
Nakamura T, Iwase A, Bayasula B et al (2015) CYP51A1 induced by growth differentiation factor 9 and follicle-stimulating hormone in granulosa cells is a possible predictor for unfertilization. Reprod Sci 22:377–384
Neal P, Baker TG (1975) Response of mouse graafian follicles in organ culture to varying doses of follicle-stimulating hormone and luteinizing hormone. J Endocrinol 65:27–32
Nekola MV, Nalbandov AV (1971) Morphological changes of rat follicular cells as influenced by oocytes. Biol Reprod 4:154–160
Nicosia SV, Wolf DP, Inoue M (1977) Cortical granule distribution and cell surface characteristics in mouse eggs. Dev Biol 57:56–74
Nilsson E, Skinner MK (2001) Cellular interactions that control primordial follicle development and folliculogenesis. J Soc Gynecol Investig 8:S17–S20
Nilsson EE, Detzel C, Skinner MK (2006) Platelet-derived growth factor modulates the primordial to primary follicle transition. Reproduction 131:1007–1015
Norris RP, Ratzan WJ, Freudzon M et al (2009) Cyclic GMP from the surrounding somatic cells regulates cyclic AMP and meiosis in the mouse oocyte. Development 136:1869–1878
Norris RP, Freudzon M, Nikolaev VO, Jaffe LA (2010) Epidermal growth factor receptor kinase activity is required for gap junction closure and for part of the decrease in ovarian follicle cGMP in response to LH. Reproduction 140:655–662
Oatley JM, Brinster RL (2012) The germline stem cell niche unit in mammalian testes. Physiol Rev 92:577–595

Ochsner SA, Day AJ, Rugg MS et al (2003a) Disrupted function of tumor necrosis factor-alpha-stimulated gene 6 blocks cumulus cell-oocyte complex expansion. Endocrinology 144:4376–4384

Ochsner SA, Russell DL, Day AJ et al (2003b) Decreased expression of tumor necrosis factor-alpha-stimulated gene 6 in cumulus cells of the cyclooxygenase-2 and EP2 null mice. Endocrinology 144:1008–1019

Oh JS, Han SJ, Conti M (2010) Wee1B, Myt1, and Cdc25 function in distinct compartments of the mouse oocyte to control meiotic resumption. J Cell Biol 188:199–207

Olsiewski PJ, Beers WH (1983) cAMP synthesis in the rat oocyte. Dev Biol 100:287–293

Packer AI, Hsu YC, Besmer P, Bachvarova RF (1994) The ligand of the c-kit receptor promotes oocyte growth. Dev Biol 161:194–205

Pangas SA, Choi Y, Ballow DJ et al (2006) Oogenesis requires germ cell-specific transcriptional regulators Sohlh1 and Lhx8. Proc Natl Acad Sci U S A 103:8090–8095

Parfenov V, Potchukalina G, Dudina L et al (1989) Human antral follicles: oocyte nucleus and the karyosphere formation (electron microscopic and autoradiographic data). Gamete Res 22:219–231

Park JY, Su YQ, Ariga M et al (2004) EGF-like growth factors as mediators of LH action in the ovulatory follicle. Science 303:682–684

Parker KL, Schimmer BP (1995) Transcriptional regulation of the genes encoding the cytochrome P-450 steroid hydroxylases. Vitam Horm 51:339–370

Parrott JA, Skinner MK (1999) Kit-ligand/stem cell factor induces primordial follicle development and initiates folliculogenesis. Endocrinology 140:4262–4271

Payne C, Schatten G (2003) Golgi dynamics during meiosis are distinct from mitosis and are coupled to endoplasmic reticulum dynamics until fertilization. Dev Biol 264:50–63

Paynton BV, Bachvarova R (1994) Polyadenylation and deadenylation of maternal mRNAs during oocyte growth and maturation in the mouse. Mol Reprod Dev 37:172–180

Pelland AM, Corbett HE, Baltz JM (2009) Amino Acid transport mechanisms in mouse oocytes during growth and meiotic maturation. Biol Reprod 81:1041–1054

Pepling ME (2012) Follicular assembly: mechanisms of action. Reproduction 143:139–149

Pepling ME, Spradling AC (2001) Mouse ovarian germ cell cysts undergo programmed breakdown to form primordial follicles. Dev Biol 234:339–351

Pescador N, Houde A, Stocco DM, Murphy BD (1997) Follicle-stimulating hormone and intracellular second messengers regulate steroidogenic acute regulatory protein messenger ribonucleic acid in luteinized porcine granulosa cells. Biol Reprod 57:660–668

Pesce M, Di Carlo A, De Felici M (1997) The c-kit receptor is involved in the adhesion of mouse primordial germ cells to somatic cells in culture. Mech Dev 68:37–44

Peter M, Dubuis JM (2000) Transcription factors as regulators of steroidogenic P-450 enzymes. Eur J Clin Invest 30(Suppl 3):14–20

Piquette GN, LaPolt PS, Oikawa M, Hsueh AJ (1991) Regulation of luteinizing hormone receptor messenger ribonucleic acid levels by gonadotropins, growth factors, and gonadotropin-releasing hormone in cultured rat granulosa cells. Endocrinology 128:2449–2456

Pirino G, Wescott MP, Donovan PJ (2009) Protein kinase A regulates resumption of meiosis by phosphorylation of Cdc25B in mammalian oocytes. Cell Cycle 8:665–670

Pratt HP (1982) Preimplantation mouse embryos synthesize membrane sterols. Dev Biol 89:101–110

Prochazka R, Nagyova E, Rimkevicova Z et al (1991) Lack of effect of oocytectomy on expansion of the porcine cumulus. J Reprod Fertil 93:569–576

Prochazka R, Nagyova E, Brem G et al (1998) Secretion of cumulus expansion-enabling factor (CEEF) in porcine follicles. Mol Reprod Dev 49:141–149

Quirk SM, Cowan RG, Harman RM (2006) The susceptibility of granulosa cells to apoptosis is influenced by oestradiol and the cell cycle. J Endocrinol 189:441–453

Rajah R, Glaser EM, Hirshfield AN (1992) The changing architecture of the neonatal rat ovary during histogenesis. Dev Dyn 194:177–192

Ralph JH, Telfer EE, Wilmut I (1995) Bovine cumulus cell expansion does not depend on the presence of an oocyte secreted factor. Mol Reprod Dev 42:248–253
Reddy P, Shen L, Ren C et al (2005) Activation of Akt (PKB) and suppression of FKHRL1 in mouse and rat oocytes by stem cell factor during follicular activation and development. Dev Biol 281:160–170
Reddy P, Liu L, Adhikari D et al (2008) Oocyte-specific deletion of Pten causes premature activation of the primordial follicle pool. Science 319:611–613
Ren Y, Suzuki H, Jagarlamudi K et al (2015) Lhx8 regulates primordial follicle activation and postnatal folliculogenesis. BMC Biol 13:39
Richani D, Wang X, Zeng HT et al (2014) Pre-maturation with cAMP modulators in conjunction with EGF-like peptides during in vitro maturation enhances mouse oocyte developmental competence. Mol Reprod Dev 81:422–435
Richard S, Baltz JM (2014) Prophase I arrest of mouse oocytes mediated by natriuretic peptide precursor C requires GJA1 (connexin-43) and GJA4 (connexin-37) gap junctions in the antral follicle and cumulus-oocyte complex. Biol Reprod 90:137
Richard FJ, Tsafriri A, Conti M (2001) Role of phosphodiesterase type 3A in rat oocyte maturation. Biol Reprod 65:1444–1451
Richards JS (1980) Maturation of ovarian follicles: actions and interactions of pituitary and ovarian hormones on follicular cell differentiation. Physiol Rev 60:51–89
Richards JS (1994) Hormonal control of gene expression in the ovary. Endocr Rev 15:725–751
Richards JS, Jahnsen T, Hedin L et al (1987) Ovarian follicular development: from physiology to molecular biology. Recent Prog Horm Res 43:231–276
Richards JS, Russell DL, Ochsner S et al (2002) Novel signaling pathways that control ovarian follicular development, ovulation, and luteinization. Recent Prog Horm Res 57:195–220
Robinson JW, Zhang M, Shuhaibar LC et al (2012) Luteinizing hormone reduces the activity of the NPR2 guanylyl cyclase in mouse ovarian follicles, contributing to the cyclic GMP decrease that promotes resumption of meiosis in oocytes. Dev Biol 366:308–316
Robker RL, Richards JS (1998) Hormone-induced proliferation and differentiation of granulosa cells: a coordinated balance of the cell cycle regulators cyclin D2 and p27Kip1. Mol Endocrinol 12:924–940
Rosenfeld CS, Wagner JS, Roberts RM, Lubahn DB (2001) Intraovarian actions of oestrogen. Reproduction 122:215–226
Salustri A, Ulisse S, Yanagishita M, Hascall VC (1990) Hyaluronic acid synthesis by mural granulosa cells and cumulus cells in vitro is selectively stimulated by a factor produced by oocytes and by transforming growth factor-beta. J Biol Chem 265:19517–19523
Salustri A, Camaioni A, Di Giacomo M et al (1999) Hyaluronan and proteoglycans in ovarian follicles. Hum Reprod Update 5:293–301
Salustri A, Garlanda C, Hirsch E et al (2004) PTX3 plays a key role in the organization of the cumulus oophorus extracellular matrix and in in vivo fertilization. Development 131:1577–1586
Sanggaard KW, Sonne-Schmidt CS, Krogager TP et al (2008) The transfer of heavy chains from bikunin proteins to hyaluronan requires both TSG-6 and HC2. J Biol Chem 283:18530–18537
Sasseville M, Ritter LJ, Nguyen TM et al (2010) Growth differentiation factor 9 signaling requires ERK1/2 activity in mouse granulosa and cumulus cells. J Cell Sci 123:3166–3176
Sato H, Kajikawa S, Kuroda S et al (2001) Impaired fertility in female mice lacking urinary trypsin inhibitor. Biochem Biophys Res Commun 281:1154–1160
Sato N, Kawamura K, Fukuda J et al (2003) Expression of LDL receptor and uptake of LDL in mouse preimplantation embryos. Mol Cell Endocrinol 202:191–194
Scarchilli L, Camaioni A, Bottazzi B et al (2007) PTX3 interacts with inter-alpha-trypsin inhibitor: implications for hyaluronan organization and cumulus oophorus expansion. J Biol Chem 282:30161–30170
Schindler R, Nilsson E, Skinner MK (2010) Induction of ovarian primordial follicle assembly by connective tissue growth factor CTGF. PLoS One 5:e12979

Schmidt D, Ovitt CE, Anlag K et al (2004) The murine winged-helix transcription factor Foxl2 is required for granulosa cell differentiation and ovary maintenance. Development 131:933–942
Schoppee PD, Garmey JC, Veldhuis JD (2002) Putative activation of the peroxisome proliferator-activated receptor gamma impairs androgen and enhances progesterone biosynthesis in primary cultures of porcine theca cells. Biol Reprod 66:190–198
Schroeder TE (1981) Microfilament-mediated surface change in starfish oocytes in response to 1-methyladenine: implications for identifying the pathway and receptor sites for maturation-inducing hormones. J Cell Biol 90:362–371
Schultz RM, Letourneau GE, Wassarman PM (1979) Program of early development in the mammal: changes in patterns and absolute rates of tubulin and total protein synthesis during oogenesis and early embryogenesis in the mouse. Dev Biol 68:341–359
Schultz RM, Montgomery RR, Belanoff JR (1983) Regulation of mouse oocyte meiotic maturation: implication of a decrease in oocyte cAMP and protein dephosphorylation in commitment to resume meiosis. Dev Biol 97:264–273
Segaloff DL, Wang HY, Richards JS (1990) Hormonal regulation of luteinizing hormone/chorionic gonadotropin receptor mRNA in rat ovarian cells during follicular development and luteinization. Mol Endocrinol 4:1856–1865
Sekar N, Garmey JC, Veldhuis JD (2000) Mechanisms underlying the steroidogenic synergy of insulin and luteinizing hormone in porcine granulosa cells: joint amplification of pivotal sterol-regulatory genes encoding the low-density lipoprotein (LDL) receptor, steroidogenic acute regulatory (stAR) protein and cytochrome P450 side-chain cleavage (P450scc) enzyme. Mol Cell Endocrinol 159:25–35
Shitsukawa K, Andersen CB, Richard FJ et al (2001) Cloning and characterization of the cyclic guanosine monophosphate-inhibited phosphodiesterase PDE3A expressed in mouse oocyte. Biol Reprod 65:188–196
Shuhaibar LC, Egbert JR, Norris RP et al (2015) Intercellular signaling via cyclic GMP diffusion through gap junctions restarts meiosis in mouse ovarian follicles. Proc Natl Acad Sci U S A 112:5527–5532
Silva JM, Price CA (2002) Insulin and IGF-I are necessary for FSH-induced cytochrome P450 aromatase but not cytochrome P450 side-chain cleavage gene expression in oestrogenic bovine granulosa cells in vitro. J Endocrinol 174:499–507
Simon AM, Goodenough DA, Li E, Paul DL (1997) Female infertility in mice lacking connexin 37. Nature 385:525–529
Simone DA, Chorich LP, Mahesh VB (1993) Mechanisms of action for an androgen-mediated autoregulatory process in rat thecal-interstitial cells. Biol Reprod 49:1190–1201
Simpson ER, Mahendroo MS, Means GD et al (1994) Aromatase cytochrome P450, the enzyme responsible for estrogen biosynthesis. Endocr Rev 15:342–355
Sirois J, Simmons DL, Richards JS (1992) Hormonal regulation of messenger ribonucleic acid encoding a novel isoform of prostaglandin endoperoxide H synthase in rat preovulatory follicles. Induction in vivo and in vitro. J Biol Chem 267:11586–11592
Smyth CD, Gosden RG, McNeilly AS, Hillier SG (1994) Effect of inhibin immunoneutralization on steroidogenesis in rat ovarian follicles in vitro. J Endocrinol 140:437–443
Sorensen RA, Wassarman PM (1976) Relationship between growth and meiotic maturation of the mouse oocyte. Dev Biol 50:531–536
Spicer LJ, Chamberlain CS, Maciel SM (2002) Influence of gonadotropins on insulin- and insulin-like growth factor-I (IGF-I)-induced steroid production by bovine granulosa cells. Domest Anim Endocrinol 22:237–254
Spicer LJ, Aad PY, Allen DT et al (2008) Growth differentiation factor 9 (GDF9) stimulates proliferation and inhibits steroidogenesis by bovine theca cells: influence of follicle size on responses to GDF9. Biol Reprod 78:243–253
Sretarugsa P, Wallace RA (1997) The developing Xenopus oocyte specifies the type of gonadotropin-stimulated steroidogenesis performed by its associated follicle cells. Dev Growth Differ 39:87–97

Sternlicht AL, Schultz RM (1981) Biochemical studies of mammalian oogenesis: kinetics of accumulation of total and poly(A)-containing RNA during growth of the mouse oocyte. J Exp Zool 215:191–200

Stocco DM (2001) Tracking the role of a star in the sky of the new millennium. Mol Endocrinol 15:1245–1254

Su YQ, Denegre JM, Wigglesworth K et al (2003) Oocyte-dependent activation of mitogen-activated protein kinase (ERK1/2) in cumulus cells is required for the maturation of the mouse oocyte-cumulus cell complex. Dev Biol 263:126–138

Su YQ, Sugiura K, Woo Y et al (2007) Selective degradation of transcripts during meiotic maturation of mouse oocytes. Dev Biol 302:104–117

Su YQ, Sugiura K, Wigglesworth K et al (2008) Oocyte regulation of metabolic cooperativity between mouse cumulus cells and oocytes: BMP15 and GDF9 control cholesterol biosynthesis in cumulus cells. Development 135:111–121

Su YQ, Sugiura K, Li Q et al (2010) Mouse oocytes enable LH-induced maturation of the cumulus-oocyte complex via promoting EGF receptor-dependent signaling. Mol Endocrinol 24:1230–1239

Sugiura K, Pendola FL, Eppig JJ (2005) Oocyte control of metabolic cooperativity between oocytes and companion granulosa cells: energy metabolism. Dev Biol 279:20–30

Sugiura K, Su YQ, Diaz FJ et al (2007) Oocyte-derived BMP15 and FGFs cooperate to promote glycolysis in cumulus cells. Development 134:2593–2603

Sugiura K, Su YQ, Eppig JJ (2009) Targeted suppression of Has2 mRNA in mouse cumulus cell-oocyte complexes by adenovirus-mediated short-hairpin RNA expression. Mol Reprod Dev 76:537–547

Suzuki T, Sasano H, Takaya R et al (1998) Cyclic changes of vasculature and vascular phenotypes in normal human ovaries. Hum Reprod 13:953–959

Thomas FH, Ismail RS, Jiang JY, Vanderhyden BC (2008) Kit ligand 2 promotes murine oocyte growth in vitro. Biol Reprod 78:167–175

Tian XC, Berndtson AK, Fortune JE (1995) Differentiation of bovine preovulatory follicles during the follicular phase is associated with increases in messenger ribonucleic acid for cytochrome P450 side-chain cleavage, 3 beta-hydroxysteroid dehydrogenase, and P450 17 alpha-hydroxylase, but not P450 aromatase. Endocrinology 136:5102–5110

Toda K, Takeda K, Okada T et al (2001) Targeted disruption of the aromatase P450 gene (Cyp19) in mice and their ovarian and uterine responses to 17beta-oestradiol. J Endocrinol 170:99–111

Trigatti B, Rayburn H, Vinals M et al (1999) Influence of the high density lipoprotein receptor SR-BI on reproductive and cardiovascular pathophysiology. Proc Natl Acad Sci U S A 96:9322–9327

Tsafriri A, Chun SY, Zhang R et al (1996) Oocyte maturation involves compartmentalization and opposing changes of cAMP levels in follicular somatic and germ cells: studies using selective phosphodiesterase inhibitors. Dev Biol 178:393–402

Tsutsumi O, Yano T, Satoh K et al (1990) Studies of hexokinase activity in human and mouse oocyte. Am J Obstet Gynecol 162:1301–1304

Tsutsumi O, Satoh K, Taketani Y, Kato T (1992) Determination of enzyme activities of energy metabolism in the maturing rat oocyte. Mol Reprod Dev 33:333–337

Uda M, Ottolenghi C, Crisponi L et al (2004) Foxl2 disruption causes mouse ovarian failure by pervasive blockage of follicle development. Hum Mol Genet 13:1171–1181

Urner F, Herrmann WL, Baulieu EE, Schorderet-Slatkine S (1983) Inhibition of denuded mouse oocyte meiotic maturation by forskolin, an activator of adenylate cyclase. Endocrinology 113:1170–1172

Van Blerkom J, Davis PW, Lee J (1995) ATP content of human oocytes and developmental potential and outcome after in-vitro fertilization and embryo transfer. Hum Reprod 10:415–424

Van Blerkom J, Davis P, Alexander S (2003) Inner mitochondrial membrane potential (DeltaPsim), cytoplasmic ATP content and free Ca2+ levels in metaphase II mouse oocytes. Hum Reprod 18:2429–2440

Vanderhyden BC (1993) Species differences in the regulation of cumulus expansion by an oocyte-secreted factor(s). J Reprod Fertil 98:219–227
Vanderhyden BC, Tonary AM (1995) Differential regulation of progesterone and estradiol production by mouse cumulus and mural granulosa cells by A factor(s) secreted by the oocyte. Biol Reprod 53:1243–1250
Vanderhyden BC, Caron PJ, Buccione R, Eppig JJ (1990) Developmental pattern of the secretion of cumulus expansion-enabling factor by mouse oocytes and the role of oocytes in promoting granulosa cell differentiation. Dev Biol 140:307–317
Vanderhyden BC, Cohen JN, Morley P (1993) Mouse oocytes regulate granulosa cell steroidogenesis. Endocrinology 133:423–426
Vanderhyden BC, Macdonald EA, Nagyova E, Dhawan A (2003) Evaluation of members of the TGFbeta superfamily as candidates for the oocyte factors that control mouse cumulus expansion and steroidogenesis. Reprod Suppl 61:55–70
Van Soom A, Tanghe S, De Pauw I et al (2002) Function of the cumulus oophorus before and during mammalian fertilization. Reprod Domest Anim 37:144–151
Varnosfaderani ShR, Ostadhosseini S, Hajian M et al (2013) Importance of the GDF9 signaling pathway on cumulus cell expansion and oocyte competency in sheep. Theriogenology 80:470–478
Vitt UA, McGee EA, Hayashi M, Hsueh AJ (2000) In vivo treatment with GDF-9 stimulates primordial and primary follicle progression and theca cell marker CYP17 in ovaries of immature rats. Endocrinology 141:3814–3820
Vivarelli E, Conti M, De Felici M, Siracusa G (1983) Meiotic resumption and intracellular cAMP levels in mouse oocytes treated with compounds which act on cAMP metabolism. Cell Differ 12:271–276
Wallace WH, Kelsey TW (2010) Human ovarian reserve from conception to the menopause. PLoS One 5:e8772
Walters KA, Middleton LJ, Joseph SR et al (2012) Targeted loss of androgen receptor signaling in murine granulosa cells of preantral and antral follicles causes female subfertility. Biol Reprod 87:151
Wang Q, Chi MM, Schedl T, Moley KH (2012) An intercellular pathway for glucose transport into mouse oocytes. Am J Physiol Endocrinol Metab 302:E1511–E1518
Watson LN, Mottershead DG, Dunning KR et al (2012) Heparan sulfate proteoglycans regulate responses to oocyte paracrine signals in ovarian follicle morphogenesis. Endocrinology 153:4544–4555
Whitelaw PF, Smyth CD, Howles CM, Hillier SG (1992) Cell-specific expression of aromatase and LH receptor mRNAs in rat ovary. J Mol Endocrinol 9:309–312
Wickramasinghe D, Ebert KM, Albertini DF (1991) Meiotic competence acquisition is associated with the appearance of M-phase characteristics in growing mouse oocytes. Dev Biol 143:162–172
Wigglesworth K, Lee KB, O'Brien MJ et al (2013) Bidirectional communication between oocytes and ovarian follicular somatic cells is required for meiotic arrest of mammalian oocytes. Proc Natl Acad Sci U S A 110:E3723–E3729
Wigglesworth K, Lee KB, Emori C et al (2015) Transcriptomic diversification of developing cumulus and mural granulosa cells in mouse ovarian follicles. Biol Reprod 92:23
Willis D, Mason H, Gilling-Smith C, Franks S (1996) Modulation by insulin of follicle-stimulating hormone and luteinizing hormone actions in human granulosa cells of normal and polycystic ovaries. J Clin Endocrinol Metab 81:302–309
Worrad DM, Ram PT, Schultz RM (1994) Regulation of gene expression in the mouse oocyte and early preimplantation embryo: developmental changes in Sp1 and TATA box-binding protein, TBP. Development 120:2347–2357
Yamamoto K, Otoi T, Koyama N et al (1999) Development to live young from bovine small oocytes after growth, maturation and fertilization in vitro. Theriogenology 52:81–89

Yoshida H, Takakura N, Kataoka H et al (1997) Stepwise requirement of c-kit tyrosine kinase in mouse ovarian follicle development. Dev Biol 184:122–137

Yoshimura Y (1998) Insulin-like growth factors and ovarian physiology. J Obstet Gynaecol Res 24:305–323

Yoshimura Y, Tischkau SA, Bahr JM (1994) Destruction of the germinal disc region of an immature preovulatory follicle suppresses follicular maturation and ovulation. Biol Reprod 51:229–233

Yoshioka S, Ochsner S, Russell DL et al (2000) Expression of tumor necrosis factor-stimulated gene-6 in the rat ovary in response to an ovulatory dose of gonadotropin. Endocrinology 141:4114–4119

Zeleznik AJ, Midgley AR Jr, Reichert LE Jr (1974) Granulosa cell maturation in the rat: increased binding of human chorionic gonadotropin following treatment with follicle-stimulating hormone in vivo. Endocrinology 95:818–825

Zhang G, Garmey JC, Veldhuis JD (2000) Interactive stimulation by luteinizing hormone and insulin of the steroidogenic acute regulatory (StAR) protein and 17alpha-hydroxylase/17,20-lyase (CYP17) genes in porcine theca cells. Endocrinology 141:2735–2742

Zhang X, Wu XQ, Lu S et al (2006) Deficit of mitochondria-derived ATP during oxidative stress impairs mouse MII oocyte spindles. Cell Res 16:841–850

Zhang Y, Zhang Z, Xu XY et al (2008) Protein kinase A modulates Cdc25B activity during meiotic resumption of mouse oocytes. Dev Dyn 237:3777–3786

Zhang M, Su YQ, Sugiura K, Xia G, Eppig JJ (2010) Granulosa cell ligand NPPC and its receptor NPR2 maintain meiotic arrest in mouse oocytes. Science 330:366–369

Zhang M, Su YQ, Sugiura K et al (2011) Estradiol promotes and maintains cumulus cell expression of natriuretic peptide receptor 2 (NPR2) and meiotic arrest in mouse oocytes in vitro. Endocrinology 152:4377–4385

Zhang H, Liu L, Li X et al (2014a) Life-long in vivo cell-lineage tracing shows that no oogenesis originates from putative germline stem cells in adult mice. Proc Natl Acad Sci U S A 111:17983–17988

Zhang H, Risal S, Gorre N et al (2014b) Somatic cells initiate primordial follicle activation and govern the development of dormant oocytes in mice. Curr Biol 24:2501–2508

Zhang H, Panula S, Petropoulos S et al (2015) Adult human and mouse ovaries lack DDX4-expressing functional oogonial stem cells. Nat Med 21:1116–1118

Zuccotti M, Ponce RH, Boiani M et al (2002) The analysis of chromatin organisation allows selection of mouse antral oocytes competent for development to blastocyst. Zygote 10:73–78

Zuelke KA, Brackett BG (1992) Effects of luteinizing hormone on glucose metabolism in cumulus-enclosed bovine oocytes matured in vitro. Endocrinology 131:2690–2696