DR LAWRENCE M. ROTH (Orcid ID : 0000-0002-5538-9231)

DR LIANG CHENG (Orcid ID : 0000-0001-6049-5293)

Article type : Review

Classical gonadoblastoma: its relationship to the "dissecting" variant and undifferentiated gonadal tissue

Lawrence M. Roth[1] & Liang Cheng [1,2]

[1]*Department of Pathology and Laboratory Medicine, Indiana University School of Medicine, Indianapolis, IN 46202*

[2]*Department of Urology, Indiana University School of Medicine, Indianapolis, IN 46202*

Running Head: Classical gonadoblastoma and its variants

Disclosures: This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors. No conflict of interest exists.

Address for correspondence: Professor L.M. Roth, Department of Pathology and Laboratory Medicine, Indiana University School of Medicine, Van Nys Medical Science Building 128, 635 Barnhill Drive, Indianapolis, IN 46202-5120, USA. e-mail: lroth@iupui.edu

Abstract: Classical gonadoblastoma occurs almost entirely in the dysgenetic gonads of an individual who has a disorder of sex development. Approximately 40% of such neoplasms are bilateral. Almost all gonadoblastomas occur in patients who have a Y chromosome or part thereof; testis specific protein Y-encoded 1 (*TSPY1*) is the putative gene. If a gonad in a patient who has a disorder of sex development contains germ cells with delayed maturation and also harbors the *TSPY1* gene, the cells can undergo transformation to classical gonadoblastoma. The latter consists of rounded islands composed of germ cells, sex cord elements, and hyaline basement membrane material surrounded by a variably cellular gonadal stroma that sometimes contains steroid cells. Classical gonadoblastoma can be interpreted as a noninvasive neoplasm that is the precursor of germinoma, and, indirectly, other more aggressive germ cell neoplasms. Undifferentiated gonadal tissue is the precursor of classical gonadoblastoma and contains germ cells with delayed maturation that express octamer-binding transcription factor 4 (OCT4); however, other germ cells show normal maturation and express TSPY1. If all germ cells in a patient with undifferentiated gonadal tissue involute, the result is a secondary streak. Undifferentiated gonadal tissue is a non-neoplastic condition that should be clearly distinguished from "dissecting gonadoblastoma," a neoplasm derived from classical gonadoblastoma that is the precursor of some germinomas. "Dissecting gonadoblastoma" is a variant of classical gonadoblastoma that has unusual growth patterns and contains both sex cord and germ cell elements. Clonal expansion of germ cells is a characteristic of the late stage of "dissecting gonadoblastoma".

Keywords: gonad, ovary, testis, disorder of sex development, gonadoblastoma, undifferentiated gonadal tissue

Introduction

In 1953, Scully [1] first described classical gonadoblastoma, and, in a seminal article 17 years later, he reported a series of 74 cases [2]. In 2006, Cools *et al.* [3] proposed that classical gonadoblastoma arises from undifferentiated gonadal tissue within the dysgenetic gonads of an individual who has a disorder

of sex development. Since that time, the concept has been widely accepted [4-8]. Recently, however, Kao *et al.* [9] proposed a different model in which classical gonadoblastoma was said to arise from the recently described entity of "dissecting gonadoblastoma," despite the fact that they also considered the latter to be the precursor of germinoma in some cases. Subsequent to the publication of the 2016 WHO classification of testicular tumours, the same group proposed that "dissecting gonadoblastoma" could be used as a synonym for undifferentiated gonadal tissue rather than to replace the latter term [10]. In this article, we investigate whether classical gonadoblastoma arises from undifferentiated gonadal tissue or from "dissecting gonadoblastoma" and discuss the relationship of classical and "dissecting" gonadoblastoma to germinoma and other malignant germ cell tumours (Figure 1). We also consider the possible implications of the usage of the term *"dissecting gonadoblastoma"* on the clinical management of patients with a disorder of sex development.

Classical gonadoblastoma and related entities

CLINICAL FEATURES

Classical gonadoblastoma occurs almost entirely in the dysgenetic gonads of an individual who has a disorder of sex development and a Y chromosome or part thereof [4, 11]. It occurs predominantly in phenotypic females with gonadal dysgenesis and an abnormal karyotype, but occurs exceptionally in apparently normal females with a 46, XX karyotype [12]. Approximately 20% of classical gonadoblastomas occur in phenotypic males, who are most often < 20 years of age [13]. The clinical presentation in the latter patients commonly includes cryptorchidism, hypospadias, and gynecomastia [2]. The most common predisposing conditions are 45, X/46, XY partial gonadal dysgenesis, 46, XY complete gonadal dysgenesis, and 46, XY disorder of sex development [4, 14]. By definition, at least one gonad is developmentally abnormal in patients who have a disorder of sex development; however, the abnormality would not be detected histologically if the gonad had been completely replaced by neoplasm prior to operation.

MOLECULAR AND GENETIC FINDINGS

The gonadoblastoma susceptibility region on the short arm of the Y chromosome is the only oncogenic area on the human Y chromosome and predisposes the dysgenetic gonads of XY individuals to develop a noninvasive tumour; testis specific protein Y-linked 1 (*TSPY1*) is the putative gene [15-18]. If a gonad in a patient who has a disorder of sex development contains germ cells with delayed maturation and also harbors the *TSPY1* gene, the cells may undergo transformation to classical gonadoblastoma [19]. The germ cells that have delayed maturation express octamer-binding transcription factor 4 (OCT4) [3].

MACROSCOPIC APPEARANCE

Pure classical gonadoblastoma typically measures < 8 cm in greatest dimension, and the consistency varies from soft and fleshy to firm. The sectioned surface of gonadoblastoma varies from brown to yellow or gray. White flecks of calcification often occur. If concurrent germinoma is present, the neoplasm can be larger, and the malignant component has a soft, pale gray appearance (Figure 2A).

HISTOPATHOLOGY AND IMMUNOPATHOLOGY

Classical gonadoblastoma is a noninvasive neoplasm composed of rounded islands or nests of cells surrounded by a variably cellular stroma. About 40% of gonadoblastomas are bilateral [7]. Histologically, the islands are composed of germ cells intimately admixed with immature sex cord derivatives, and the latter often surround hyaline basement membrane deposits or occasionally calcifications (Figure 2B). At times, the stroma between the islands is cellular, resembling gonadal stroma, and, in some instances, stromal steroid cells are noted. Crystals of Reinke are not identified. The germ cells of individual cases of gonadoblastoma are heterogeneous, being composed of both mature and immature forms [7]. Frequently, classical gonadoblastoma shows involutional changes

including deposits of hyalinized basement membrane material and calcification. Rarely, the involutional changes are extensive resulting in a calcified mass without any viable neoplastic cells. We refer to such cases as involuted gonadoblastoma, although such cases were originally described as "burnt out" gonadoblastoma [2]. Classical gonadoblastoma contains 2 types of germ cells; the germinoma-like cells are considered to be the precursor of the malignant germ cell neoplasms associated with gonadoblastoma; however, other germ cells resemble spermatogonia [20]. The mature germ cells express TSPY1, whereas the immature germ cells express OCT4 (Figure 2C) [21]. Only a small subpopulation of germ cells in classical gonadoblastoma coexpresses both antibodies [21]. The sex cord cells show nuclear expression of steroidogenic factor 1 and cytoplasmic expression of α-inhibin (Figure 2D).

The transcription factors SRY-box 9 (*SOX9)* and forkhead box L2 (*FOXL2)* are required for male and female mammalian gonadal development, respectively [22]. In patients with a disorder of sex development, testicular differentiation in dysgenetic gonads can be visualized utilizing SOX9, and ovarian differentiation can be visualized using FOXL2 [22].

UNDIFFERENTIATED GONADAL TISSUE

In a seminal article, Cools *et al.* [3] first introduced the concept of undifferentiated gonadal tissue**.** The latter is defined as gonadal tissue containing germ cells not enclosed in seminiferous tubules or follicles that either resides without apparent organization in a background of cellular gonadal stroma or is organized together with sex cord elements into cord-like structures [3]. Undifferentiated gonadal tissue exclusively occurs within dysgenetic gonads of patients who have a Y chromosome or part thereof and has been described morphologically as surviving germ cells with delayed development within immature sex cords or isolated in the interstitium [3]. As such, undifferentiated gonadal tissue is strictly a histological diagnosis. Expression of OCT4 in undifferentiated gonadal tissue is useful in determining maturation delay of germ cells and, thus, the risk for tumorigenesis in dysgenetic gonads [23].

Macroscopically, the dysgenetic gonads of a patient with a disorder of sex development in cases lacking a neoplasm have the appearance of a streak. It is important to realize that the gonads of a patient with a disorder of sex development and a Y chromosome or part thereof are often histologically heterogenous showing various differentiation patterns as illustrated in Fig. 2G of Cools *et al.* [4]. Areas of undifferentiated gonadal tissue, ovarian tissue, dysgenetic testicular tissue, secondary streak, as well as neoplasms can occur in any combination. Undifferentiated gonadal tissue is typically a component of the dysgenetic gonad in patients who have a Y chromosome or part thereof. Maturation-delayed germ cells occurring in dysgenetic testicular tubules should be distinguished from undifferentiated gonadal tissue in these gonads [24].

At low magnification, dysgenetic gonads have a convex surface that overlies cellular gonadal stroma (Figure 3A). In some instances, the sex cord elements of undifferentiated gonadal tissue are inconspicuous (Figure 3B)**.** However, even when they are more abundant, they blend into the surrounding gonadal stroma in a seamless manner on hematoxylin and eosin stains (Figure 3C). Immunocytochemical stains, however, serve to accentuate the cord-like structures. The sex cord elements show nuclear expression of steroidogenic factor 1 and cytoplasmic expression of α-inhibin (Figure 3D)**.**

STREAK GONADS

Macroscopically, streak gonads are elongated pale white structures located in the mesosalpinges [25]. They are best known for their occurrence in Turner syndrome in patients with a 46, X karyotype. These patients lack a Y chromosome or part thereof. However, Cools *et al.* [3] described a different type of streak that occurs as a result of loss of germ cells from undifferentiated gonadal tissue in patients who have a Y chromosome or part thereof. We refer to the latter form as a secondary streak in contrast to the streaks that occur in Turner syndrome that we refer to as primary streaks.

Histologically, dysgenetic (streak) gonads have a convex surface that overlies cellular gonadal stroma (Figure 4A). In well-preserved tissue from a streak gonad**,** a surface lining of mesothelium that overlies a thin tunica albuginea is apparent (Figure 4B). The latter, in turn, overlies

cellular gonadal stroma (Figure 4C). Residual sex cords lacking germ cells can be identified in some secondary streaks, a histological feature that distinguishes the latter from primary streak gonads (Figure 4D). Although patients who have a primary streak gonad and lack the Y-chromosome or part thereof (Turner syndrome) have a negligible risk of developing classical gonadoblastoma or malignant germ cell tumour, an important finding of our review of the data of Cools *et al.* [3] and Beaulieu Bergeron *et al.* [5] is that patients with a secondary streak have a risk of developing gonadoblastoma similar to that of undifferentiated gonadal tissue. This observation suggests that classical gonadoblastoma develops before the complete loss of germ cells in the dysgenetic gonads of patients with a disorder of sex development.

MATURATION DELAY OF GERM CELLS

In patients who have a disorder of sex development with a Y chromosome or part thereof, delayed maturation of germ cells is a histological feature of their dysgenetic gonads. Maturation delay of germ cells occurs in both undifferentiated gonadal tissue and dysgenetic testicular tissue, and an association exists between the two [24]. Undifferentiated gonadal tissue is the precursor of gonadoblastoma, and dysgenetic testicular tissue is the precursor of germ cell neoplasia in situ [3]. Maturation delay of germ cells is a protective mechanism for survival of germ cells in a harsh environment; however, prolonged expression of OCT4 in maturation-delayed germ cells is considered to be a risk factor for malignant transformation.

CLASSICAL GONADOBLASTOMA

Undifferentiated gonadal tissue in dysgenetic gonads has been proposed as the precursor of classical gonadoblastoma [3]. In 67% of cases of classical gonadoblastoma that contained adjacent gonad, undifferentiated gonadal tissue was identified [3]. The germ cells in both undifferentiated gonadal tissue and classical gonadoblastoma are heterogeneous; they can express TSPY1, OCT4, or

can co-express the two [21]. Clonal expansion of germ cells and final organization in undifferentiated gonadal tissue may be the last step in the transition to classical gonadoblastoma [3]. It is of interest that the cellular gonadal stroma characteristic of undifferentiated gonadal tissue is sometimes also a component of classical gonadoblastoma, although it is considerably less conspicuous in the latter.

DYSGENETIC TESTICULAR TISSUE

In 2006, Cools *et al.* [3] recognized and in the following year Looijenga *et al.* [23] described in greater detail a second precursor of germ cell malignancy in patients with a disorder of sex development and a Y chromosome or part thereof, germ cell neoplasia in situ originating in dysgenetic testicular tissue. Although classical gonadoblastoma is the precursor of germinoma in dysgenetic gonads lacking obvious testicular tissue, germ cell neoplasia in situ is the immediate precursor of germinoma in testicular areas [4].

In patients with undervirilization syndromes, it is often difficult to distinguish maturation delay from germ cell neoplasia in situ in dysgenetic testicular tissue; however, patient age and the position of OCT4-expressing cells within dysgenetic seminiferous tubules may be helpful in some cases [19]. In dysgenetic testicular tissue with maturation delay, the germ cells expressing OCT4 are located in the central portion of the tubule, whereas in germ cell neoplasia in situ, they have migrated toward the basement membrane. Expression of the c-KIT ligand, stem cell factor was demonstrated to be useful in distinguishing germ cell neoplasia in situ from maturation delay of germ cells in dysgenetic gonads [26]. Stem cell factor was expressed in all cases of germ cell neoplasia in situ and gonadoblastoma but not in infantile testicular tissue.

In 2016, Lepais *et al.* [24] proposed a novel histological approach for assessment of gonads of patients with a disorder of sex development. They studied 175 samples from 86 patients and analyzed gonads according to a strict histological interpretation protocol. They used the term *testicular "dysplasia"* to describe the architectural disorganization of the testicular portion of the gonad and specifically excluded the use of the term *gonadal dysgenesis* as a pathologic diagnosis. However,

there is a precedent for the use of the term *testicular dysgenesis syndrome* in reference to pathological findings, as it was previously applied to describe cases of germ cell neoplasia in situ associated with testicular environmental disorders [27]. Moreover, we believe that the terminology of Lepais *et al.* [24] is suboptimal, since the term *testicular "dysplasia"* had been used previously to describe a different testicular lesion. In the original usage, the term *cystic dysplasia of the testis* was used to describe cystic dilatation of the rete testis with secondary compression of the testicular parenchyma [28]. Furthermore, we are not aware of prior usage of the term "*dysplasia"* in patients with a disorder of sex development. In the field of pathology, the term *dysplasia* refers to alteration in size, shape, and organization of adult cells. We prefer the established term *dysgenetic testicular tissue* that refers more specifically to defective development.

Lepais *et al.* [24] identified five histological categories for the gonads of patients with a disorder of sex development: normal gonad, hypoplastic testis, "dysplastic" testis, streak gonad, and ovotestis. Criteria for dysgenetic testicular tissue, our preferred term for "dysplastic" testis, include irregular, branched seminiferous tubules, thin tunica albuginea, and fibrous interstitium [24]. Gonadal biopsies would not be adequate for use of this classification since variability from area to area in the dysgenetic gonad could result in sampling error.

"DISSECTING GONADOBLASTOMA"

Late in Dr. Robert E. Scully's career, he recognized two histological growth patterns of gonadoblastoma that were easily confused with germinoma; the first was large confluent expanded nests and the second consisted of clusters and cords [9]. He referred to this histological variant as "dissecting (infiltrating) gonadoblastoma;" however, these observations were not published during his lifetime.

In a recent investigation of 50 cases of classical gonadoblastoma, Kao *et al.* [9] described a morphological variant in 38 of these cases that they referred to as "dissecting gonadoblastoma." They considered the latter to be a variant of classical gonadoblastoma that showed similar

immunohistochemical findings. Involutional changes are less frequent in the "dissecting" variant than in classical gonadoblastoma suggesting that the latter is a more stable lesion (Figure 5A). In their model, classical gonadoblastoma was proposed to be the intermediate lesion between the cord-like/anastomosing phase and the solid/expansive form of "dissecting gonadoblastoma."

The prime importance of this variant is that several growth patterns in which the number of sex cord cells appear reduced can be misinterpreted as germinoma [9]. They described 3 patterns: solid expansive, anastomosing, and cord-like. The solid expansive and cord-like patterns, in particular, can be confused with similar patterns that have been described in germinoma resulting in more aggressive clinical treatment than necessary (Figure 5B). The application of α-inhibin or steroidogenic factor 1 immunocytochemical stains facilitates the identification of residual sex cord elements and is often useful in distinguishing "dissecting gonadoblastoma" from germinoma [9].

DISTINCTION OF UNDIFFERENTIATED GONADAL TISSUE FROM "DISSECTING GONADOBLASTOMA"

The relationship of "dissecting gonadoblastoma" to undifferentiated gonadal tissue is controversial and has not been completely resolved. Recently, it was proposed that "dissecting gonadoblastoma" either replace or be used as a synonym for the term *undifferentiated gonadal tissue* [9, 10]. In our opinion, however, it is preferable to continue to use the well-established term *undifferentiated gonadal tissue* for those cases that are considered to be the precursor of classical gonadoblastoma, as was recommended in the latest WHO classification of testicular tumours (p. 237), and to limit the use of the term *"dissecting gonadoblastoma"* to those cases with solid expansive, cord-like, or other patterns that can be confused with germinoma [7]. We do not believe that the term "*dissecting gonadoblastoma*" should be used as either a substitute or synonym for undifferentiated gonadal tissue [8].

Both undifferentiated gonadal tissue and "dissecting gonadoblastoma" occur in patients with a disorder of sex development; they differ, however, in that undifferentiated gonadal tissue is a non-neoplastic condition, whereas "dissecting gonadoblastoma" is a neoplasm with the potential for

progression to germinoma. Because of the current controversy in the literature, we will discuss in detail the clinical, macroscopic, and histological differences between the two lesions and also examine the clinical importance of making this distinction.

Macroscopically, streak gonads are elongated pale white structures located in the mesosalpinges. Classically, primary streaks occur in patients with 46, X Turner syndrome and are unaccompanied by a concomitant neoplasm, whereas undifferentiated gonadal tissue and secondary streaks occur in patients who have a Y chromosome or part thereof. By way of contrast, "dissecting gonadoblastoma" would occur macroscopically as a gonadal mass in the great majority of cases since the cases of Kao *et al.* [9] were invariably associated with classical gonadoblastoma.

Histologically, undifferentiated gonadal tissue consists of gonadal stroma that contains maturation-delayed germ cells either individually within the gonadal stroma or within immature sex cords. The individual germ cells and immature sex cords blend into the underlying gonadal stroma on hematoxylin and eosin stains (Figure 6A). Both the individual germ cells and the sex cords appear to belong in and be an element of the surrounding gonadal stroma. In contrast, the aggregates of "dissecting gonadoblastoma" are sharply demarcated on hematoxylin and eosin stains and stand out from the underlying gonadal stroma in a fashion similar to that of classical gonadoblastoma (Figure 6B). A caveat, however, is the observation that the cords in undifferentiated gonadal tissue stand out and are accentuated by immunohistochemical stains; thus, the distinction between undifferentiated gonadal tissue and "dissecting gonadoblastoma" must be made on hematoxylin and eosin stained sections in order to avoid a possible misinterpretation.

Gonadal stroma is typically the predominant component of undifferentiated gonadal tissue, whereas the germ cell and sex cord component is usually more prominent in "dissecting gonadoblastoma;" however, exceptions are not infrequent. In some instances, "dissecting gonadoblastoma" has an expansive growth pattern with proliferation of transformed germ cells; however, no desmoplastic reaction is identifiable. Clonal expansion of germ cells in "dissecting

gonadoblastoma" is the likely immediate precursor of some germinomas, and, therefore, is a helpful criterion for its diagnosis, when present. Although there is no direct association of undifferentiated gonadal tissue with malignant germ cell tumours, there is a clear association of classical, involuted, and "dissecting" gonadoblastoma to the aforementioned neoplasms.

In summation, we believe that it is essential for optimal patient care to distinguish undifferentiated gonadal tissue from "dissecting gonadoblastoma" because the former is a non-neoplastic condition whereas the latter is a neoplasm that is a precursor of germinoma in some cases. Failure to do so might result in some patients with a disorder of sex development receiving more aggressive treatment than necessary.

RELATIONSHIP OF CLASSICAL AND "DISSECTING" GONADOBLASTOMA TO MALIGNANT GERM CELL TUMOURS

Cools *et al.* [4] classified patients with a disorder of sex development into 4 risk categories, high, intermediate, low, and unknown, depending on their risk of developing a malignant germ cell neoplasm. In 60% of cases of classical gonadoblastoma, an associated malignant germ cell tumour is encountered, and in 80% of the latter cases, the neoplasm is germinoma (Fig. 7A) [29]. We are aware of 3 precursors of a malignant germ cell neoplasm in patients with a disorder of sex development. Classical gonadoblastoma represents a major precursor of germinoma [30]. However, "dissecting gonadoblastoma" is the immediate precursor of germinoma in some cases (Figure 7B). A third, less common, precursor is germ cell neoplasia in situ that arises in dysgenetic testicular tissue [4, 17, 24]. Patients with classical gonadoblastoma and germinoma sometimes develop a more aggressive germ cell neoplasm, including yolk sac tumour, embryonal carcinoma, or teratoma [31]. Germinoma occurred in 22 cases of classical gonadoblastoma that also had a "dissecting" component, and 3 germinomas were associated with another malignant germ cell component [9]. No non-germinomatous malignant germ cell neoplasm occurred in the absence of germinoma. Although the numbers are small, this observation, suggests that other malignant germ cell tumours not uncommonly arise from germinoma.

NOMENCLATURE

The term *"dissecting gonadoblastoma*" was introduced in 2016; however, its solid/expansive pattern, in particular, seems inconsistent with the term *"dissecting."* An alternative term to be considered would be *gonadoblastoma variant with unusual growth patterns*. However, we believe that in order to avoid possible misinterpretation in the literature, the choice of preferred terminology should be left to a future consensus committee. However, we believe that in order to avoid possible misinterpretation of the literature, the choice of preferred terminology should be left to a future consensus committee.

PATIENT ADVOCACY GROUPS

Patient advocacy groups currently are demanding a more conservative clinical approach whenever possible with regard to possible gonadectomy in patients with a disorder of sex development [4]. More specifically, the conservative approach includes delay of gonadectomy in patients whenever medically feasable to allow for maximum patient and parental input into gender assignment and, thus, such an approach ensures that the patient's sex assignment best fits their wishes [32]. The most important factors that influence gender assignment include an accurate clinical and histological diagnosis, the appearance of the external genitalia, the possible surgical options, the potential for fertility, the risks of gonadal malignancy, and, importantly, the gender perception of the patient and parents [32]. Full disclosure and complete involvement of the parents in making decisions concerning gender assignment and/or genital surgery must be a factor in the basic medical care for infants and young children with a disorder of sex development.

Conclusions

It is important to distinguish undifferentiated gonadal tissue from "dissecting gonadoblastoma" because patient management, risk of progression, and prognosis are distinctly

different for the two entities. Undifferentiated gonadal tissue occurs in the dysgenetic gonads of patients who have a Y chromosome or part thereof and is the precursor of classical gonadoblastoma, whereas "dissecting gonadoblastoma" is a variant of classical gonadoblastoma that can progress to germinoma.

Patient advocacy groups currently are demanding a more conservative approach whenever possible with regard to possible gonadectomy in patients with a disorder of sex development. More specifically, the conservative approach includes delay of gonadectomy in patients whenever medically feasable to allow for maximum patient and parental input into gender assignment. The ultimate functionality of the gonad is highly relevant to this decision.

Acknowledgements

LMR wrote the manuscript and LC edited it.

Conflicts of interests

No conflicts of interest are declared.

Figure legends

Figure 1. Presumptive pathway of tumourigenesis in patents having a disorder of sex development and a Y chromosome or part thereof shows origin of classical gonadoblastoma from undifferentiated gonadal tissue and malignant germ cell tumours from classical gonadoblastoma with "dissecting gonadoblastoma" as an intermediate step in some cases. The germ cells of undifferentiated gonadal tissue sometimes completely involute, and the result is a secondary streak gonad. In an alternate pathway, germ cell neoplasia in situ, another precursor of germinoma, develops within seminiferous tubules in dysgenetic testicular tissue. Rectangles with solid lines indicate disorder or tumour; broken lines, process.

Figure 2. Classical gonadoblastoma and variants. (A), Sectioned surface of the gonadal neoplasm shows yellow-tan classical gonadobastoma with minute white calcifications to the left and homogenous gray-white germinoma to the right. The spermatic cord and testicular adnexa occupy the left portion of the field. (B), Rounded islands of classical gonadoblastoma are separated by cellular gonadal stroma. Areas of dystrophic calcification are prominent to the right. (C), Islands of classical gonadoblastoma show strong nuclear expression of OCT4 in many but not all germ cells. (D), The sex cord elements in variably sized islands of gonadoblastoma show cytoplasmic expression of α-inhibin,

Figure 3. Undifferentiated gonadal tissue. (A), The convex surface of the gonad overlies cellular gonadal tissue. Sex cord elements are barely visible at this magnification. Collagenous connective tissue and fat are noted in the lower portion of the field. (B), Sparse sex cords containing germ cells and immature sex cord elements cells blend into cellular gonadal stroma. (C), The sex cords are more numerous but blend into the cellular gonadal stroma. The germ cells have abundant clear cytoplasm. (D), Cytoplasmic expression of α-inhibin highlights the sex cord elements.

Figure 4. Secondary streak gonad. (A), Note the convex surface and the cellularity of the gonadal stroma. (B), Celomic mesothelium covers a secondary streak. A thin tunica albuginea overlies more cellular gonadal stroma. (C), The gonadal stroma is cellular. (D), The identification of residual sex cord elements lacking germ cells histologically distinguishes the secondary streak from a primary streak that occurs in 46, X Turner syndrome.

Figure 5. Classical and "dissecting" gonadoblastoma. (A), Rounded island of classical gonadoblatoma with extensive dystrophic calcification in the right upper portion of the field is separated by cellular gonadal stroma from a tennis racket-shaped aggregate of "dissecting gonadoblatoma" in the center of the field. (B), Cords of "dissecting gonadoblastoma" containing germ cells with large round nuclei and clear cytoplasm and sex cord elements with small oval to spindle shaped nuclei and scant cytoplasm are separated by cellular fibrous stroma.

Figure 6. Distinction of undifferentiated gonadal tissue from "dissecting gonadoblastoma." (A), In areas of undifferentiated gonadal tissue, the individual germ cells and immature sex cords blend

seamlessly with the gonadal stroma. (B), Irregular cords of "dissecting gonadoblastoma" stand out sharply from the cellular gonadal stroma. Note the barbell-shaped cord in the central part of the field (arrow). The bar is sectioned tangentially.

Figure 7. (A), Rounded island of classical gonadoblastoma is composed of germ cells with clear cytoplasm, sex cord elements, and hyalinized nodules of basement membrane material some of which are calcified. Inset shows germinoma from the same case. The neoplasm is composed of uniform malignant germ cells separated by septa infiltrated by lymphocytes. (B), "Dissecting gonadoblastoma" with a cord-like pattern in the left upper portion of the field is separated from germinoma in the lower right by a thick wavy band of collagenous tissue.

References

1. Scully RE. Gonadoblastoma; a gonadal tumor related to the dysgerminoma (seminoma) and capable of sex-hormone production. *Cancer* 1953;**6**;455-463.

2. Scully RE. Gonadoblastoma. A review of 74 cases. *Cancer* 1970;**25**;1340-1356.

3. Cools M, Stoop H, Kersemaekers AM *et al.* Gonadoblastoma arising in undifferentiated gonadal tissue within dysgenetic gonads. *J. Clin. Endocrinol. Metab.* 2006;**91**;2404-2413.

4. Cools M, Looijenga LH, Wolffenbuttel KP, Drop SL. Disorders of sex development: Update on the genetic background, terminology and risk for the development of germ cell tumors. *World J. Pediatr.* 2009;**5**;93-102.

5. Beaulieu Bergeron M, Lemieux N, Brochu P. Undifferentiated gonadal tissue, Y chromosome instability, and tumors in XY gonadal dysgenesis. *Pediatr. Dev. Pathol.* 2011;**14**;445-459.

6. Ulbright TM, Young RH. Gonadoblastoma and selected other aspects of gonadal pathology in young patients with disorders of sex development. *Semin. Diagn. Pathol.* 2014;**31**;427-440.

7. Moch H, Humphrey PA, Ulbright TM, Reuter VE. *Classification of Tumours of the Urinary System and Male Genital Organs*. Lyon: International Agency for Research on Cancer (IARC) Press, 2016.

8. Roth LM, Lyu B, Cheng L. Perspectives on testicular sex cord-stromal tumors and those composed of both germ cells and sex cord-stromal derivatives with a comparison to corresponding ovarian neoplasms. *Hum. Pathol.* 2017;**65**;1-14.

9. Kao CS, Idrees MT, Young RH, Ulbright TM. "Dissecting gonadoblastoma" of Scully: A morphologic variant that often mimics germinoma. *Am. J. Surg. Pathol.* 2016;**40**;1417-1423.

10. Idrees MT, Ulbright TM, Oliva E *et al.* The World Health Organization 2016 classification of testicular non-germ cell tumours: A review and update from the International Society of Urological Pathology Testis Consultation Panel. *Histopathology* 2017;**70**;513-521.

11. Talerman A, Roth LM. Recent advances in the pathology and classification of gonadal neoplasms composed of germ cells and sex cord derivatives. *Int. J. Gynecol. Pathol.* 2007;**26**;313-321.

12. Kurman RJ, Carcangiu ML, Herrington CS, Young RH. *WHO Classification of Tumours of Female Reproductive Organs, 4th edition*. Lyon, 2014.

13. Ulbright TM, Young RH. *Tumors of the Testis and Adjacent Structures*. Silver Spring, MD: American Registry of Pathology, 2013.

14. Jacobsen JK, Talerman A. *Atlas of Germ Cell Tumours*. Copenhagen: Munksgaard, 1989.

15. Tsuchiya K, Reijo R, Page DC, Disteche CM. Gonadoblastoma: Molecular definition of the susceptibility region on the Y chromosome. *Am. J. Hum. Genet.*1995;**57**;1400-1407.

16. Lau Y, Chou P, Iezzoni J, Alonzo J, Komuves L. Expression of a candidate gene for the gonadoblastoma locus in gonadoblastoma and testicular seminoma. *Cytogenet. Cell Genet.* 2000;**91**;160-164.

17. Cools M, Drop SL, Wolffenbuttel KP, Oosterhuis JW, Looijenga LH. Germ cell tumors in the intersex gonad: Old paths, new directions, moving frontiers. *Endocr. Rev.* 2006;**27**;468-484.

18. Li Y, Vilain E, Conte F, Rajpert-De Meyts E, Lau YF. Testis-specific protein Y-encoded gene is expressed in early and late stages of gonadoblastoma and testicular carcinoma in situ. *Urol. Oncol.* 2007;**25**;141-146.

19. Cools M, van Aerde K, Kersemaekers AM *et al.* Morphological and immunohistochemical differences between gonadal maturation delay and early germ cell neoplasia in patients with undervirilization syndromes. *J. Clin. Endocrinol. Metab.* 2005;**90**;5295-5303.

20. Jørgensen N, Muller J, Jaubert F, Clausen OP, Skakkebaek NE. Heterogeneity of gonadoblastoma germ cells: Similarities with immature germ cells, spermatogonia and testicular carcinoma in situ cells. *Histopathology* 1997;**30**;177-186.

21. Kersemaekers AM, Honecker F, Stoop H *et al.* Identification of germ cells at risk for neoplastic transformation in gonadoblastoma: An immunohistochemical study for

22. Hersmus R, Kalfa N, de Leeuw B *et al.* FOXL2 and SOX9 as parameters of female and male gonadal differentiation in patients with various forms of disorders of sex development (DSD). *J. Pathol.* 2008;**215**;31-38.

23. Looijenga LH, Hersmus R, Oosterhuis JW, Cools M, Drop SL, Wolffenbuttel KP. Tumor risk in disorders of sex development (DSD). *Best Pract. Res. Clin. Endocrinol. Metab.* 2007;**21**;480-495.

24. Lepais L, Morel Y, Mouriquand P *et al.* A novel morphological approach to gonads in disorders of sex development. *Mod. Pathol.* 2016;**29**;1399-1414.

25. Gondos B, Riddick DH. *Pathology of Infertility : Clinical Correlations in the Male and Female*. New York: Thieme Medical Publishers, 1987.

26. Stoop H, Honecker F, van de Geijn GJ *et al.* Stem cell factor as a novel diagnostic marker for early malignant germ cells. *J. Pathol.* 2008;**216**;43-54.

27. Skakkebaek NE, Rajpert-De Meyts E, Main KM. Testicular dysgenesis syndrome: An increasingly common developmental disorder with environmental aspects. *Hum. Reprod.* 2001;**16**;972-978.

28. Nistal M, Regadera J, Paniagua R. Cystic dysplasia of the testis. Light and electron microscopic study of three cases. *Arch. Pathol Lab Med* 1984;**108**;579-583.

29. Clement PB, Young RH. *Atlas of Gynecologic Surgical Pathology*. 3rd ed. London: Saunders/Elsevier, 2014.

30. Pauls K, Franke FE, Buttner R, Zhou H. Gonadoblastoma: Evidence for a stepwise progression to dysgerminoma in a dysgenetic ovary. *Virchows Arch.* 2005;**447**;603-609.

31. Simon RA, Laughlin TS, Nuccie B, Wang N, Rothberg PG, Wang X. A 46 XY phenotypic female adolescent with bilateral gonadal tumors consisting of five different components. *Int. J. Gynecol. Pathol.* 2008;**27**;407-411.

32. Mendonca BB. Gender assignment in patients with disorder of sex development. *Curr. Opin. Endocrinol. Diabetes Obes.* 2014;**21**;511-514.