THE NATURE OF SOMATIC CELL INTERACTIONS IN THE SEMINIFEROUS TUBULE

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I. INTRODUCTION

It is generally accepted that the hormonal control of mammalian spermatogenesis is mediated by direct actions of hormones on testicular somatic cells, and that the optimal expression of programs for germinal cell development is dependent upon the microenvironment provided by adjacent somatic cells (Fig. 1) (for reviews, see 16, 17).

In this presentation, we shall focus upon the nature of interactions between Sertoli cells and peritubular cells, with emphasis on observations from our laboratory on the properties of these cells from the rat testes in monoculture and in co-culture. Before becoming totally immersed in this rather specialized topic, however, it may be useful to consider some of the recent developments in male reproductive biology which have led to current views of the hormonal control of spermatogenesis, and to describe in general terms the basis for the growing importance attributed to the role of testicular somatic cells in the formation and maintenance of a unique local environment in the seminiferous tubule.

It has long been known that germinal cell development is dependent upon the maintenance of an appropriate hormonal milieu. Yet, germ cells do not appear to respond directly to FSH, LH or to androgens. These hormones, which are required to restore spermatogenesis in hypophysectomized animals, elicit their overall effects by modulating the functions of testicular somatic cells. The hormonally-stimulated somatic cells interact at several levels with adjacent germinal cells, providing in some manner all that is required for the orderly progression of spermatogonia to spermatozoa. Within this framework, much remains to be defined and delineated:

1) The easiest and most straightforward task consists simply of cataloguing the list of functional activities of Sertoli cells and peritubular cells which can be directly modulated by various hormones. Advances in our knowledge in this area are progressing rapidly, along with a better comprehension of hormone actions.
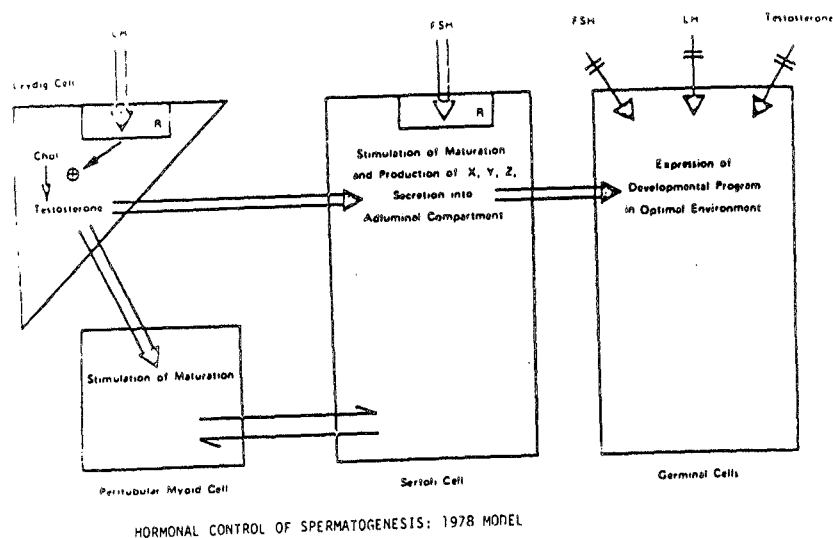
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HORMONAL CONTROL OF SPERMATOGENESIS: 1978 MODEL

Fig. 1. Hormonal control of spermatogenesis (Fritz, 1978).

2) The next project is to try to understand the nature of the responses of Sertoli cells and peritubular cells, singly and cooperatively; and to determine which responses may be most relevant in forming the microchemical environment (49, 63) and cytoarchitectural arrangements (13, 44) prevailing within the seminiferous tubule. It is in this area that we shall soon be presenting data and speculations. While a beginning of acquisition of knowledge is becoming evident, the unknown portions predominate.

3) It would be highly rewarding to discover the precise local conditions required for the progress of specific stages of spermatogenesis. Which of the components produced by Sertoli cells and peritubular cells are essential for the expression of programs encoded within germinal cells at various stages of development? As suggested by the nature of the question very little is known in this area. Similarly, only bits of information are available on the biochemical changes taking place within germinal cells which are responsible for the well-described structural changes occurring during development (3, 7).

4) Virtually nothing is known about mechanisms involved in the control of spermatogenesis from the vantage point of molecular biology. This is hardly surprising, since an understanting of the cellular targets for hormonal actions on spermatogenesis has been gained only recently, and biochemists are just beginning to comprehend possible mechanisms of cell-cell interactions (15).

Within this context, we shall initially consider the characteristics of peritubular cells alone, and those of Sertoli cells alone. We shall then concentrate upon the nature of interactions between these two types of cells, and

speculate upon the biological relevance of these interactions in establishing the unique microchemical and cytoarchitectural arragements within the seminiferous tubule required for germinal cells to develop. Finally, we shall attempt to relate these observations to the general field of mesenchymal cell-epithelial cell interactions.

II. PROPERTIES OF PERITUBULAR CELLS

A. Overview

In the intact seminiferous tubule, spermatogonia and a relatively small percentage of the basal surfaces of Sertoli cells rest upon a basal lamina which separates these cells from peritubular myoid cells in the boundary tissue. The tubule wall in rat testis is comprised of layer of cells and cell-free matrix, commencing with the basal lamina or basement membrane adjacent to the seminiferous epithelium; a relatively clear zone containing a reticulum of collagen fibrils; a thin basal lamina-like structure which provides an inner coating for the continuous layer of peritubular myoid cells; another clear zone; and finally an outer layer of flattened endothelial cells that line the peritubular lymphatic sinusoids (11). Structural and ultrastructural characteristic of peritubular myoid cells have been well described (5, 6, 11, 13, 30, 31, 42). The development of peritubular myoid cells *in vivo* is androgen-dependent (5), and peritubular cells have been shown to contain androgen receptors (47, 62).

The peritubular myoid cell layer provides an incomplete barrier to the passage of molecules from the interstitial space into the basal compartment of the seminiferous tubule (11). Some peritubular cells are in close apposition, joined by tight junctional complexes which exclude the passage of small molecules such as lanthanum nitrate. However, sufficiently large intercellular clefts exist between other peritubular myoid cells at various stages of the cycle of the seminiferous epithelium to permit the penetration of lanthanum past the boundary tissue. In these apparently randomly located regions of the tubule, which are estimated to represent about 15% of the length of the tubule, lanthanum injected intravenously has been visualized in the space between the peritubular myoid cell layer and the germinal cells (11). Data reviewed permit the conclusion that peritubular cells form an initial barrier which partially exludes the passage of molecules past the tubule wall. These properties are insufficient to account for those of the seminiferous tubule barrier (often inappropriately called the "blood-testis" barrier).

Other functions of the peritubular cells are not clear, but they are thought to play a role in tubular contractions (22).

Table 1 Characteristics of Monocultures of Sertoli Cells and Peritubular Cells Prepared From Testes of Immature Rats and Plated onto a Plastic Substratum

Property	Sertoli Cells	Peritubular Cells
Plating and Initial Migration	Attach as aggregates; migration initially from periphery, leaving cluster of cells within aggregates for first 24-48 hrs. Good plating efficiency in serum-free MEM.	Uniformly distributed; migrate rapidly, forming layer(s) within 24 hrs; requires serum in MEM for efficient plating and migration.
Types of Layer(s) Formed	Spread to form contiguous monolayer with epithelial borders.	Form flattened monolayers and multilayers with uneven diffuse borders.
Shapes of Non-Dividing Cells	Squamous to low columnar; polygonal becoming rounder and flatter during culture, with characteristics surface structures evident by SEM	Attenuated, flat cells, ranging from triangular to polyhedral, with smooth free surfaces evident by SEM. Often spindle-shaped at high density.
Types of Boundaries Between Cells	Pavement or mosaic type; tight junctional complexes in apical regions away from sites of attachment; no criss-crossing; some sites of interdigitation among filopodia of adjacent cells.	Frequent criss-crossing; no tight junctional complexes evident.
Ultrastructural Characteristics	One or two nucleoli per irregularly shaped nucleus; paranucleolar karyosomes occasionally evident; well developed SER; RER less abundant and usually not dilated; rich in lipid droplets.	Multiple nucleoli in oblong-shaped nucleus; RER abundant and frequently dilated; lipid droplets rare; frequent arrays of subplasmalemnal microfilaments (4-7 nM diameter) and characteristic dense bands.
Biochemical Properties	FSH elicits structural changes; increased adenylate cyclase activity; increased synthesis of DNA, estrogens, inositol and several proteins, such as ABP, PA, etc.; androgens increase formation and release of ABP; synthesize and secrete collagen IV, laminin and proteoglycans containing chondroitin sulfate and heparin sulfate.	No detectable effects of FSH; patterns of proteins synthesized and secreted are different from those produced by Sertoli cells; androgens increase formation of P Mod-S activity; synthesize and secrete collagen IV and I, fibronectin and proteoglycans containing chondroitin sulfate.

For details, see the text, and references 51-61.

Fig. 2. Transmission electron micrographs of peritubular cells isolated from testes of 20-day-old rats and maintained in culture for 10 days (panel A) or 6 days (panel B) in modified Eagle's's minimal essential medium containing 10% calf serum. Panel A: 4000×; Panel B: 16000×. For details, see the original publications (57, 59).
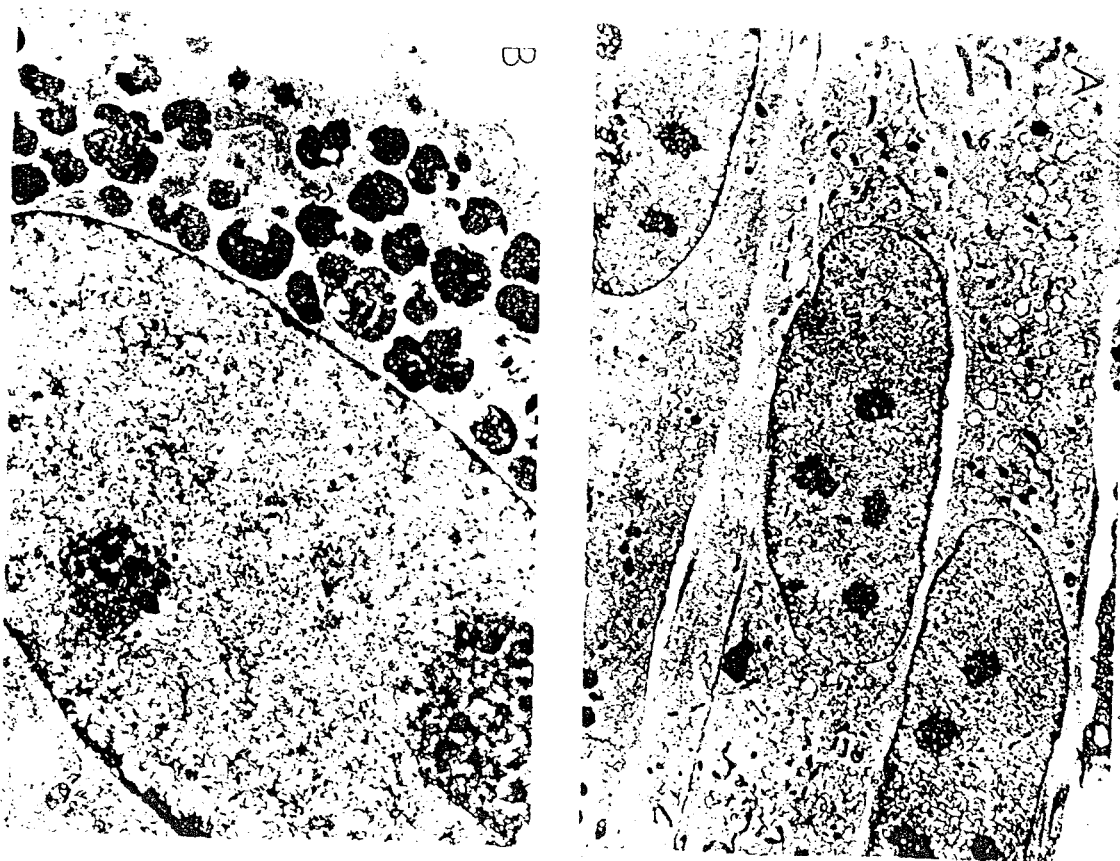
B. Properties of Peritubular Cells Maintained In Culture

Many of the structural characteristics of peritubular cells (5, 11) are retained in preparations in culture (Table 1 and Fig. 2) (34, 57, 58, 59, 61).

Peritubular cells readily spread to form monolayers or multilayers when plated on various substrata. However, the addition of serum is required for efficient plating, subculture and growth. No chemically defined-serum-free

Table 2. Levels of Soluble Extracellular Matrix (ECM) Components Produced by Peritubular Cells or Sertoli Cell Maintained in Serum-Free MEM in Monoculture or Co-Culture

Cell Type in Culture	Levels of ECM Componets in Medium During 48 Hour Collection Period Starting on Day 5 (ng/µg DNA)		
	Fibronectin	Collagen I	Collagen IV
Peritubular Cells	432 ± 68	254 ± 37	350 ± 54
Sertoli Cells	0	0	322 ± 52
Peritubular and Sertoli	182 ± 34	96 ± 15	197 ± 30

All values are expressed as the mean ± SEM for 3 separate experiments, each of which was analyzed in triplicate (n = 9). For details of the competitive ELISA assays, and other information, consult the original publication (55).

has thus far been found which can support these functions of peritubular cells in monoculture. After cells have grown to confluence, they can survive for several weeks in serum-free medium. Under these conditions, peritubular cells have the capacity to synthesize fibronectin (61); types I and IV collagen (55); and proteoglycans containing chondroitin sulfate (51) (Table 2). The gel electrophoretic patterns of other proteins synthesized by peritubular cells in culture and released into the medium have been described (24, 61). The presence of specific protein(s) synthesized by peritubular cells which modulate the functional activities of Sertoli cells has recently been detected in conditioned medium (52). This is discussed below in the section on peritubular cell — Sertoli cell interactions.

III. PROPERTIES OF SERTOLI CELLS

A. Overview

The structure and functions of these cells have been exhaustively reviewed (12, 16, 17, 35, 36, 39, 40, 41, 46, 63). A three-dimensional reconstructions of a rat stage V Sertoli cell has recently been achieved, allowing an increased appreciation of the extraordinary complexity and diversity of interactions of Sertoli cells with all classes of cells in the seminiferous tubule (44, 64, 66). It is abundantly clear from information reviewed that Sertoli cells form the basic architecture and organization of the seminiferous tubule, including the generation and maintenance of the testis barrier separating the adluminal from the basal compartments (12). The barrier, which functions to maintain the unique chemical composition of fluids in the lumen of the seminiferous tubule and in the rete testis (49, 63), also serves to provide an immunologically privileged environment for germinal cells in the adluminal compartment (38).

The list of components known to be synthesized by Sertoli cells is rapidly growing (Table 3). Most of this information has been obtained by investigating the secretory products of Sertoli cell-enriched preparations in culture. Additional information which strengthens interpretations of the physiological relevance of in vitro observations has been obtained with experiments on seminiferous tubules in organ culture, and with analyses of testes depleted of germ cells by irradiation in utero (for reviews see, 36, 63). In the cases of those proteins isolated from testes or rete testis fluid to which antibodies have been generated, immunocytological examination of testes has demonstrated the presence in Sertoli cells of androgen binding protein (ABP) (1); clusterin (60a); dimeric acidic glycoprotein (20); transferrin (20) and plasminogen activator (Tung et al., in preparation). However, none of these proteins is necessarily unique to Sertoli cells (17).

Table 3. Products Synthesized and Released by Primary Cultures of Sertoli Cell-Enriched Preparations

Proteins:

Androgen Binding Protein	Collagen IV
Anti-Mullerian Hormone	Proteoglycans
Transferrin	Clusterin
Ceruloplasmin	Dimeric Acidic Glycoprotein
Plasminogen Activator	Others ("Cyclic" Proteins; "Lactalbumin", etc)

Steroids: 17 ß-Estradiol

Carbohydrates: Inositol
                Lactate, Pyruvate

For details, see references 16, 17, 20, 34, 35, 36, 39, 40, 46 and 50.

B. Properties of Sertoli Cells Maintained in Culture

As in the case of peritubular cells, many of the in vivo structural characteristics of Sertoli cells (12) are retained in cells cultured in serum-free, chemically defined medium (Table 1, and Fig. 3) (56, 59). The normal cytoarchitecture is better maintained in Sertoli cells cultured on an extracellular matrix substratum than in cells plated on an uncoated plastic substratum (60). Evidence of maturation of Sertoli cells in culture has been presented (56).

Patterns of proteins synthesized by Sertoli cell-enriched preparations have been described (9, 24, 65, 67). The gel electrophoretic profile of proteins secreted by Sertoli cells is clearly different from patterns of proteins secreted by peritubular cells (24, 61). For example, Sertoli cells in culture do not synthesize Type I collagen or fibronectin, but peritubular cells do (61) (Table 2). Sertoli cells secrete transferrin (53), ceruloplasmin (53a), and a host of other proteins (Table 3) which are not synthesized by peritubular cells.
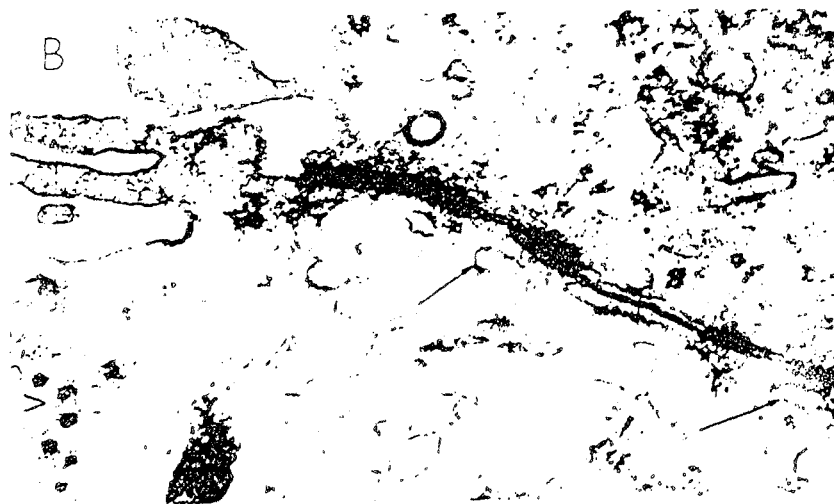
Fig. 4 Transmission electron micrographs of Sertoli cells isolated from testes of 20-day-old rats maintained in culture for 5 days (panel A) or 10 days (panel B) in modified Eagle's minimal essential medium containing 10% calf serum. Arrow in panel A points to complex interdigitations between cells; and arrows in panel B point to flattened cisternae adjacent to the tight junctional complex. Panel A: 7800 x; Panel B: 60000 x. For details, see the original publication (56).

As already indicated, considerable information is being acquired about some of the products which comprise only a small percentage of total proteins synthesized, such as ABP (2, 18), and plasminogen activator (26, 27, 28), especially with respect to factors modulating the formation of these proteins. The hormonal regulation of the production of specific proteins, lactate, inositol and estrogen by Sertoli cells in culture has been extensively documented in several reviews (16, 17, 20, 35, 36, 40, 50, 63). It is well established that Sertoli cells can readily respond to FSH and to androgens, and also to insulin. More recently, IGF-II (MSA) has been reported to increase transferrin synthesis by Sertoli cells (54), and both IGF-I and IGF-II have been shown to increase the synthesis of lactate, DNA and protein by Sertoli cell-enriched preparations from testes of immature rats (4).

IV. INTERACTIONS OF SERTOLI CELLS WITH PERITUBULAR CELLS

A. Extracellular Matrix Formation and Deposition

Sertoli cell-enriched aggregates plated on top of peritubular cells spread to form a monolayer. Sertoli cells reaggregate, and then form mounds, nodules, and eventually complex protrusions as adjacent nodules merge (58). Aggregates are surrounded by ribbons of peritubular cells, separated by a limiting membrane and collagen fibrils (Figure 4B). The structure formed in vitro in the co-cultured system resembles the histologic appearance of seminiferous tubules in situ (Figure 4A). Basal lamina formation is evident (Figure 4B). In contrast, peritubular cells and Sertoli cells in monoculture spread to form relatively uniform layers, devoid of mounds, nodules or protrusions (25, 58, 61). Sertoli cells and peritubular cells in co-culture can survive in serum-free MEM for months, whereas neither cell type can do so in monoculture (58). ABP production by Sertoli cells is enhanced and sustained by the presence of peritubular cells in co-culture (23, 58).

These remarkable changes in properties of each cell type when placed in co-culture provided a strong incentive to investigate the nature of the apparent cooperativity. We first explored the influences of cell interactions on the formation and deposition of extracellular matrix components (Table 2). Sertoli cells, which did not produce Type I collagen during the first week of culture, stimulated peritubular cells to synthesize more soluble Type I collagen during co-culture. In addition, co-culture resulted in a greater deposition in extracellular fibrils of Type I collagen. Type IV collagen was synthesized by each populations of cells; fibronectin was synthesized by peritubular cells but not by Sertoli cells; and laminin was detectable in Sertoli cells but not in peritubular cells. Deposition of all ECM components investigated was more pronounced in co-cultures of Sertoli cells and peritubular cells than in monocultures of either cell type (Table 2; 55). We interpret data presented to indicate

that the two cell types in co-culture act cooperatively to form the basal lamina-like structure observed *in vitro* (Figure 4B). We propose that the same processes may occur *in vivo*.

B. Evidence for the Existence of Paracrine Factors

1. P Mod-S

Conditioned medium obtained from peritubular cells in culture stimulates Sertoli cell to increase the formation and release of ABP (52) (Table 4). As indicated previously, ABP production by Sertoli cells is augmented when peritubular cells are present during co-culture (23, 58). We interpret data shown to indicate that a protein, designated "P Mod-S", is released by

Table 4. Effects of P Mod-S on Levels of Androgen Binding Protein (ABP), Transferrin and Plasminogen Activator (PA) Produced by Primary Cultures of Rat Sertoli Cells

Additions to Basal Medium	Levels of Protein Released Into Medium By Sertoli Cells in Culture		
	ABP (ng/µg DNA/72h)	Transferrin (ng/µg DNA/72h)	PA (units/µg DNA/72h)
Control	4.5 ± 0.5	58 ± 10	60 ± 50
FIRT*	16.0 ± 4.0	180 ± 20	210 ± 20
P MOD-S**	14.5 ± 3.5	200 ± 15	35 ± 5.0

*FIRT represents follicle stimulating hormone (oFSH, NIH S-16, 100 ng/ml); insulin (5 µg/ml); retinol (0.35 µM); and testosterone (1 µM) in serum-free MEM.
**P Mod-S is the partially purified 50-100 kDa fraction prepared from 200X-concentrated serum-free conditioned medium obtained from secondary cultures of peritubular cells. The concentration of P Mod-S added to the Sertoli cell culture medium was 1.5 µg protein/ml. All values are given as the mean ±S.D. for n = 9. For details consult the original publication (52).

peritubular cells (P), and this factor modulates functions of Sertoli cells (S). Addition of P Mod-S stimulates the formation of transferrin to as great an extent as it augments the synthesis of ABP by Sertoli cells (Table 4). Addition of a mixture of FSH, insulin, retinol and testosterone (FIRT) is required to elicit maximal stimulation of transferrin formation by Sertoli cells (54a). The same mixture increased the synthesis of ABP and transferrin to an extent comparable to that observed in Sertoli cells cultured in the presence of P Mod-S, but it should be noted that P Mod-S was more stimulatory than FSH, insulin, retinol or testosterone alone (Table 4). The formation of plasminogen activator (PA) is also greatly stimulated by addition of FIRT, in confirmation of observations previosuly reported on the effects of FSH on PA synthesis by Sertoli cells in culture (26, 27, 28). P Mod-S addittion did not enhance PA levels in the medium. Instead, the formation of PA by Sertoli cells cultured in the presence of P Mod-S appeared to be reduced slightly (Table 4).

Fig. 4 Transmission electron micrographs relationship between peritubular cells and Sertoli cells *in situ* (panel A) and in a co-culture of peritubular cells and Sertoli cells prepared from testes of 20-day-old rats. The micrograph of the seminiferous epithelium of a 10-day-old rat shows the basal region of a Sertoli cell (SC) and a spermatogonium (Sp) resting on the basal lamina, which is adjacent to the dense layer of microfilaments (arrow), near the plasma membrane of the Sertoli cell. Between the amorphous basal lamina (about 30 nm wide) and the layer of peritubular cells, numerous collagen fibers are evident (56). Panel B is a micrograph of peritubular cells and Sertoli cells which had been in co-culture for 28 days in MEM containing 10% calf serum. Arrows point to the basal lamina, and numerous collagen fibers are discernible between Sertoli cells and peritubular cells in the space between basal laminae. Panel A: 40000 x; Panel B: 6000 x. For details consult the original publications (56, 58).

... ... of Sertoli cells and peritubular cells than in monocultures of either cell type (Table 2; 55). We interpret data presented to indicate

Mechanisms involved are unknown, but it may be concluded that P Mod-S influences Sertoli cell functions in a manner which is not identical to the actions of any of the components present in the FIRT mixture.

Experiments are in progress to isolate P Mod-S and to determine its site(s) of action (52). Thus far, our observations indicate that P Mod-S is a protein having an apparent molecular mass of 70 KDa, and that its addition increases the incorporation of [^{35}S]-methionine into several proteins released into the medium by Sertoli cells. In primary cultures of peritubular cells maintained in the presence of androgens (testosterone or dihydrotestosterone), levels of P Mod-S activity secreted are greater than those detected when peritubular cells are maintained in MEM alone or in MEM containing estrogens (52). We have also recently observed that the stimulation by androgens of ABP production by populations of Sertoli cells nearly devoid of contaminating peritubular cells is less than that obtained in Sertoli cell-enriched preparations containing peritubular cells. Reconstitution experiments have shown that androgen stimulation of ABP production by Sertoli cells is enhanced by the presence of peritubular cells (52a).

We interpreted these data to indicate that androgens stimulate peritubular cells (stromal or mesenchymal cells) to synthesize P Mod-S, which then modulates the functions of adjacent Sertoli cell (epithelial cells). In this manner, P Med-S may serve as a local mediator of androgen actions on Sertoli cells. The actions postulated are independent of the direct effects of testosterone on Sertoli cells (32, 33, 46).

2. Speculations on Other Paracrine Factors Produced by Testicular Cells

The existence of other paracrine factors has been suggested by several lines of evidence. Since Sertoli cells alter the functions of peritubular cells in co-culture (58), we postulate the existence of a putative S Mod-P, a component secreted by Sertoli cells which modulates the activities of peritubular cells. The influences of LH on the formation of interstitial fluid of the testis (50), and the influences of hormone on the properties of capillary distribution and permeability in the testis during gonadal maturation (25) suggest the presence of factors produced by testicular cells under hormonal regulation which may affect the functions of vascular endothelial cells. The presence of androgen receptors in testicular endothelial cells has recently been reported (37), suggesting the possibility that these cells may directly respond to androgens. Alternatively, peritubular cells could secrete a factor (P-Mod-V) which alters the properties of vascular endothelial cells. Several investigators have reported that FSH modulates the synthesis of androgens by Leydig cells (for reviews see 16, 45). These data suggest the possibility that Sertoli cells (the only cells known to respond directly to FSH) can produce S-Mod-L, a factor which modulates the properties of Leydig cells. Finally, several types of interactions between Sertoli cells and germinal cells have been reported (for review, see

39). Not only do Sertoli cells secrete products which affect the development of germ cells, but the germinal cells association pattern can modulate the functions of Sertoli cells. For example, basal rates of formation of plasminogen activator by Sertoli cells are over 10-fold greater in cells in stages VII and VIII of the cycle of the seminiferous epithelium (19, 29).
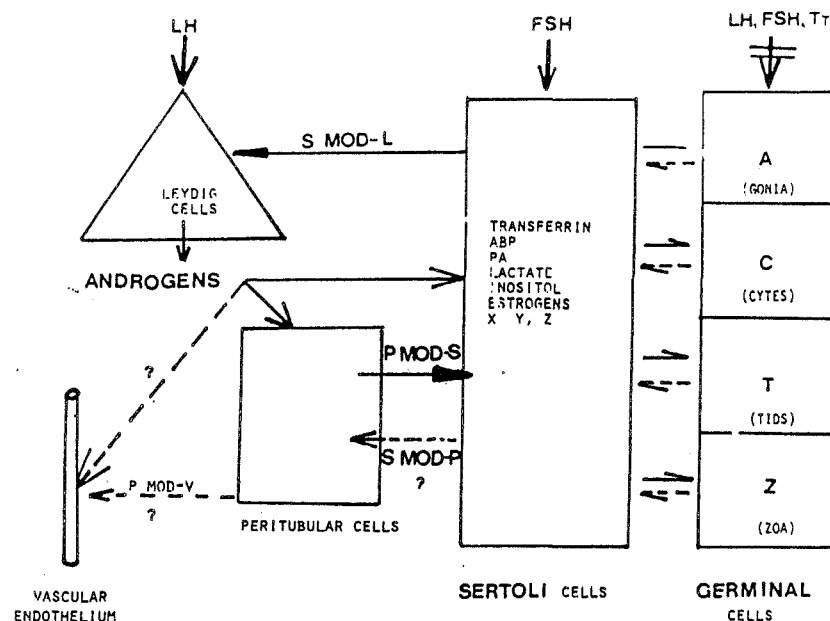

Fig. 5. Hormonal control of spermatogenesis: 1984 model. Solid arrows represent known actions or effects, while dotted arrows indicate postulated actions. In the germinal cell column, «A» represents spermatogonia, «c» represent spermatocytes, «T» represents round and elongating spermatids, and «Z» represents spermatozoa released into lumen. «P-Mod-S» designates the protein(s) secreted by peritubular cells which modulate the activities of Sertoli cells (52). «S. Mod-P» and «S-Mod-L» designate hypothetical protein(s) secreted by Sertoli cells which modulate the activities of peritubular cells (58), and Leydig cells (45), respectively. «P Mod-V» designates the hypothetical protein(s) secreted by peritubular cells which modulate the activities of vascular endothelial cells. For details, see the text and (17).

The likelihood emerges that factors may be produced by many type of cells in the seminiferous tubule which exert local control, and that these paracrine or autocrine components may play an integral role in establishing the nature of the microenvironment required for spermatogenesis. The 1978 version of the hormonal control of spermatogenesis presented earlier (Figure 1) is therefore accordingly modified to reflect these new concepts (Figure 5). Somatic cells of the seminiferous tubule are regulated directly by systemic hormones (primar-

ily FSH and androgen). In response to these hormones, the somatic cells are stimulated to synthesize and secrete modulating factors (such as somatomedins, P Mod-S, etc.) which may exert local control of neighboring cells. In addition, Sertoli cells would be stimulated to secrete components which provide necessary nutrients and factors (such as lactate, inositol, transferrin, ceruloplasmin, etc.). These metabolites are postulated to be required by neghboring germinal cells for their maintenance, and for the expression of their encoded programs. The role of specific paracrine factors in these processes remains to be determined. We postulate that peritubular cells and Sertoli cells form a functional unit required to maintain and alter cytoarchitectural arragements in the seminiferous tubule during various stages of spermatogenesis. Some of these effects could be mediated via cooperative effects on ECM deposition. In addition, peritubular cells and Sertoli cells may act cooperatively to modulate changes required during the translocation of clones of preleptotene spermatocytes from the basal to the adluminal compartments. We presume that a restructuring of the extracellular matrix takes place as advancing cytoplasmic extensions of Sertoli cells begin to separate clones of preleptotene spermatocytes from the basal lamina at stages VII-IX of the cycle of the seminiferous epithelium (19, 43, 55).

The complexity of cell interactions demanded during spermatogenesis suggests that processes involved may be as intricate as those being uncovered in the immune system. Immunologists have discovered large number of interactions among B cells, T cells and macrophages during immune responses (48). If similar phenomena are occuring in the testis, we may anticipate encountering cell-mediated interactions, as well as a host of local humoral agents in the seminiferous tubule.

The specificity of components being discovered in the testis such as seminiferous growth factor (14); LHRH-like peptide (50); or P Mod-S (52) remains to be delineated. It is possible that one or more of the peptides may be similar to families of modulating factors and growth factors isolated from the other sources (for review, see 21). Continuing investigations of peritubular cell-Sertoli cell interactions are almost certain to reveal new surprises, and they may well show a relationship to investigations of mesenchymal cell-epithelial cell interactions in other organs, such as developing mammary gland (10) or the epithelia of the genital tract (8). In several systems, the hormonal modulation of epithelial responses has been shown to be mediated by mesenchymal cells under direct hormonal control (8). This may also prove to be the case in stromal cell (peritubular cell) — epithelial cell (Sertoli cell) interactions in the seminiferous tubule.

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