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Chapter

Sertoli Cell Phagocytosis: An Essential Event for Spermatogenesis

Fei Wang and Daishu Han

Abstract

During spermatogenesis, most male germ cells undergo apoptosis, and the cytoplasmic portions of the elongating spermatids are shed as residual bodies (RB). Both apoptotic germ cells (AGC) and RB must be phagocytosed by Sertoli cells, which are essential to maintain testicular homeostasis for normal spermatogenesis. The phagocytosis of AGC and RB by Sertoli cells confers various meanings, including elimination of apoptotic components, removal of autoantigens, and the recycle of degenerated substrates as an energy source. Sertoli cell phagocytosis can be regulated by various mechanisms. The impairment of Sertoli cell phagocytosis may disrupt tissue homeostasis in the testis, thereby impairing to testicular function and spermatogenesis. This chapter discusses the mechanisms underlying phagocytic removal of AGC and RB by Sertoli cells and the consequences of this biological event for spermatogenesis and male fertility.

Keywords: Sertoli cell, phagocytosis, spermatogenesis, apoptosis, male fertility

1. Introduction

Mammalian spermatogenesis is a cell-organized differentiation process of male germ cells in the testis. This process includes spermatogonium mitosis, spermatocyte meiosis, and spermatid morphogenesis. Throughout spermatogenesis, Sertoli cells tightly embrace differentiating germ cells in the seminiferous epithelium and create a microenvironment essential for germ cell differentiation. In addition to physical support, Sertoli cells provide nutrition for developing germ cells and take up apoptotic components. During spermatogenesis, most of male germ cells undergo apoptosis, and those that finalize differentiation process will shed their most cytoplasmic components as residual bodies (RB). Apoptotic germ cells (AGC) and RB must be timely eliminated by Sertoli cells via phagocytosis.

The phagocytic elimination of AGC and RB by Sertoli cells has been proposed to contribute to spermatogenesis in several ways: (1) reducing space competition for enormous male germ cells to finalize differentiation process, (2) preventing noxious cellular contents that may be released by necrosis of apoptotic germ cells, (3) removing autoantigens that may induce an autoimmune response, and (4) recycling of AGC and RB as an energy source for Sertoli cells.

Various mechanisms are involved in the regulation of Sertoli cell phagocytosis of AGC and RB. The interaction of class B scavenger receptor type I (SR-BI) expressed

on phagocytes and phosphatidylserine (PS) exposed on apoptotic cell surfaces is a universal mechanism by which phagocytes engulf apoptotic cells [1]. This mechanism is also involved in the regulation of the phagocytosis of AGC and RB by Sertoli cells [2]. Tyro3, Axl, and Mer (TAM) tyrosine kinase receptors and their functional common ligand, growth arrest specific gene 6 (Gas6), are also essential for optimal phagocytosis of AGC by Sertoli cells. Several other genes that regulate Sertoli cell phagocytosis of AGC have been recognized. The mechanisms underlying phagocytic removal of AGC by Sertoli cells are the main focus of this article.

Impairment of Sertoli cell phagocytosis is associated with pathogenesis and dysfunction of the testis, thus impairing male fertility. The inhibition of Sertoli cell phagocytic ability disrupts spermatogenesis [3]. Gene mutation that impairs Sertoli cell phagocytosis may lead to autoimmune orchitis and male infertility [4]. The pathogenic conditions due to impaired Sertoli cell phagocytosis are mentioned in the text.

2. Germ cell apoptosis and Sertoli cell phagocytosis

The mammalian testis consists of two distinct compartments: the seminiferous tubules and the interstitial spaces among the tubules (**Figure 1**). The two major functions of the testis include spermatogenesis, occurring within the seminiferous epithelium, and steroidogenesis by Leydig cells that are in the interstitial spaces.

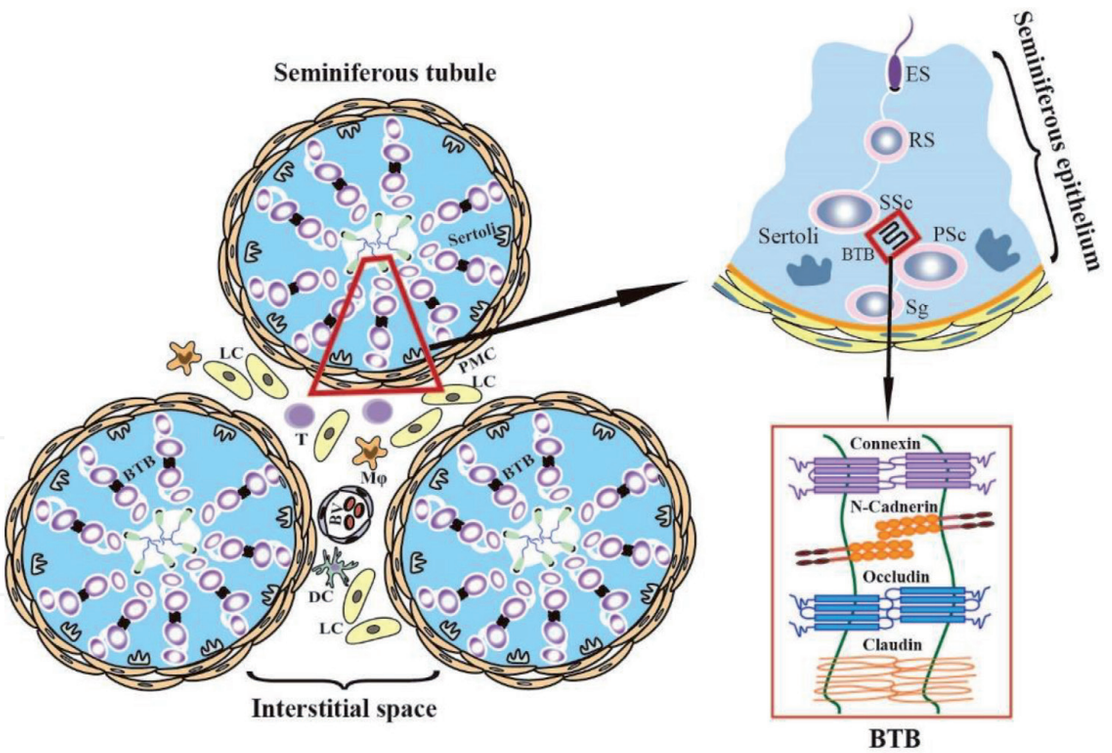

Figure 1.
Human testicular schematic of histological structure and cellular constituents. The testis consists of two separate regions, namely, seminiferous tubules and interstitial spaces (left panels). The seminiferous tubules are composed of multiple layers of peritubular myoid cells (PMC) that constitute a tubular wall and Sertoli cells embracing different stages of male germ cells to form the seminiferous epithelium where spermatogenesis is fulfilled (right upper panel). The seminiferous epithelium is divided into two compartments, namely, the basal compartments and adluminal compartments, by the BTB that is formed by various junctions (right low panel) between two adjacent Sertoli cells, near the basal side. Different stages of developing germ cells, including spermatogonia (Sg), primary spermatocytes (PSc), secondary spermatocytes (SSc), round spermatids (RS), and elongated spermatids (ES), are localized on the seminiferous epithelium from the basal compartments to adluminal compartments. The interstitial spaces are composed of various cell types, the majority of which are Leydig cells (LC), but also macrophages (Mφ), as well as minor dendritic cells (DC) and T lymphocytes (T).

The seminiferous tubules possess a special microenvironment essential for spermatogenesis, which is composed of columnar Sertoli cells tightly encompassing developing germ cells. The blood-testis barrier (BTB) that is formed by two adjacent Sertoli cells near the basal side of the tubules is critical for maintaining the tissue homeostasis and immune microenvironment for normal germ cell development. During spermatogenesis, more than 75% germ cells have been estimated to undergo apoptosis [5, 6]. Apoptosis can occur at any stage of germ cells. Male germ cell survival and apoptosis are highly regulated by endocrine hormones [7]. In particular, follicle-stimulating hormone (FSH) produced by the pituitary and testosterone synthesized in Leydig cells is essential for healthy spermatogenesis. Low level of FSH increases germ cell apoptosis. The administration of testosterone in vivo inhibits germ cell apoptosis. Both FSH and testosterone could not act on germ cells because these cells do not express the receptors of two hormones. By contrast, FSH and testosterone can regulate the functions of Sertoli cells that express the hormonal receptors. Therefore, FSH and testosterone indirectly regulate germ cell apoptosis via Sertoli cells. The cascade of caspase activation is involved in the initiation of germ cell apoptosis [8]. Caspase 2 activation initiates the caspase cascade, in which BAX is involved in the cleavage of caspases.

Like other apoptotic cells, the translocation of phosphatidylserine (PS) to the surface of the cellular membrane is a characterization of male germ cell apoptosis [9]. The PS on the surface of apoptotic cells can be recognized by SR-BI and engulfed by phagocytes. At the final stage of germ cell development, most of the cytoplasm portions of spermatozoa are shed as RB before spermatozoa release to the lumen of the seminiferous tubules. However, immunohistochemical staining only detects a limited number of AGC. The RB are also rarely observed by histological analysis. These phenomena are assumed due to the rapid removal of apoptotic cells and RB through phagocytosis, a common way for engulfing apoptotic cells [1]. In accordance with this speculation, the inhibition of Sertoli cell phagocytosis in vivo greatly increases AGC numbers within the seminiferous tubules [3].

Phagocytosis of AGC and RB by Sertoli cells can be assessed by various approaches [10]. Confocal and transmission electron microscopy are reliable approaches that can distinguish apoptotic components ingested by Sertoli cells. However, these expensive and time-consuming approaches are not suitable for routine tests. Several simplified protocols to indirectly measure Sertoli cell phagocytosis have been reported [11–13]. These protocols require further optimization to avoid data misinterpretation. Lipid droplets are cyclically formed in the cytoplasm of Sertoli cells, which coincides with the spermatogenic cycle [14]. Therefore, it has been proposed that lipid droplets in Sertoli cells might result from the degradation of apoptotic components, including RB and AGC. An in vitro study confirmed that phagocytosis of AGC by Sertoli cells resulted in lipid droplet formation in Sertoli cells, which was used for evaluation of phagocytosis of AGC by Sertoli cells [13].

3. Mechanisms underlying phagocytosis of AGC and RB by Sertoli cells

3.1 Role of SR-BI/PS system

SR-BI is a receptor for high-density lipoprotein and can bind to acidic liposomes and apoptotic cells [15–17]. PS is a type of phospholipid that is located on the inner leaflet of the plasma membrane bilayer of healthy cells [18]. However, PS translocates to the outer leaflet of the cellular membrane during cell apoptosis and is exposed on the surface of apoptotic cells [9]. PS on the apoptotic cell surface can be recognized and bound by SR-BI on phagocytes, which is a key mechanism by

which phagocytes engulf apoptotic cells (**Figure 2**). The interaction of PS and SR-BI induces cytoskeletal changes that form phagocytic cup, thereby resulting in the engulfment of apoptotic cells. As shown in **Figure 3** (right side), SR-BI is expressed in Sertoli cells, and PS is exposed on the surfaces of AGC and RB [19–21]. Several in vitro studies provide evidence that Sertoli cells engulf AGC and RB through the interaction of SR-BI and PS. The phagocytosis of AGC and RB by Sertoli cells can be inhibited by the presence of annexin V that specifically binds to PS on the surfaces of AGC and RB [3, 21]. Moreover, an antibody against SR-BI disables the phagocytosis of AGC by Sertoli cells [19]. The SR-BI/PS-mediated phagocytosis of AGC and RB by Sertoli cells is confirmed in vivo, in which injection of anti-SR-BI antibody and annexin V into the seminiferous tubules increases the number of AGC [3]. Therefore, both in vitro and in vivo studies confirm that Sertoli cells recognize and engulf AGC and RB in the SR-BI/PS-dependent fusion.

3.2 Role of TAM receptors in mediating Sertoli cell phagocytosis

TAM receptors belong to a subfamily of the transmembrane receptor tyrosine kinases (**Figure 4**), which include three members, Tyro3, Axl, and Mer [22]. Gas6 is a functional common ligand of TAM receptors [23]. The TAM/Gas6 system regulates cell survival, innate immune response, and phagocytosis of apoptotic cells [24–27]. TAM receptors are involved in several pathological conditions, such as chronic inflammatory and autoimmune diseases [28, 29], viral infection [30–32], and cancer [33–35]. Notably, TAM receptors are essential for spermatogenesis and male fertility [36, 37].

The mechanisms by which the TAM/Gas6 system regulates testicular functions have been intensively investigated [38]. TAM receptors and Gas6 are abundantly expressed in Sertoli and Leydig cells [39]. All three Tyro3, Axl, and Mer receptors are expressed in Sertoli cells, whereas Leydig cells express Axl and Mer. Gas6 is uniquely expressed in Leydig cells. TAM receptors negatively regulate the expression of pro-inflammatory cytokines in both Sertoli and Leydig cells [40, 41], which might contribute to the immunoprivileged status of the testis [42]. In particular, the TAM receptors and Gas6 are essential for the phagocytic removal of AGC by Sertoli cells [43]. TAM receptors cooperatively regulate Sertoli cell phagocytosis of AGC. All three Tyro3, Axl, and Mer receptors participate in recognizing and binding AGC to Sertoli cells, whereas Mer is responsible for triggering phagocytic intracellular signaling that promotes engulfment of AGC. Any individual TAM receptors in Sertoli cells exhibit similar binding ability to AGC. However, Sertoli cells lacking all three TAM receptors remarkably decrease the binding between Sertoli cells and AGC. The TAM-mediated binding of Sertoli cells to AGC cannot be homologous

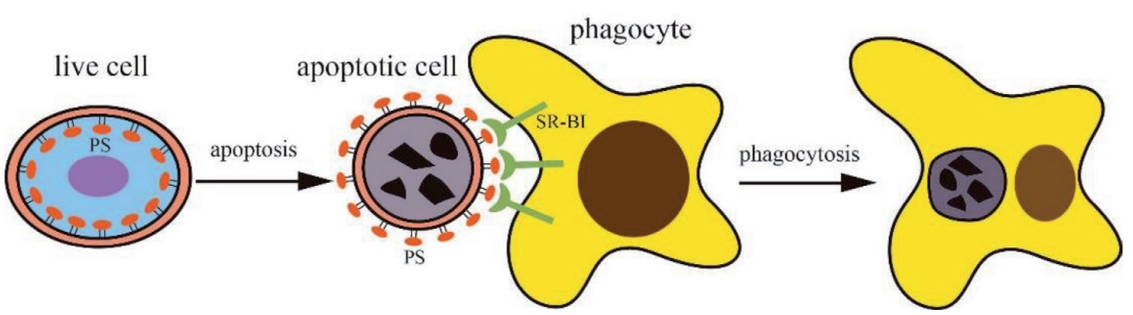

Figure 2.
SR-BI/PS-mediated phagocytosis. PS is translocated from the inner leaflet to outer leaflet of cellular membrane during apoptosis. PS is recognized by SR-BI located on the surface of phagocytes, and subsequently apoptotic cell is engulfed by phagocytosis via cytoskeletal changes.

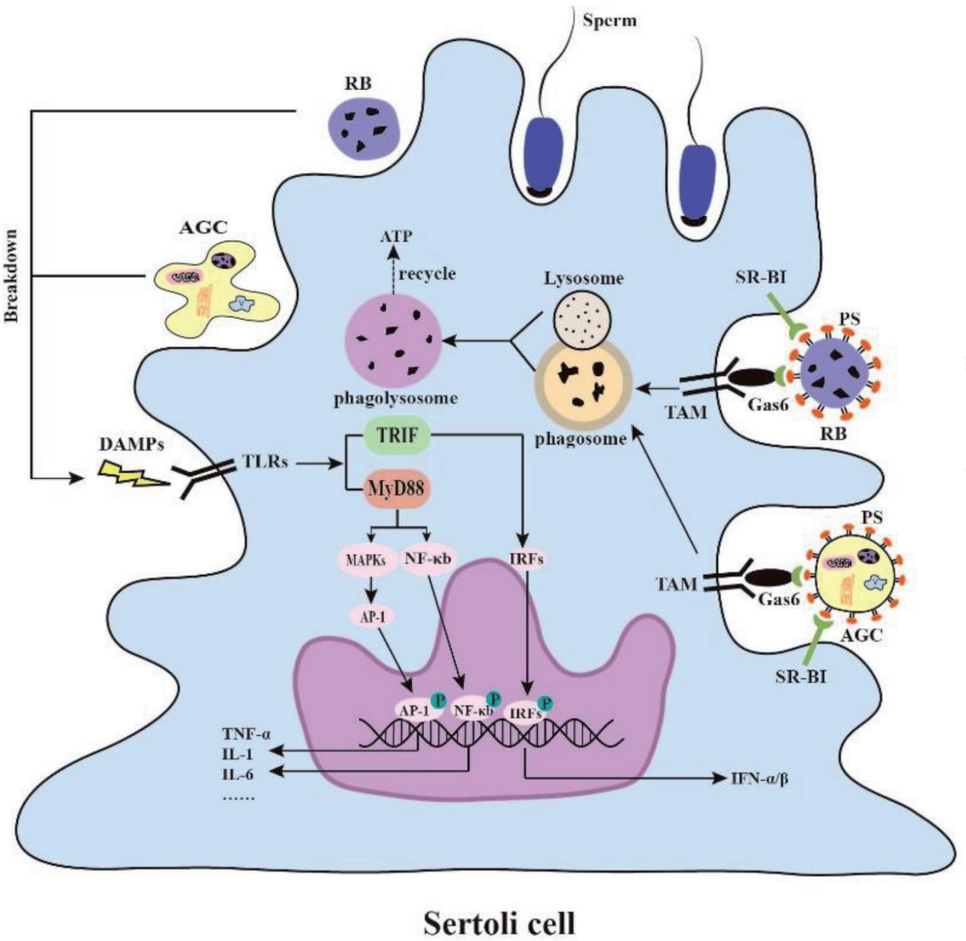

Figure 3.
Mechanisms and consequences of Sertoli cell phagocytosis of apoptotic germ cells (AGC) and residual bodies (RB). AGC and RB are phagocytized by Sertoli cells through two mechanisms (right side). SR-BI expressed on the Sertoli cell membrane binds with phosphatidylserine (PS) located on the surfaces of AGC and RB, thereby engulfing AGC and RB. TAM receptors mediate the phagocytosis of AGC and RB by Sertoli cells through Gas6 that bridges TAM receptors on Sertoli cell membrane and PS on the surfaces of AGC and RB. After phagocytosis, AGC and RB fuse with lysosomes and are recycled as energy sources for ATP production. If AGC and RB are not efficiently engulfed by Sertoli cells (left side), AGC and RB break down and release damage-associated molecular patterns (DAMPs). DAMPs can be recognized by toll-like receptors (TLRs) and initiate innate immune responses through TRIF and MyD88 signaling pathways. These pathways activate nuclear factor kappa B (NF-κB), mitosis antigen protein kinases (MAPKs), and interferon regulatory factors (IRFs), thereby inducing the expression of inflammatory cytokines, including TNF-α, IL-1, IL-6, INF-α, and IFN-β.

adhesion because germ cells do not express any TAM receptors. Gas6 is required for TAM-mediated phagocytosis of AGC by Sertoli cells. The N-terminal region of Gas6 binds to PS on the surface of AGC, and the C-terminal of Gas6 is recognized by TAM receptors, allowing Gas6 to bridge the binding between Sertoli cells and AGC (**Figure 3**, right side). Gas6 also plays a role in mediating Sertoli cell phagocytosis of AGC through the activation of Mer, thus triggering intracellular phagocytic signaling that modulates the cytoskeleton of Sertoli cells for engulfing AGC.

3.3 Other molecules regulating Sertoli cell phagocytosis

Sertoli cells abundantly express dynamin 2, and dynamin 2 is involved in the regulation of Sertoli cell phagocytosis [12]. Dynamin 2 regulates the actin assembly in Sertoli cells during phagocytosis. A dynamin 2 inhibitor reduces Sertoli cell phagocytosis through the impairment of phagocytic cup formation. Knockdown of dynamin 2 perturbs actin polymerization and recruitment to target liposomes. The role of dynamin 2 in regulating Sertoli cell phagocytosis requires the interaction between dynamin and amphiphysin 1 [44]. Dynamin 2 and amphiphysin 1

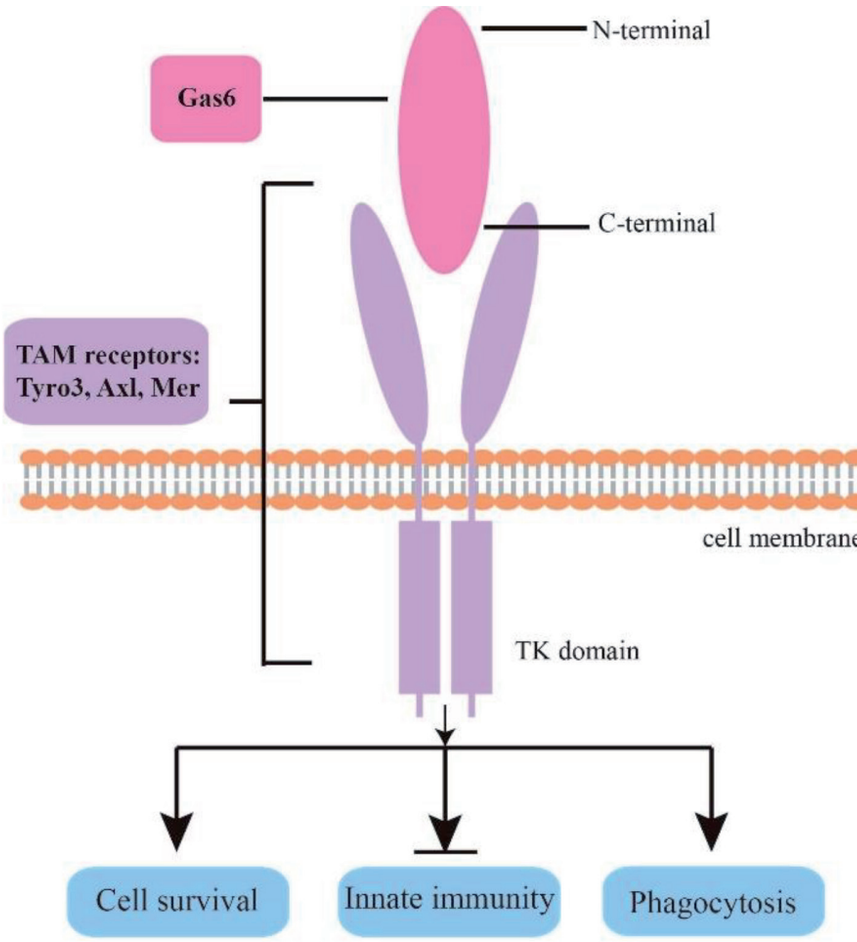

Figure 4.
TAM receptors and Gas6 system. TAM receptors belong to transmembrane proteins. The extracellular N-terminal region of TAM receptors binds to C-terminal domain of Gas6. The binding of Gas6 to TAM receptors results in the activation of intracellular tyrosine kinase (TK) domain of TAM receptors, thereby promoting (→) cell survival and phagocytosis of apoptotic cells and inhibiting (⊣) innate immunity.

can be specifically bound and simultaneously accumulated at ruffles of phagocytic cups. The interaction of dynamin 2 and amphiphysin 1 depends on the PS exposure on AGC.

Dimeric transferrin inhibits phagocytosis of RB by Sertoli cells in an autocrine manner [45]. Transferrin is a glycoprotein that transports iron and is highly expressed in Sertoli cells. Iron is essential for the inhibitory effect of transferrin on Sertoli cell phagocytosis. Transferrin can be physiologically secreted by Sertoli cells and inhibits the phagocytic removal of RB in autocrine manner.

ELMO1 is an evolutionarily conserved engulfment protein that mediates the internalization of apoptotic cells. However, ELMO1-deficient mice are viable and largely normal except for evident testicular pathology [46]. The seminiferous epithelium is disrupted, and AGC number is increased in the testis of ELMO1-deficient mice, therefore reducing sperm output. ELMO1 mediates the phagocytic removal of AGC by Sertoli cells. The engulfment receptors BAL1 and RAC1 (upstream and downstream of ELMO1, respectively) are involved in ELMO1-mediated Sertoli cell phagocytosis of AGC.

Noncoding miRNA regulates Sertoli cell phagocytosis. An early study showed that Dicer, a key enzyme that processes miRNA precursors into its functional form, is required for Sertoli cell function [47]. Dicer knockout mice are fetal lethal. The conditional Dicer knockout in Sertoli cells remarkably increases AGC numbers and leads to primary infertility, suggesting that miRNAs are involved in Sertoli

cell function and spermatogenesis in mice. Whether phagocytic ability of Sertoli cells is impaired by Dicer mutation remains unclear. However, the miR-471-5p has been recently identified to regulate phagocytosis of AGC by Sertoli cells [48]. The overexpression of miR-471-5p in Sertoli cells increases AGC number due to a defective phagocytic ability of Sertoli cells in transgenic mice. The role of miR-471-5p in regulating Sertoli cell phagocytosis requires its interaction with the autophagy protein LC3. Interestingly, androgen favors Sertoli cell phagocytosis by regulating the expression of miR-471-5p and its target proteins.

4. Pathophysiological meaning of Sertoli cell phagocytosis

The sperm production and testosterone synthesis are two major functions of testis. To fulfill these functions, the testis is highly organized, considering its anatomical location, histological structure, and cellular compositions. The testis is constituted by several types of tissue-specific cells. In addition to numerous germ cells, major testicular somatic cell types, including Leydig and Sertoli cells, are crucial for spermatogenesis. Leydig cells, localizing in the interstitial spaces of the testis, synthesize testosterone essential for spermatogenesis and multiple other extratesticular target organs. Sertoli cells embrace developing germ cells and constitute the seminiferous epithelium within the seminiferous tubules where spermatogenesis occurs (**Figure 1**). Sertoli cells are the only type of somatic cells in the seminiferous epithelium and play critical roles in regulating spermatogenesis by building a niche for germ cell development, providing nutrition to germ cells, and removing AGC and RB by phagocytosis. Sertoli cell phagocytosis is the most noticeable. Several consequences of phagocytotic removal of AGC and RB by Sertoli cells have been proposed. Removal of AGC and RB provides appropriate spaces in the seminiferous epithelium for healthy spermatogenesis. AGC can release autoantigens when necrosis occurs, which may induce autoimmune responses. Therefore, timely elimination of AGC before releasing autoantigens prevents autoimmune responses. After phagocytosis of AGC and RB, Sertoli cells recycle these apoptotic components as an energy source. This energy source would be important for Sertoli cells because circulating nutrients barely reach to the seminiferous epithelium due to the BTB and lacking blood vessels.

4.1 Space saving

Based on origin, phagocytes can be classified into professional or nonprofessional phagocytes, respectively [49]. The hematopoietic phagocytes belong to professional and can infiltrate into the infected sites to ingest microbes and clean up damaged cells, which is critical for the innate defense against microbial infection. However, circulating phagocytes cannot migrate into tissues separated by the BTB, where resident tissue-specific phagocytes, which are considered as nonprofessional, are essential for maintaining tissue homeostasis by phagocytic removal of apoptotic substrates. The typical example is the mammalian testis. More than one hundred million sperms are produced each day in men during their whole reproductive age. Since a large number of male germ cells develop simultaneously within the seminiferous epithelium, there is a competition for space and nutrient if all the germ cells would develop into spermatozoa. Therefore, before maturing to sperm, most developing germ cells die through apoptosis, and the remaining spermatids shed most of their cytoplasmic portions as RB. Since the number of germ cells that Sertoli cells can support for finalizing their development is limited, we can speculate that the elimination of AGC and RB by Sertoli cells is important to ensure

enough spaces for a healthy germ cell production and maintain tissue homeostasis. However, this speculation lacks experimental evidence. By contrast, there is a body of substantial evidence that the phagocytic removal of AGC and RB prevents an autoimmune response.

4.2 Removal of autoantigens

Male germ cells, which are mostly developed after the establishment of central immune tolerance, express a large number of novel proteins. These new proteins of male germ cells can be recognized as "foreign antigens" by the immune system. However, male germ cells do not induce an autoimmune response in the male reproductive tracts under physiological conditions due to their special immune microenvironment. The testis is a distinct immunoprivileged organ. Immune privilege represents a special immunological status that exists in several mammalian organs, including the eye, brain, pregnant uterus, and testis, where allografts or/and xenografts can survive without evoking immune rejection [50]. The testis tolerates both alloantigens and immunogenic autoantigens [51]. Various mechanisms are involved in the maintenance of testicular immune privilege [42], in which Sertoli cells play crucial roles.

Sertoli cells modulate testicular immune privilege with different mechanisms (**Figure 5**). The BTB protects the majority of the antigenic germ cells by sequestering autoantigens behind the BTB from immune components in the interstitial spaces. The BTB is formed between adjacent Sertoli cells near the basal membrane of the seminiferous epithelium (**Figure 1**). Several cellular junctions, including tight junction, gap junction, and basal ectoplasmic specialization, are involved in the BTB formation. The BTB divides the seminiferous epithelium to the basal and adluminal compartments [52]. The BTB limits the access of immune contents residing in the interstitial spaces into the adluminal compartment and sequesters the germ cell autoantigens within the adluminal compartments. Therefore, the BTB plays an important role in maintaining immune privilege within the adluminal compartments of the seminiferous epithelium. Although the BTB sequesters the late stage of germ cells in the adluminal compartments, the early stage of germ cells, including preleptotene spermatocytes and spermatogonia that reside outside the BTB, also produces antigenic proteins [53]. Moreover, certain germ cell antigens behind the BTB can egress into the interstitial spaces, and these antigens do not induce an immune response in the testis [54]. These observations suggest that the BTB cannot completely sequester germ cell antigens and should be only partially responsible for testicular immune privilege. In fact, the interstitial spaces outside the BTB also enjoy immunoprivileged status. A dense network, including the tissue structure, local immunosuppressive milieu, and systemic immune tolerance, coordinately regulates the immunoprivileged environment in the testis [42, 55]. In addition to the BTB, Sertoli cells produce various anti-inflammatory factors that regulate the testicular immune microenvironment [56]. Sertoli cells express activin A and activin B [57]. Activin A inhibits the expression of pro-inflammatory cytokines, thereby suppressing the testicular inflammatory responses. TGF-β is also predominantly produced by Sertoli cells in the testis. As an anti-inflammatory factor, TGF-β1 protects islet β-cell grafts after co-transplantation with Sertoli cells [58]. Moreover, Sertoli cells express Fas ligand (FasL) and programmed death ligand-1 (PD-L1), two negative immunoregulatory ligands which are both involved in the maintenance of testicular immune privilege [59, 60].

Phagocytosis is a biological process that regulates immunity [61]. The phagocytic removal of AGC and RB by Sertoli cells is critical for timely elimination

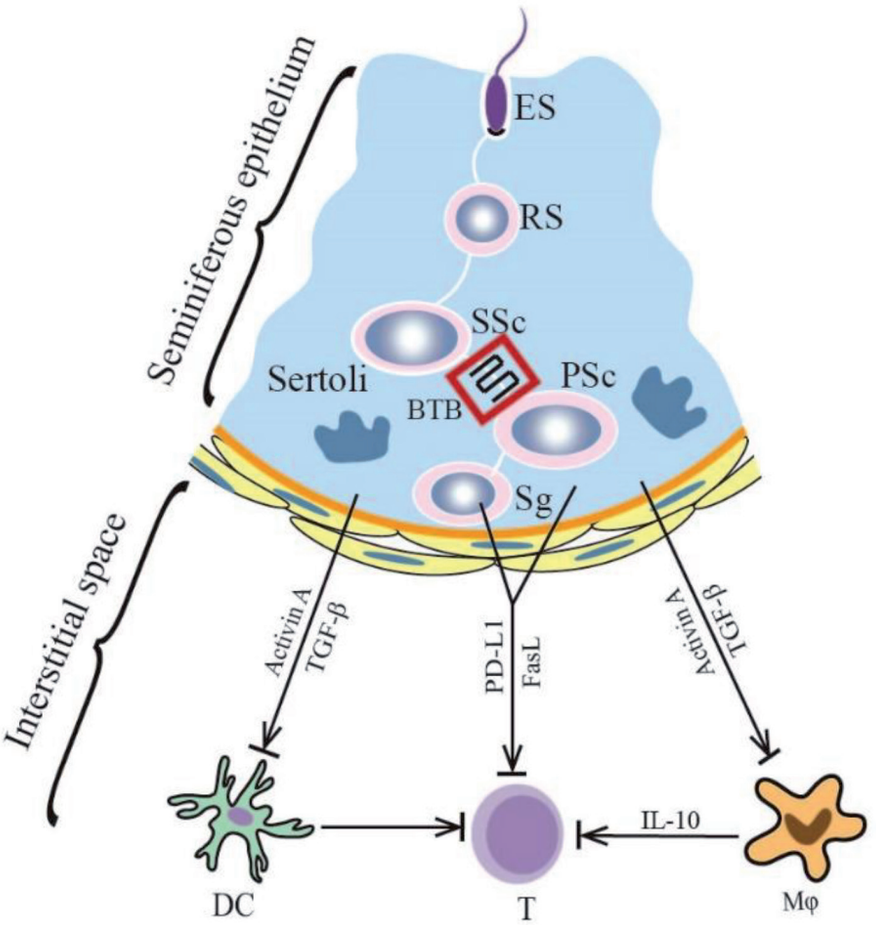

Figure 5.
Role of Sertoli cells in testicular immune privilege. Sertoli cells produce various anti-inflammatory cytokines, including activin A and TGF-β, which inhibit immune response of dendritic cells (DC) and testicular macrophages (Mφ). Sertoli cells, together with germ cells, also express high level of Fas ligand (FasL) and programmed death ligand 1 (PD-L1) that can inhibit immune response by inducing apoptosis of T lymphocytes. Sg, spermatogonia; PSc, primary spermatocyte; SSc, secondary spermatocyte; Rs, round sperm; ES, elongated sperm.

of autoantigens that may be released if AGC and RB are broken down. Toll-like receptors (TLRs) belong to a subfamily of pattern recognition receptors that initiate innate immune responses. Several TLRs are expressed in testicular cells and can be activated by their relative ligands [41, 62, 63]. Damaged tissues and necrotic cells may release endogenous TLR ligands, namely, damage-associated molecule patterns (DAMPs), which can induce noninfectious inflammatory response (**Figure 3**, left side). Various DAMPs, including high-mobility group box 1 (HMGB1) and several heat stock proteins (HSPs), have been recognized to activate TLR2 and TLR4 [64, 65]. Notably, HMGB1 and HSPs are abundantly expressed in male germ cells and can be released under stress conditions [66, 67]. Therefore, necrotic germ cells and RB breakdown may release endogenous TLR ligands, thus inducing inflammatory responses. Accordingly, physical trauma and chemical noxae that may damage germ cells are risk factors of chronic testicular orchitis [68]. An impaired removal of AGC leads to autoimmune orchitis [69]. The damaged male germ cells (DMGCs) induce the expression of various inflammatory mediators, including pro-inflammatory factors and chemokines, in Sertoli cells, thereby promoting leukocytes' infiltration to the testis [70]. The DMGC-induced inflammatory cytokine expression and immune cell infiltrations require TLR2 and TLR4 in Sertoli cells. Therefore, timely removal of AGC and RB by Sertoli cells is essential for maintaining immune homeostasis in the testis to prevent autoimmune orchitis.

4.3 Providing energy

Another meaning of male germ cell death and removal of AGC and RB serves as energy sources for Sertoli cells [71]. After phagocytosis by Sertoli cells, AGC and RB fuse with lysosomes. AGC and RB are subsequently broken down and recycled as energy sources for ATP production. The most noticeable phenotype of Sertoli cells is the formation of numerous lipid droplets in the cell cytoplasm. These lipid droplets result from the breakdown of engulfed AGC and RB [13, 72]. Unlike the majority of cell types that mainly use glucose as an energy source, Sertoli cells predominantly use lipids to produce ATP [71]. The lipids from AGC and RB should be the main energy sources for Sertoli cells. Sertoli cells provide essential physical and environmental support for spermatogenesis, which are energy consumers. Corresponding to their function, Sertoli cells exhibit an active energy metabolism and produce high levels of ATP [71, 73]. While lipids and glucoses can be substrates for ATP production within cells under physiological conditions, the majority of cell types use glycogen to produce ATP, whereas lipids serve as energy storage. Only minor cell types, such as adipocytes, myocardial cells, and Sertoli cells, have been confirmed to actively utilize lipids to produce ATP. Why these cell types predominantly use lipids as energy sources remains unclear. However, the active usage of lipids by Sertoli cells is compatible with the special microenvironment in the testis. Sertoli cells are barely reached by the nutrition from the peripheral circulation due to the barriers in the basement membrane, BTB, and the absence of blood capillaries within the seminiferous epithelium. The simplest way for Sertoli cells to have enough energy to support spermatogenesis is to recycle the lipid contents of AGC and RB. Therefore, the phagocytic removal of AGC and RB by Sertoli cells is necessary for Sertoli cells to ensure their functions, which confers a novel meaning for a large number of germ cells to undergo apoptosis during spermatogenesis.

5. Conclusive remarks

The mammalian testis possesses a special microenvironment for fulfilling its functions. The adluminal compartments of the seminiferous epithelium are separated from blood circulation by the BTB, and the circulating phagocytes cannot reach these regions. Therefore, Sertoli cells are responsible for the clearance of numerous AGC and RB during spermatogenesis. The phagocytic removal of apoptotic components by Sertoli cells is not only for waste disposal but also confers more meaning. In addition to prevention of autoimmune responses by removing autoantigens, recycling of apoptotic components can be used as an energy source for Sertoli cells. These biological processes would be particularly important in the tissues where immunogenic autoantigens are produced and seldom reached by circulating nutritious substrates. The mechanisms behind cell death and their removal by phagocytes, and their tissue-specific significance, are worthwhile to investigate in depth.

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