CHAPTER

8

Sertoli Cell Secreted Regulatory Factors

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I. INTRODUCTION

The Sertoli cell provides the microenvironment and cytoarchitectural support for the developing spermatogenic cells and directly regulates the reproductive endocrinology of the male. This is in large part accomplished through the secretion of a wide variety of proteins. These secretory products include transport proteins to provide nutrient support to the germ cells, extracellular matrix and cell adhesion molecules to promote appropriate cell–cell interactions, and proteases to allow tissue remodeling during spermatogenesis. These Sertoli cell secreted factors can be categorized as nutritional factors (e.g., transport proteins) that support the nutrient requirements of the germ cells, environmental factors (e.g., extracellular matrix) that influence the physical content and extracellular environment between cells, and regulatory factors (e.g., growth factors) [1].

Regulatory factors are defined as factors that, through receptor-mediated signal transduction events, influence cellular function, growth, or differentiation on a molecular level. These regulatory factors can act as autocrine agents to influence Sertoli cells or paracrine factors to influence neighboring cells such as spermatogenic cells, Leydig cells, or peritubular myoid cells [1]. These regulatory factors are essential for the careful control of testis development, spermatogenesis and male fertility, and male reproductive endocrinology. Two major subcategories of regulatory factors are growth factors and hormones.

Growth factors are defined as factors that influence cell proliferation and tissue growth. Although growth factors directly influence the cell cycle, growth factors can also have effects on a variety of other cellular functions and cell differentiation. Properties of a variety of growth factors are presented in Table 8.1. All the growth factors listed have been shown to be Sertoli cell secreted factors and are discussed in more detail later. These Sertoli cell secreted regulatory factors are all required for normal testis function and male fertility.

Another subcategory of Sertoli cell secreted regulatory factors is hormones. The local actions of hormone-like factors are often required to directly influence cellular function and differentiation. In contrast to growth factors, hormones generally do not directly influence cell proliferation. However, indirect effects mediated through the production of a growth factor do occur. These hormones are also needed to act as an endocrine agent at distant tissue and organs to regulate the reproductive endocrinology of the male. The positive and negative feedback systems built into endocrine regulation require the local production of hormones. Sertoli cell secreted hormones are discussed later.

TABLE 8.1 Properties of Several Common Growth Factors

Growth factor	Abbreviation	Approximate size (kDa)	Examples of physiological action	Receptor(s)
Insulin-like growth factor I	IGF-I	7.5	Skeletal growth	IGF-I receptor
Insulin-like growth factor II	IGF-II	7.5	Fetal development	IGF-I and IGF-II
Epidermal growth factor	EGF	6	Tissue growth	EGF receptor
Transforming growth factor α	TGFα	5	Tissue growth	EGF receptor
Transforming growth factor β	TGFβ	25/dimer	Growth inhibition/tissue repair	TGFβ, type 1, 2, and 3 receptors
Fibroblast growth factor	FGF	17	Angiogenesis/tissue growth	FGF receptor
Neurotropins	NT-3	13	Neuronal development	TrkC receptor
Interleukin 1	IL-1	17	Immune response/inflammation	IL-1 receptor
Kit ligand/stem cell factor	KL/SCF	30	Tissue growth/germ cells	KL receptor
Glial cell–derived neurotrophic factor	GDNF	34	Cell growth	Ret/GFRα receptor

The initial review of this topic was published more than 10 years ago in *The Sertoli Cell* [2]. The advances since that time have been significant and will be the focus of the current review. Both Sertoli cell secreted growth factors and hormones are discussed. These secreted factors play a critical role in the development, growth, and maintenance of testis function. Sertoli cells are the principal cell regulating the process of spermatogenesis and these secreted regulatory factors are essential for Sertoli cell function.

II. GROWTH FACTORS

The development of the testis and maintenance of spermatogenesis require a precise growth regulation [3, 4]. All cell populations proliferate during embryonic testis development and in the early postnatal period. Sertoli cells become postmitotic and terminally differentiate during the early pubertal period once spermatogenesis is initiated [3–5]. The other somatic cells in the testis (e.g., Leydig, peritubular) have a slowed but continuous rate of growth in the adult [4–8]. Spermatogenic cells require a rapid rate of mitosis and meiosis at the onset of puberty that continues throughout adult life. The waves of spermatogenic cell proliferation require local control by Sertoli cells. The secretion of growth factors by Sertoli cells will have a role in the precise cell growth regulation required in the developing and adult testis.

A. Insulin-Like Growth Factors

Growth factors with structural similarity to insulin are in a family of factors termed insulin-like growth factors (IGFs). These include IGF-I and IGF-II. IGF-I is a critical factor required for cell cycle progression and DNA synthesis in all proliferating cells. For this reason IGF-I is expressed and acts on nearly all cells. The serum level of IGF-I is high (e.g., 100 ng/mL) and is a principal liver product [9]. A family of IGF binding proteins exists to regulate the level of IGF bioactivity available to a cell and influence the homeostasis of the extracellular growth factors.

All testicular somatic cells express and respond to IGF-I including Sertoli cells [10–13]. IGF-I influences DNA synthesis and increases lactate and transferrin production by Sertoli cells [14, 15]. IGF-I also can influence the actions of FSH on Sertoli cells [16, 17] through unique signaling events [17]. Although the IGF-I produced by Sertoli cells [10–13] can act as an autocrine factor on the Sertoli cells, the high concentration of IGF-I in the interstitial fluid is available to the basal surface of the Sertoli cell. The blood–testis barrier created by Sertoli cells sequesters the developing spermatogenic cells. IGF-I available in the interstitium does not pass this barrier. Therefore, IGF-I secreted by Sertoli cells will likely have a critical role as a paracrine factor on germ cells (Fig. 8.1). Germ cells appear to be a site for IGF-I actions [18, 19]. Local production of IGF binding proteins by Sertoli cells has been shown to influence IGF-I actions and will contribute to this proposed Sertoli cell–germ cell interaction [12, 20, 21]. The ability of IGF-I produced by Sertoli cells to influence Leydig cell function has also been proposed [22]. Although these interactions can be observed *in vitro*, the physiological importance *in vivo* is questioned due to the levels of IGF-I and binding proteins in the interstitium [23]. It is likely Sertoli cell secreted IGF-I will be critical for spermatogenic cells, but actions on other

testis somatic cells is questioned due to other sources of IGF-I for these cells.

IGF-II has also been shown to mediate Sertoli cell–spermatogenic cell interactions [24–27]. The IGF-II receptor is present on both Sertoli cells and germ cells [25]. This receptor can also influence gene expression in the spermatogenic cells [26]. The IGF-II receptor is also termed the mannose-6-phosphate receptor and can bind a variety of additional ligands with appropriate carbohydrate specificity. Therefore, this IGF-II receptor system appears to mediate interactions between Sertoli cells and germ cells through a variety of potential ligands [27].

B. Transforming Growth Factor α

Transforming growth factor α (TGFα) is a member of the epidermal growth factor (EGF) family and binds to the same EGF receptor as EGF [28, 29]. TGFα is expressed as a transmembrane precursor that is proteolytically processed to release the mature peptide (Table 8.1). TGFα is secreted by a large number of normal cell types and promotes cell proliferation in normal and transformed cells.

EGF has been shown to influence spermatogenesis [30], and removal of serum levels of EGF reduces sperm numbers [31]. Circulating concentrations of EGF, however, are thought to be too low to have physiological influences [32]. EGF is not expressed in the rodent testis or in Sertoli cells [33]. In contrast, TGFα is expressed by Sertoli cells [33]. Both peritubular myoid cells and Leydig cells also produce TGFα [33, 34]. Peritubular cells and Leydig cells proliferate in response to TGFα [33]. TGFα has been shown to influence Sertoli cell lactate and estrogen production [35] and can influence Sertoli cell DNA synthesis if the cells are derived from prepubertal animals [36, 37]. TGFα also influences transferrin production by Sertoli cells [38]. The actions of TGFα on Sertoli cells and the testis are dependent on the developmental stage. Embryonic testis growth requires TGFα expression and action on most cell types [4]. At the onset of puberty, the Sertoli cells differentiate and become postmitotic. Sertoli cell responsiveness to TGFα/EGF declines as the adult stage of development is obtained [39]. Other cell types continue to utilize TGFα for growth regulation. The spermatogenic cells also utilize TGFα, and Sertoli cell–derived TGFα is postulated to be an important paracrine interaction of this Sertoli cell regulatory factor [40] (Fig. 8.1). Sertoli production of TGFα is also capable of influencing adjacent peritubular cells [39, 40].

C. Transforming Growth Factor β

Transforming growth factor β (TGFβ) has three mammalian isoforms ($TGF\beta_1$, $TGF\beta_2$, and $TGF\beta_3$) that act as inhibitors of cell proliferation and facilitate cellular differentiation in a wide variety of cells [41]. TGFβ acts by inhibiting the actions of growth stimulators (e.g., TGFα) and promoting the expression of extracellular matrix components. Most cell types contain the receptors for TGFβ isoforms. Although found in transformed cells, TGFβ regulates most normal cell types [41].

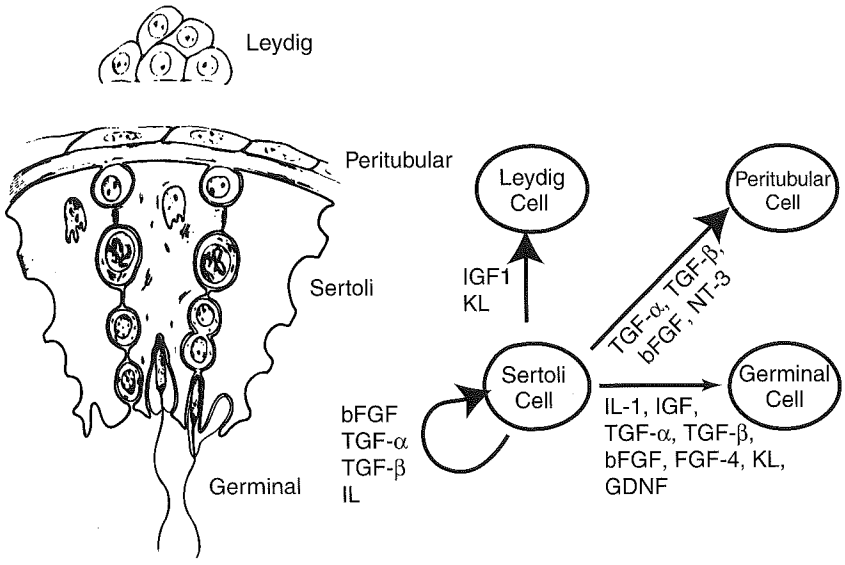

FIGURE 8.1. Sertoli cell secreted regulatory factors.

TGFβ isoforms are expressed by Sertoli cells [40, 42, 43] and Sertoli cells are one of the few tissues that express TGFβ3 [40, 44]. Sertoli cells express all three isoforms under distinct hormonal regulation [40, 45]. The developmental regulation of TGFβ expression in the testis of the different isoforms suggests distinct functions and/or differential regulation [45, 46]. TGFβ expression by Sertoli cells is critical for embryonic testis development [46, 47]. At the onset of puberty, TGFβ expression by Sertoli cells also corresponds to alterations in cell growth [45]. $TGF\beta_1$ is high prepubertally and then declines to low levels in the adult. $TGF\beta_2$ expression is similar and FSH appears to promote the decrease in expression by Sertoli cells [45]. $TGF\beta_3$ has a transient increase in expression at the onset of puberty [45].

The site of action for Sertoli cell secreted TGFβ is suggested from the analysis of TGFβ receptor localization. All cell types in the testis contain type I and II TGFβ receptors [48–50], including spermatogenic cells. Peritubular cells produce and respond to TGFβ with a decline in cellular proliferation induced by TGFα [42, 45]. Peritubular cells also produce more extracellular matrix components in response to TGFβ [42]. TGFβ also induce peritubular cell contractility that is needed for sperm transport in the seminiferous tubule [51]. TGFβ does not have major effects on Sertoli cell proliferation or functional genes, but does increase production of plasminogen activator [52]. TGFβ also appears to influence the tight junctions between Sertoli cells [53, 54]. Leydig cells are also a site of TGFβ production and action. TGFβ inhibits Leydig cell steroidogenesis [55–57]. Therefore, critical targets for the Sertoli cell secreted TGFβ isoforms are spermatogenic cells and peritubular cells (Fig. 8.1). Abnormalities in TGFβ expression have been suggested as causal in testicular diseases such as fibrosis [58]. The bioactivity of TGFβ produced by Sertoli cells is also a factor in the function of TGFβ [59]. TGFβ also can be inhibited by specific proteins such as Bambi [60], which can influence the actions of the Sertoli cell expressed TGFβ. TGFβ is a critical regulatory factor produced by Sertoli cells to influence testis development, function, and spermatogenesis.

D. Fibroblast Growth Factor

Several members of the fibroblast growth factor (FGF) family have been shown to influence testis function. FGF family members regulate cellular growth and differentiation, as well as tissue angiogenesis [61]. FGFs have widespread tissue distribution and each of the family members has distinct receptors and specific functions.

Basic FGF (bFGF) is one of the most studied FGF family members. Initially bFGF was identified in whole testis [62, 63] and subsequently was found to be expressed by Sertoli cells [64, 65]. bFGF production by Sertoli cells was found to be increased by FSH [65]. bFGF has been shown to stimulate Sertoli cell growth prepubertally [66]. The ability of FSH to simulate Sertoli cell growth prepubertally may in part be indirectly mediated through bFGF production [65]. Early postnatal actions of FGF also suggest potential indirect effects on gonocyte proliferation [67]. Sertoli cells also respond to bFGF by an increase in the production of proteoglycans [68], lactate, and glucose metabolism [69]. Localization of the bFGF receptor (FGFR type 1) demonstrated receptors in Sertoli cells, Leydig cells, peritubular cells, and germ cells [70]. Therefore, the actions of bFGF secretion by Sertoli cells could mediate cellular interactions with spermatogenic cells and peritubular cells, as well as autocrine actions on Sertoli cells (Fig. 8.1).

Another member of the FGF family, Hst-1/FGF-4, has been shown to be expressed by Sertoli cells [71]. A conditional knockout of FGF-4 resulted in impaired fertility and a knockin caused enhanced spermatogenesis [72]. This observation suggests a potential Sertoli cell–spermatogenic cell interaction mediated by FGF-4 (Fig. 8.1). The overexpression of FGF-4 was also found to protect the testis from the chemotherapeutic drug adriamycin [72]. Therefore, Sertoli cell expression of FGF-4 appears to be important for normal testis function.

FGF-9 is another member of the FGF family that has been shown to be important for early embryonic testis development [73]. FGF-9 appears to be a downstream gene to Sry and is expressed by Sertoli cells to influence male sex differentiation through the adjacent mesenchymal tissue. A knockout of FGF-9 results in sex reversal and impaired testis development [73]. Functions of FGF-9 later in development have not been thoroughly investigated.

E. Neurotropins

The neurotropin family of growth factors includes nerve growth factor (NGF), neurotropin 3 (NT-3), neurotropin 4/5 (NT-4/5), and brain-derived neurotrophic factor (BDNF). The high-affinity receptors for these factors are the trk receptors and the low-affinity ones are the LNGFR. Although these neurotropins are critical for neurons and associated cells, they also have been shown to be expressed by a number of nonneuronal tissues including the testis.

NGF has been the most highly investigated neurotropin in the testis. NGF appears in the adult testis to primarily be expressed by spermatogenic cells [74–76], whereas the NGF receptor trkA is expressed in Sertoli cells [76–78]. The NGF precursor protein is highly expressed in round spermatids, while the NGF acts to

promote Sertoli cell viability [76] and cellular functions such as lactate and estrogen production [78]. Although some studies have suggested NGF expression in the adult Sertoli cell [79], the majority have suggested NGF mediates a spermatogenic cell–Sertoli cell interaction and adult Sertoli cells do not express NGF [76–78, 80]. In contrast to adult Sertoli cells, NGF does appear to be expressed by late embryonic and early postnatal Sertoli cells [81]. The NGF receptor (trkA) was expressed in Sertoli cells, peritubular cells, and interstitial cells at these stages of development [81]. Therefore, NGF is a Sertoli cell secreted growth factor, but its expression by Sertoli cells appears to be more important in the perinatal period than in the adult.

Other neurotropins expressed in the adult testis have not been extensively studied. NT-3 was found to localize in spermatogenic cells, whereas BDNF may be expressed by adult Sertoli cells [82]. In contrast, BDNF is not expressed in the embryonic or postnatal testis [81]. More thorough investigation of other neurotropins is needed to clarify roles in the adult testis. Observations suggest that Sertoli cell expressed neurotropins play a critical role during development, but in the adult, spermatogenic cells influence Sertoli cells through neurotropin production.

NT-3 appears to be a critical neurotropin secreted by Sertoli cells during the initial stages of male sex determination and testis development [81, 83]. At the time of Sertoli cell fate differentiation by the testis determining factor Sry, Sertoli cells express NT-3 as a potential immediate downstream gene to Sry [81, 83]. This Sertoli cell secreted NT-3 acts on adjacent mesonephros cells as a chemotactic factor and promotes migration into the testis, and the migrating mesonephros cells become peritubular cells and promote seminiferous cord formation, which is the first morphological event in testis development [81, 83, 84]. The NT-3 receptor trkC is on the migrating mesonephros cells and blocking the actions of this receptor inhibits cord formation and testis development [83]. This has also been recently confirmed in the human fetal testis [85]. NT-3 also can act as a direct chemotactic agent to induce mesonephros migration into the developing testis [84]. Observations suggest Sertoli cell secretion of NT-3 is a downstream event to Sry and critical to early embryonic testis development by acting on early developing peritubular cells. In contrast, adult Sertoli cells do not appear to express NT-3, such that this is primarily an embryonic function of Sertoli cells.

F. Cytokines

Cytokines are defined as growth factors involved in immune cell communication and the immune system. However, these proteins also influence nonimmune cells and tissues. A number of different cytokines that have been shown to have functions in the testis are outlined next.

The interleukins (ILs) are a family of cytokines produced by activated lymphocytes and macrophages. Interleukin 1 beta (IL-1β) is secreted by lymphocytes, whereas interleukin 1 alpha (IL-1α) is produced by a wide variety of nonimmune tissues. IL-1α has been shown to be expressed by Sertoli cells [86] and appears to influence spermatogenic cells [87]. The expression of IL-1α by Sertoli cells appears dependent on the presence of germ cells [88]. IL-1α can directly influence prepubertal Sertoli cell growth [89], transferrin gene expression [90], and lactate production [91]. Interestingly, Sertoli cells also express an antagonist form of IL-1α termed IL-1ra [92, 93]. These positive and negative acting forms of IL-1α are proposed to have a role in regulating cellular interactions between Sertoli cells and germ cells [92–94]. In contrast to IL-1α, IL-1β is not expressed by Sertoli cells [92, 93]. However, IL-1β can induce the expression of IL-1α by Sertoli cells [95].

In addition to IL-1α, Sertoli cells produce interleukin 6 (IL-6) in response to the autocrine actions of IL-1α [96–98]. Residual bodies induce Sertoli cell expression of IL-1α that in an autocrine manner induces Sertoli cell production of IL-6. Analysis of the hormonal regulation of IL-1 and IL-6 demonstrates distinct regulation [99] and secretion into different tubule compartments [100]. Although autocrine actions of IL-6 on Sertoli cells have been postulated [101, 102], an important function of Sertoli cell secreted interleukins is communication with the immune cells in the interstitial compartment [103, 104]. Leydig cells also produce these interleukins, probably for a similar function [105, 106].

Other cytokines produced by Sertoli cells include interferon α (IFN-α) [107] and α_2-macroglobulin (α_2-MG) [108]. Both are proposed to interact with immune cells in the interstitium and possibly mediate other cell–cell interactions. Macrophage populations in the interstitium also are targets for macrophage migration inhibiting factor (MIF), which is primarily produced by Leydig cells [109, 110], but under appropriate conditions is also produced by Sertoli cells [109]. These and other cytokines [111] are likely to promote critical interactions with immune cells and Sertoli cell secreted cytokines will have a role in these cellular interactions.

G. Kit Ligand/Stem Cell Factor

Kit ligand (KL), also known as stem cell factor (SCF), acts at the kit receptor and is known to be important

for primordial germ cell viability and cell migration to the embryonic gonad. KL is required for normal germ cell proliferation and fertility of both the male and female. Sertoli cells are known to secrete KL and the spermatogonia express the kit receptor [112, 113]. Inhibition of KL reduces spermatogenesis and specifically the early mitotic events of germ cell development. Therefore, KL produced by Sertoli cells is needed in the embryo for germ cell survival and proliferation, early postnatally for gonocyte development [114], and in the adult for spermatogonia proliferation. This is a classic Sertoli cell–spermatogenic cell paracrine interaction in that the growth factor is produced only by one cell and receptor is in the target germ cell (Fig. 8.1). Gonadotropins regulate the expression of KL by Sertoli cells and various agents such as gonadotropin-releasing hormone (GnRH) [115], growth-hormone–releasing hormone (GHRH) [116], and somatostatin [117] can influence KL production. The physiological importance of this Sertoli cell secreted factor is shown in forms of male infertility. Impaired expression of kit [118] and mutated forms of KL [119] result in subfertility in human infertility patients. In addition to the Sertoli cell produced KL acting on spermatogonia, Leydig cells have been shown to potentially be a target during development or regeneration of the testis [120, 121].

H. Glial Cell–Derived Neurotrophic Factor

Glial cell–derived neurotrophic factor (GDNF) is a member of the TGFβ superfamily and neurotropin family. GDNF has been shown to be secreted by Sertoli cells and acts on spermatogonia through the ret/GFR receptor specifically located on spermatogonia [122]. GDNF appears to promote spermatogonia stem cells to initiate development [122, 123]. Spermatogonia cell proliferation is influenced by GDNF [124, 125]. A related growth factor, neurturin, also has similar functions [125]. Therefore, similar to KL, GDNF is secreted by Sertoli cells to act on early-stage spermatogonia to promote cell proliferation and development (Fig. 8.1). Disruption of GDNF expression alters spermatogenesis [126] and overexpression of GDNF causes a nonmetastatic germ cell tumor by hyperproliferation of spermatogonia [127]. Although GDNF can influence Sertoli cell proliferation prepubertally, the spermatogonia cell is the primary target in the adult [128].

III. HORMONES

Sertoli cells are endocrine cells in that they respond to hormones (e.g., FSH and androgen) and also produce hormones. In addition to the secretion of growth factors by Sertoli cells, hormones are also important regulatory factors secreted. The major hormones produced by Sertoli cells are reviewed next.

A. Inhibin/Activin

More than 70 years ago, Sertoli cells were shown to produce a regulatory agent termed inhibin [129] that inhibits FSH production by the pituitary [130, 131]. The two inhibin gene product subunits α and β form a dimer for inhibin, whereas a β homodimer produces activin that can stimulate FSH production. Both inhibin and activin are now known to have a wide variety of biological functions and are produced by a number of tissues. Sertoli cells under the control of FSH produce inhibin [132–135]. Although the major role of inhibin is to feed back information to the pituitary to regulate FSH production, local actions within the testis are also known [136, 137]. Sertoli cells can respond to activin to influence postnatal Sertoli cell proliferation [138]. Inhibin may influence germ cells [139, 140] and germ cells can influence the expression of inhibin by Sertoli cells [141]. The cellular interactions mediated by inhibin and activin in the testis are complicated by their expression by multiple cell types. Leydig cells can produce both inhibin and activin [142–145]. Recently, activin has been shown to be produced by peritubular cells and may regulate Sertoli cells through a paracrine interaction [146, 147]. Therefore, the specific paracrine and autocrine roles for inhibin and activin remain to be fully elucidated. However, the endocrine roles for these factors are better established. Abnormalities in inhibin levels correlating with abnormal pubertal development and testicular dysfunction have been documented [148–150].

B. Müllerian Inhibiting Substance

Müllerian inhibiting substance (MIS), also termed anti-Müllerian hormone (AMH), is secreted by embryonic Sertoli cells and promotes the regression of the Müllerian duct and female reproductive tract development [151, 152]. MIS was first identified in the fetal and neonatal testes [153] and was subsequently cloned and found to be expressed by Sertoli cells [154–156]. MIS is a TGFβ family member. MIS expression is induced upon male sex determination by Sertoli cells and then declines to negligible levels in the adult [157, 158]. FSH inhibits the expression of MIS correlating with the decline during pubertal testis development [159]. MIS acts at specific receptors [151] to promote regression of the Müllerian duct and to regulate androgen production by Leydig cells [160]. The regulation of MIS expression has been examined on the transcriptional level

and demonstrated that the SF1, GATA-4, and Sox8 transcription factors regulate the MIS promoter [161–163]. FSH, thyroid hormone, and androgen can regulate the expression of MIS in Sertoli cells [164, 165]. MIS/AMH appears to be a Sertoli cell secretory factor that has a critical role during embryonic development as an endocrine hormone. Genetic mutations in MIS or its receptor cause human mutations in premature developmental defects, persistent Müllerian ducts, and azoospermia [166–168].

C. Estrogen

Sertoli cells express aromatase prepubertally so they can utilize androgen production by Leydig cells to produce estrogen. As Sertoli cells differentiate, aromatase expression drops such that negligible estrogen is produced in the adult testis. The knockout null mutations in the estrogen receptor (ER) demonstrated alterations in spermatogenesis and infertility [169]. The phenotype was found to be primarily mediated through rat testis and efferent duct abnormalities [170, 171]. However, estrogen production by Sertoli cells appears to also have direct effects on testis function and spermatogenesis [172]. Localization of the ERα demonstrated expression in Leydig cells and spermatogonic cells [173], whereas ERβ was found in Sertoli cells in the adult stage and in the fetal stage in Sertoli, Leydig, peritubular, and germ cells [174]. Estrogen can act on Sertoli cells and influence FSH actions [175]. As stated, FSH inhibits estrogen production and IGF-1 can alter these effects [16]. Sertoli cell secretion of estrogen appears to be primarily a role for the prepubertal period and as the testis develops in response to FSH, estrogen production becomes negligible. Estrogen may have a role in suppressing prepubertal androgen produced by Leydig cells and may influence germ cells at later stages. Specific roles for estrogen in the adult remain to be elucidated.

IV. OTHER REGULATORY FACTORS

A. Uncharacterized Factors

Several regulatory factors have been postulated to be produced by Sertoli cells and have specific roles in mediating interactions with other cell types. These factors have not been characterized beyond their biological activity. The first was seminiferous growth factor (SGF), which had mitogenic activity and was distinct from FGF and stimulated proliferation of several Sertoli cell lines [176–178]. The second was Sertoli cell secreted growth factor (SCSGF), which had some similarities to TGFα and also stimulated the growth of several Sertoli cell lines [179]. These factors may well be some of the factors described earlier and have similar biological activities.

A number of studies have suggested that Sertoli cells secrete a factor(s) that influences Leydig cell function and steroidogenesis. Abnormal physiological and environmental insults that affect Sertoli cell function have been shown to indirectly influence Leydig cell function [180–192]. Sertoli cell conditioned medium has been used to identify factors that stimulate cell proliferation and suppress steroidogenesis [193]. The evidence for such an activity is very convincing, but the factor(s) have not been characterized. Again a number of the Sertoli cell secreted factors mentioned earlier have this activity on Leydig cells (e.g., TGFα, bFGF).

Sertoli cells have also been shown to produce factors such as erythropoietin [194], leukemia inhibitory factor (LIF), and ciliary neurotrophic factor (CNTF) [195], but the specific expression and actions of these factors needs to be assessed. Recently, the genomic microarray analysis of Sertoli cell gene expression has suggested the expression of a number of secreted regulatory factors [196]. Examination of these factors primarily provided the same list as that discussed earlier. It is likely that new factors secreted by Sertoli cells will be determined, but a more genomic approach should facilitate the identification and initial detection versus the more classic protein biochemistry approaches used in the past.

B. Lipids

In addition to protein regulatory agents, nonprotein factors such as lipids are also found to have dramatic biological activities. Although many lipids have been primarily localized within the cell, some lipids such as lysophospholipids and thromboxanes are soluble and are known to bind specific G-protein coupled receptors on the surface of cells [197, 198]. These lipid-induced receptor-mediated events alter cellular functions in a similar manner to protein growth factors [197]. The testis has unique forms of metabolic enzymes generating these lipids [199–201], such that lipid factors may be secreted by Sertoli cells. Sertoli cells have been shown to produce soluble sphingosine molecules and other lysophospholipids [202–204]. The specific lipids secreted and functions remain to be elucidated, but due to the important functions of these extracellular soluble lipids it is likely they will be important regulators of testis function and spermatogenesis.

V. SUMMARY

The secretion of regulatory factors by Sertoli cells is critical for normal testis development and function. Abnormalities in the expression or action of these factors often result in infertility or endocrine defects. As shown in Figure 8.1 and Table 8.2, a number of growth factors are secreted by Sertoli cells to influence spermatogenic cells. Because Sertoli cells are the principal regulatory cells for spermatogenesis, it is not surprising that many of the growth factor's primary targets are germ cells. Since the original review of this topic, several major observations have been made regarding Sertoli cell secreted growth factors [2]. Knowledge that the production of KL and GDNF by Sertoli cells directly regulates spermatogonia cell proliferation and development represents a significant advance in our understanding of the control of the initiation of the spermatogenic wave and regulation of the spermatogenic stem cell population. Recently, human male infertility conditions have been linked to abnormalities in the expression and action of these factors. Other specific factors that have the characteristics of the growth factor being expressed by Sertoli cells and actions only on a specific stage of germ cells are likely to be identified. The number of growth factors listed in Table 8.2 will likely grow further during the next decade as well. The microenvironment within the testis and local cell–cell interactions mediated by regulatory factors such as those listed in Table 8.2 require elucidation for us to understand how hormones regulate testis function and how therapeutic treatments for male infertility can be developed.

TABLE 8.2 Sertoli Cell Secreted Growth Factors

Growth factor	Site of action	Function
Insulin-like growth factor I (IGF-I)	Germ cells Sertoli cells Leydig cells	Cell cycle progression and growth (i.e., S-phase DNA synthesis)
Insulin-like growth factor II (IGF-II)	Germ cells	Cell–cell signaling
Transforming growth factor α (TGFα)	Germ cells Sertoli cells Peritubular cells	Cell growth initiation
Transforming growth factor β (TGFβ)	Germ cells Sertoli cells Peritubular cells	Growth inhibition and increase in cell differentiated function
Basic fibroblast growth factor (bFGF)	Germ cells Sertoli cells Peritubular cells	Cell growth initiation
Neurotropin 3 (NT-3)	Peritubular cells	Cell migration (i.e., mesonephros) and growth
Interleukin 1 and 6 (IL-1 and IL-6)	Germ cells	Cell–cell signaling
Kit ligand/stem cell factor (KL/SCF)	Germ cells Leydig cells	Cell growth (i.e., spermatogonial)
Glial cell–derived neurotrophic factor (GDNF)	Germ cells	Cell growth (i.e., spermatogonial stem cells)

TABLE 8.3 Sertoli Cell Secreted Hormones

Hormone	Site of action	Function
Inhibin	Pituitary	Reduce FSH production
Müllerian inhibiting substance/anti-Müllerian hormone (MIS/AMH)	Müllerian duct	Regress female reproductive tract development
Estrogen	Leydig cells	Reduce steroidogenesis and cell–cell signaling

The Sertoli cells also secrete several critical hormones, as listed in Table 8.3. Sertoli cells are critical to male reproductive endocrinology because they are a target for several hormones (e.g., FSH and testosterone) and they produce hormones. Feedback pathways are always required in a biological process and endocrine system, so it is not surprising that a critical inhibitory feedback protein like inhibin is produced. In addition, the negative feedback estrogen provides prepubertally to suppress Leydig cell androgen production is also anticipated. The information that has developed during the past decade [2] indicates that these factors also have other roles in mediating regulatory interactions between cells within the testis. Although the specific functions of the local actions of the inhibin, activin, and estrogen need to be elucidated, clearly they have local paracrine functions independent of the endocrine role of these factors. This will likely be an intense area of research for a number of years.

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