MASCULINIZING OVARIAN TUMOR; AN ARRHENOBLASTOMA

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The purpose of this paper is to report a case of arrhenoblastoma. It intends to describe the diverse histologic patterns within one and the same tumor, as well as present the close histogenetic relationship existing between cells of the Sertoli and Leydig types appearing in the tumor, chiefly as regards their modes of differentiation in the neoplastic tissue.

CASE REPORT

A. W., twenty-five year old married woman, was admitted to the hospital with a complaint of indisposition in November 1954. Previous to 1952, menstruation appeared regularly at intervals of 28 days.

She had one full term normal pregnancy, giving birth to a boy in February 1952. Menstruation recurred in April and took a regular course until October 1952.

The second pregnancy was interrupted by an artificial abortion conducted in December of that year. Thereafter small amounts of genital bleeding ocurred almost daily for about half a year. In July 1953 she was diagnosed by a docter as metropathia hemorrhgica and treated with a hormone preparation (estrogen) for one and one half month, given every other day.

Bleeding ceased but amenorrhea occurred again from September to November 1953. She was treated again by another docter with a hormone preparation under the diagnosis of ovarian dysfunction for about 50 days.

The last menstruation occurred on June 18, 1954, and was followed by complete amenorrhea which led to a misdiagnosis of pregnancy to be made. The family came to believe that the baby in the womb would be a male judged from the masculine taint in her facies, that appeared gradually since about August, —a Japanese folk-belief says that if the countenance of a pregnant woman changes from "womanly" to "manly", the baby in the womb would more probably be a male.— At the same time a moderate degree of hirsutism began to be noticed, so that she had to shave her whiskers daily. Further she came to have abundant acne eruptions in the forehead and around the mouth. Her breasts became more flattened and her well developed breast curve began to be lost.

As a whole, obese as she was especially when about 18 years of age, she

Received for publication March 14, 1956.

started to lose fat gradually and became muscular and rather lean especially during the last one year.

She was of middle build, light brown in colar, muscular with scanty fat and looked masculine, especially around the eyebrows and eyes, accompanied by a moderate growth of moustache and whiskers. The breasts appeared flat. The dense pubic hair extended far up to the abdomen and the enlarged clitoris was found to be about 4 times the average size. Her voice was rather deep. Acne eruptions had almost disappeared by the time she was seen.

Pelvic examination revealed a movable tumor, the size of a fist, on the left side. No other findings mentionable and no pregnancy were noted.

Operation was performed on 26th, November 1954. The right ovary was normal, the uterus looked slightly small. Otherwise nothing pathological was present in the pelvic organs except for the left ovary.

Postoperative course was good and menstruation was reestablished from 20th, December 1954. Recurrence or metastasis has not occurred so far.

PATHOLOGICAL FINDINGS

The tumor was spherical, of the size of a fist about $8\times6\times6$ cm, and enclosed by a smooth glistening tenacious capsule.

Consistency was relatively soft as a whole, but the cut surface revealed soft and firm parts mingled with hemorrhagic areas partly though almost solid throughout, with colours ranging from yellow to brown.

There were no large cystic changes but small ones.

Histological picture of this tumor varied from one area to another.

The outermost part of the tumor was occupied by thick collagenous fibers provided with scanty fibrobalstic cells.

The neoplastic cells grew into the loose edematous stroma composed of relatively small spindle cells with darkly staind nucleus that were scattered to form a net work under the pseudocapsule. The findings presented as chief patterns that transfer each other in places, are described below;

1. Pattern in which cord-like arrangement of epithelial-like cells predominated (Fig. 1), (Fig. 9), (Fig. 10).

There were irregular cell columns assuming a zig-zag course anastomosed with and bordered by small amount of spindle-shaped cells among which irregular polygonal to somewhat round shaped cells of relatively large size with eosinophilic cytoplasm containing granules were present. The cells composing the cell-columns were relatively tall or rather cylindric with scanty protoplasm. The cell possesed a not so clearly definable borderline and its cytoplasm was soft, fragile and looked flocculent in fixed material. The outline of the individual cell was recognized with difficulty, though its polarity was maintained.

The nuclei were elongated oval, more or less curved in, and with abundant chromatin. The nucleolus was single and occassionally visible. The nuclear membrane was thin and soft, containing very fine granular chromatin distributed diffusely. The nucleus appeared to retain slightly some characters resembling a vesicular nucleus. Mitotic figures were rarely found.

The cells were arranged parallel to the long axis of the nucleus *i.e.* vertical to the basis, to form a cord or tubule-like structure, so that in cross sections, there were seen as rosettes, in places, whose central part was occupied by a faintly eosin stainable mass. No clearly acceptable duct formation was seen here.

The cells, much less in number, occupying the space surrounding the cell-cords were almost fusiform and possesed elongated oval nuclei of character very similar to that of the former. In some areas, it was difficult to distinguish one from the other when an individual cell was observed.

Some of these fusiform cells appeared to be transforming themselves into the large polygonal eosinophilic cells with round vesicular nuclei mentioned above by way of short spindle cells.

Structural relation between the cells composing the cell-column and the surrounding cells was not so intimate in some parts so that tissue crevices were found.

In addition to these type cells, there were seen much more loosely arranged small spindle cells whose appearance were embryonal conective tissue cells and one layer of capillary endothelial cells accompanied.

2. *Pattern with well marked characters of epithelial cells* (Fig. 2), (Fig. 11).

The cuboidal cells with round clearly vesicular nuclei composed the cell-columns and did not accompany the large polygonal eosinophilic cells in the space but smaller spindle cells.

The cells were more cuboidal than the columnar cells of the former pattern. They had a round or slightly oval nucleus with scanty chromatin and one distinctly visible nucleolus. The cytoplasm was relatively rich, faintly stained with eosin, looked clear, soft and fragile. The outline was recognizable by the faintly discernable cell membrane. The cell columns were surrounded by small amounts of relatively small spindle cells with nuclei rather rich in chromatin. There were no longer any polygonal eosinophilic cells between the cell-cords, while edematous stroma could be seen in some parts. Distinct duct formation was also not seen but the cells revealed a more epithelial character and a tendency to form tubules.

3. *Pattern composed of fusiform cells of immature type predominating; sarcoma-like pattern* (Fig. 3), (Fig. 12).

Inversely proportional to the epithelial-like cells, the fusiform cell with nuclei similar in character to that of the former increased in number and were arranged in bundle-forms separated by capillaries accompanied. The whole appeared like the histologic picture of a sarcoma but cell polymorphism was less in degree and not so very anaplostic, even though these cells showed immature types. In some parts, the central portion of the bundle showed more or less a tendency to transform from fusiform cells to epitholial-like cells of immature type. No large eosinophilic cells were found here, and the cells were of a highly undifferentiated type.

4. A modified type of the first pattern showing much resemblance to an embryonal gonad (Fig. 4).

With an increase in the epithelial-like cells arranged as cell cords, they grew into multilayered cell strata enclosed by fusiform cells with tissue crevices around themselves. When the whole is seen, in section, it resembled renal glomeruli to a certain degree, though they formed only irregular cell clumps, or resembled more a developmental stage of the testis. This pattern could be shown as a derivative of patterns 1 and 3.

5. Pattern chiefly composed of intermediate cell type between the above epithelial-like and fusiform cells and seen as an alveolar structure or in sheet. (Fig. 5), (Fig. 6).

The main cells looked epithelial in character from their structural arrangement but differed from the cells mentioned in the former patterns (Fig. 13). The shape of the cells was rather short spindle, polygonal to slightly plump with scanty protoplasm. The nucleus was round to oval with a lesser amount of chromatin and an almost always visible nucleolus within a thin nuclear membrane. The character of the main cells showed intermediate types to epithelial-like and fusiform cells and were more or less matured. In addition to these cells, large polygonal or cresent shaped eosinophilic cells with slightly eccentric round vesicular nucleus appeared in groups mingled with the former. Transition types to the main cell could be traced in some areas. These large eosinophilic cells were very similar to the cells that appeared arround the cell-cords in the first pattern and they were identical, by the cell characters.

They together with the above described cells grew into broad hands forming an alveolar structure of variable amount or as a sheet, formed by loosely arranged edematous stroma with vessel elements.

6. Pattern composed chiefly of more clearly definable spindle cells with large eosinophile cells in groups (Fig. 7), (Fig. 8).

The more well definable spindle-shaped tumor cells grew diversely, in places, aggregating together to islet form or scattered about in the very loose edematous stroma composed of small spindle-shaped young cells interpreted as fibrolastic cells with very thickly stained nucleus. In some areas, a clear cut appearance of the large polygonal to cresent shaped eosinophilic cells were seen in groups in contact with each other, some of them almost fused, among the spindle cells.

In the area composed of more loosely distributed tumor cells they sometimes appeared round or oval in shape separately (Fig. 14). Often with transition types to the spindle-shaped tumor cells. Some of these large eosinophile cells with granules contained rod-like crystalloids in the protoplasm (Fig. 15). Distribution of these cells was closely correlated to that of tumor cells too. Blood vessels were present that possessed edematous walls of somewhat swollen cells.

Besides, some small cysts surrounded by flattened tumor cells were found here and there (Fig. 7).

These several main histologic patterns were transformable from one to the other, evidenced by the pictures shown in the tumor. Hemorrhage occurred diffusely in some areas, but no necrotic area was seen at all.

In the capsular region several of the tumor cells were found in groups or scattered amoung thick collagenous fibered strata (Fig. 16). The polygonal eosinophilic cells appeared in very small numbers with clearly visible transition type cells to short spindle and to rather cuboidal tumor cells. No ovarian tissue was present.

COMMENT

That this case belongs to a type of arrhenoblastoma is well ascertainable from the clinical course and from the histological findings described above. The findings showed that in spite of the diverse histologic pictures presented, the chief components of the tumor are immature type cells that tend to differentiate into two directions *i.e.* to epithelial cell character and to stromal cell character —the latter does not mean cells composing the stroma in this case—. They show variable differentiation phases and variable structures which are sarcoma-like or carcinoma-like, etc. but in some areas they appear similar to a stage of embryonal gonad. These immature type cells are represented as epithelial-like cells and fusiform cells in the first and third patterns, and these two are alike in character especially in nuclear appearance, and are very similar to each other as regards the individual cell. They are interpreted as being the same in nature, in a highly undifferentiated phase (Fig. 3).

The epithelial-like cells grow, decreasing in the basophility of the cell body and nuclear chromatin, and show increase in nuclear size and amount of protoplasm, to become cuboidal cells with round vesicular nucleus, with a tendency to form tubules. The latter can most probably be interpreted as Sertoli cells (Fig. 11).[5)2)]

The fusiform cells appearing sarcoma-like as in Fig. 3 are transformed to more clearly definable spindle-shaped cells which may aggregate in islet form or scattered in the very loose stroma and intermingled with it, as shown in Figs. 7 and 8.

These cells resemble the cells composing the stroma. Both are spindle in shape, the question arises as to how to discriminate the tumor cells from the young fibroblastic cells in this area morphologically. Generally, the nuclear character of the former shows a more or less large slightly ovoid nucleus of thin fine unclear membrane containing very fine diffusely distributed chromatin tending to make visible a nucleolus, that is resembled rather more the nucleus of the cell found in Fig. 5, if it is compared with the latter whose nucleus is more sharp spindle in shape and contains very darkly stained chromatin, as seen in the central portion of Fig. 2. However the close resemblance between the spindle-shaped tumor cells and young fibroblastic cells may produce a problem concerning the immature cell type and histogenesis of this tumor, depending on the interpretation of the latter.

The cells called intermediate type by the author and shown as the main component of patter 5, can be derived from the immature cells of the neoplasm formerly mentioned and are rather more matured, but they have the peculiarity with a tendency to retain an epithelial character resembling that of cells belonging to the Sertoli cell series, as well as a possibility to transform them-

selves into large polygonal eosinophilic cells (Fig. 6). Moreover they are very much similar to the spindle-shaped tumor cells scattered in the stroma which are also transformable into the large eosinophilic cells (Fig. 8).

Now letting aside these somewhat complex modifications in the appearance mode of the large polygonal eosinophilic cells, they may belong to cells of the stromal cell character, *i.e.* originate from the spindle or short spindle shaped cells in general. They can be interpreted as being identical to the Leydig cells of the testis by their large eosinophilic granular cytoplasm containing rod-like crystalloids (Reinke), Sudan III positive substance, sometimes by the vacuoles, and by their eccentric round vesicular nucleus, in spite of the shape and mode of distribution they may be variable (Fig. 15).[15)14)17)]

The shapes varied from round, oval to crescent-like columnar when in groups, or polygonal respectively, depending on the situation in which the cells appeared. The appearance may also depend somewhat on the distribution mode of tumor cells which are their precursors. When these tumor cells are distributed loosely, they appear round or oval, and when compactly distributed, they appear as cresents in a group and in contact with each other often with cracks, and occasionally fused or form a cord. In addition, these shapes may also be due to the maturing phase of the cells or functional phase to some degree, for example, if old they tend to fuse, their cytoplasm may become more abundant and grossly granular or vacuolated, become ill definable in boundary or may sometimes contain rod-like crystalloids of Reinke. Besides, in some places, their appearances are characteristic and appear as if several of the cells arise as abrupt transformations without any transitional type cells (Fig. 7). This can be explained by the analogy of the basal cell carcinoma undergoing clear cut cornifiction in very limited areas showing almost no transitions. At any rate it is evident that the tumor cells can differentiate themselves into the "Leydig cells." So it is reasonable to think that that these "Leydig cells" preserve their neoplastic character even in such a differentiated, or mature phase. Of course they are not derivatives of non-neoplastic stroma cells by induction of tumor cells, nor hyperplasia of the remaining elements of the ovary. The loosely arranged myxomatous small spindle cells interpreted as young fibroblastic and vessel elements compose the stroma of this tumor, though these cells look embryonic and almost indistinguishable from the tumor cells in some area, morphologically, and in this case, it is less reasonable to assume that these stroma cells give rise to the above mentioned epithelial-like cells or the fusiform cells.

On histogenesis of the Leydig cells of the testis in normal development, there are different views that they have their origin in fibrous connective tissue, of epithelial, and of double origin according to phase of development, that is, the primary, epithelial, secondary, of connective tissue.[15)] The former may be generally accepted.[7)] But the results from the tumor observed here support the double origin theory with a slight modification of the term epithelial-like instead of epithelial.

As the neoplasm can represent a duplication of some developmental stage of the tissue from which the tumor is derived, even though in modified or abridged form, the large eosinophilic cells above mentioned which are, in this

case, evidently derived from immature fusiform or epithelial-like through spindle or "intermediate" type cells, can be interpreted as a homologue of the primary Leydig cells of the testis, because it cannot be denied that Leydig cells of the testis of postnatal life are derivatives of the stromal connective tissue cells. For the peculiarity of their mode of appearance in this tumor and their assuming a more epithlial character, this assumption may be useful for an explanantion. Now, the afore-mentioned fact that these tumor cells undergo differentiation into the cells of the Sertoli type as well as into cells of the Leydig type is worthy of note. In other words, the main components of this tumor are immature cell materials possesing bipotency to differentiate, *i.e.* a common precursor from which both types cells are derived.

In spite of some inconsistencies in opinions regarding the detailed histogenesis of Sertoli and Leydig cells in normal development, the common source of both type cells may be ultimately traced to the sexual coelomic mesenchymal cells, and if they appear sometimes like connective tissue cells morphologically, they should already have the special characters of an early developmental stage. It may be related to this bipotency of the cells that areas of this tumor where the Sertoli cell character is prominent, have a lesser amount of Leydig cells, while areas where the latter is abundant show often spindle shaper cells of characters resembling less the Sertoli cells. The presence, in the same tumor, of these two kinds of cells and their precursor of neoplastic character, will lead one to think that this case tumor is embryonic in type, similar to a nephroblastoma whose cells undergo differentiation into epithelial tubules and the mesenchymal tissue, and to meduloblastoma that has a bipotency to differentiate into both neuroblastic and spongioblastic series, even though this case occurs in more aged woman.

If the tumor cells become differentiated and matured only in one direction along the aforementioned series, or derived from one sidely differentiated cell material, the tumor may show a type of Sertoli cell tumor (Testicular adenoma of pick) or Leydig cell tumor of the ovary, respectively.[17) 9)]

Accordingly it is reasonable that tumors composed of possible variations in amount of these two types cells and of immature type cells, should be belonging to the same group.

As types of arrhenoblastoma, undifierentiated, intermediate and mature are used for classification in general and the last is represented by the testicular adenoma type. That the case discussed here belongs to the intermediate type is apparent.

If this classification method is used, in addition to the above three types of tumor belonging to the arrhenoblastoma group, it is reasonable to add another to the mature type, namely the "Leydig cell tumor" by reason of the previously mentioned histogenetic relationship made clear from an interpretation of this tumor.

The view that Sertoli-cells and granulosa cells are homologues, and Leydig cell and hilus cell or stromal (Lutein) cell are equivalent in male and female has been supposed also by the author,[1)] and this tumor, especially the pattern composed of epithelial-like cells arranged in columns resembles that of a type of granulosa cell tumor to a certain degree, so in a highly undifferentiated stage,

they may be more closely related. The masculinizing phenomenon are to be ascribed to the overgrowth of Leydig cells that are neoplastic in nature, and to its functioning. At least the cells of Leydig type with the crystalloid of Reinke play an indispensable role.

As terminology, the term "arrhenoblastoma" introduced by R. Meyer seems to have a rather wide concept which represents a group of tumors, and G. Teilum's "androblastoma" also a wide group that includes the former.

G. Teilum left the term arrhenoblastoma for the group of tumors which are characterized clinically as virilizing, so the term is defined by its clinico-pathological contents. By his assertion, therefore, arrhenoblastoma should contain Leydig cell tumor of the ovary too.

From the morphological stand point the author would like to use the term arrhenoblastoma for the tumor of so-called intermediate type or mixed type belonging to the arrhenoblastoma group, and for mature type the term Sertoli cell tumor and Leydig cell tumor of the ovary respectively.

SUMMARY

1. A case of Arrhenoblastoma of intermediate type with typical clinical symptoms is reported with illustrations of the several histologic patterns.

2. From histological study, it is reasonable to think that this tumor originates from the cell material that has bipotency to differentiate into both Sertoli and Leydig type cells.

3. When it takes only one course of differentiation or is derived from the cell material being differentiated into one direction only, it may show a Sertoli cell tumor or Leydig cell tumor of the ovary.

4. This case, an intermediate type of arrhenoblastoma, is considered to be an embryonic type tumor as nephroblastoma, morphologically though it occur in a more aged woman.

5. Masculinizing symptoma are ascribed to the Leydig cell growth in the tumor.

6. The Leydig cell tumor of the ovary should be classified as the arrhenoblastoma group as well as the Sertoli cell tumor by reason of the above mentioned.

The author wishes to express his deep gratitude to Prof. Dr. Fukuzo Oshima for his encouragement and to Dr. R. Nakamura for the chance to study this case.

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EXPLANATION OF FIGURES

FIG. 1. (Pattern 1). Cord-like arrangement of epithelial-like cells, borderd by a small amount of spindle shape cells among which irregular polygonal relatively large eosinophilic cells are present.

FIG. 2. (Pattern 2). Cuboidal cells with vesicular nuclei compose cell columns without acoompanying large polygonal eosinophilic cells. The central portion of the picture shows edematous stroma.

FIG. 3. (Pattern 3). Fusiform cells of undifferentiated type arranged as an irregularly distributed bundle with the capillaries. Transitional picture to the first pattern suggested by the cell arrangement.

FIG. 4. (Pattern 4). Cell clumps enclosed by fusiform cells which are more matured than that shown in Fig. 3, are characteristic. The whole resembles a stage of development of the testis.

Fig. 5. (Pattern 5). The intermediate type cells between the epithelial-like and the fusiform cells ars growing as alveolar structure or in sheets. The large polygonal or crescent shape cells with round vesiculsr nucleus appear in groups or are mingled with the former.

FIG. 6. (Pattern 5). Tumor cells of "intermediate type" arranged in alveolar structure among the loose edematous stroma the fomer are transforming themselves into the large eosinophilic cells.

FIG. 7. (Pattern 6). Well defined spindle shaped cells growing in sheets among which large eosinophilic cells appear in groups. Small cysts are shown.

Fig. 8. (Pattern 6). Spindle shape tumor cells scattered in the loose edematous stroma. Discrimination of the stroma cells from tumor cells becomes difficult, as in dividual cell. Note round or oval eosinophilic cells with round vesicular nucleus transformed from the short spindle cells in the right lower portion.

FIG. 9. High power magnification of undifferentiated epithelial like cells shown in pattern 1.

FIG. 10. High power magnification of Fig. 1.
Rather more differentiated cells in "Sertoli" cell series, showing cord-like arrangement.

FIG. 11. High power magnification of a part of Fig. 2.
Cuboidal cells with round vesicular nuculeus arranged into cord or tubule formation. The cells are interpreted as "Sertoli" cell.

FIG. 12. High power magnification of a part of Fig. 3.
Undifferentiated spindle shaped tumor cells show sarcoma-like pattern.

FIG. 13. High power magnification of a part of Fig. 5.
"Intermediate" type cells show alveolar structure. Some of these cells are being transformed into large eosinophilic cells with an eccentric nucleus.

FIG. 14. Appearance mode of the large eosinophilic cells from precuorser cells. Short spindle tumor cells, being loosely arranged.
Note. The cells show round or oval shape with round vesicular nucleus less granules and no crystalloid, but these cells are transferred further to cells interpreted as "Leydig" cells shown Fig. 15.

FIG. 15. Large eosinophilic cells with a round vesicular eccentric nucleus and granules identitical to "Leydig" cell. *Note*: Utmost right side cell containing the rod-like crystalloid of Reinke. The utmost left cell also contained crystalloid.

FtG. 16. Peripheral part of the tumor. Cuboidal shaped tumor cells in columus among thick collagenous stratum.

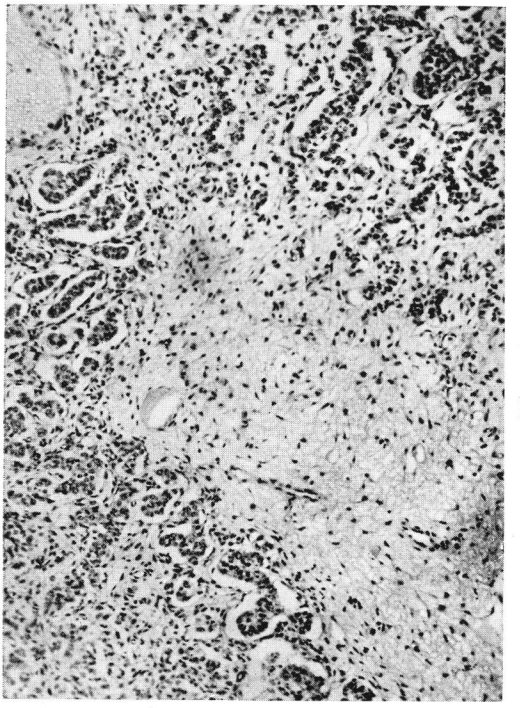

FIG. 2

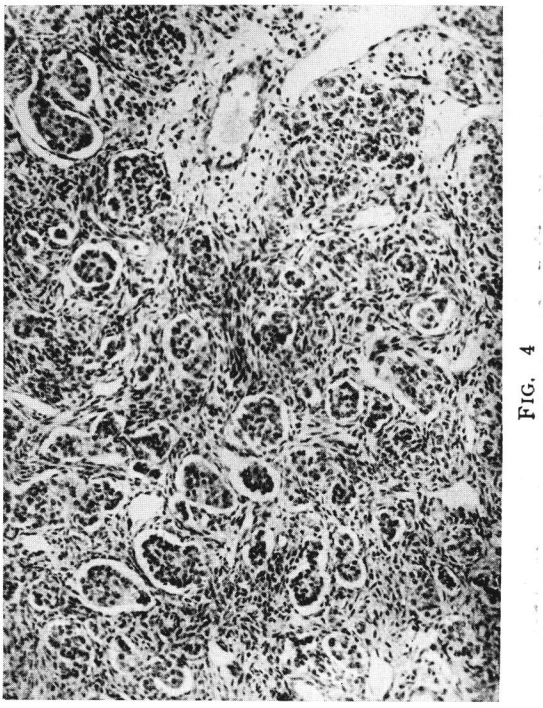

FIG. 4

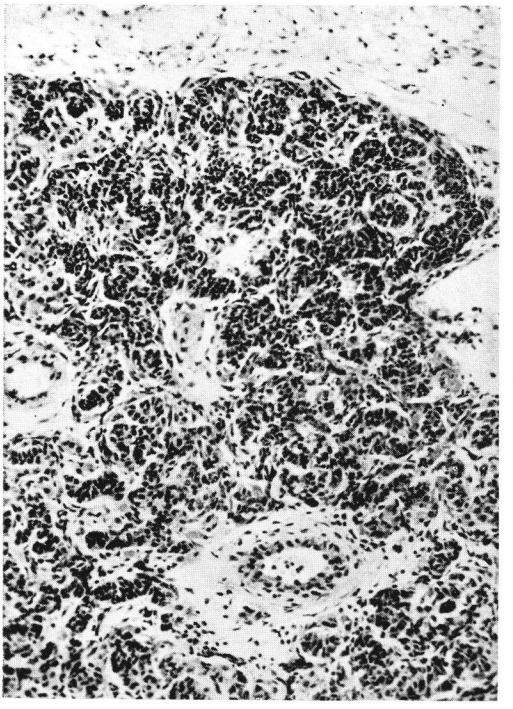

FIG. 1

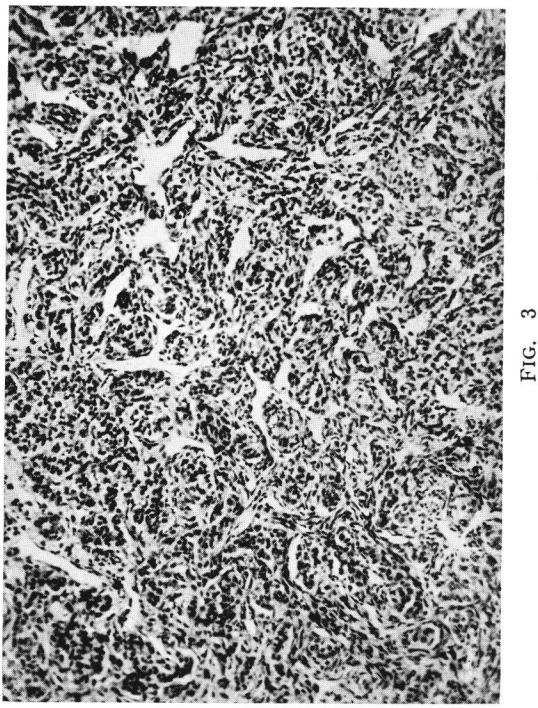

FIG. 3

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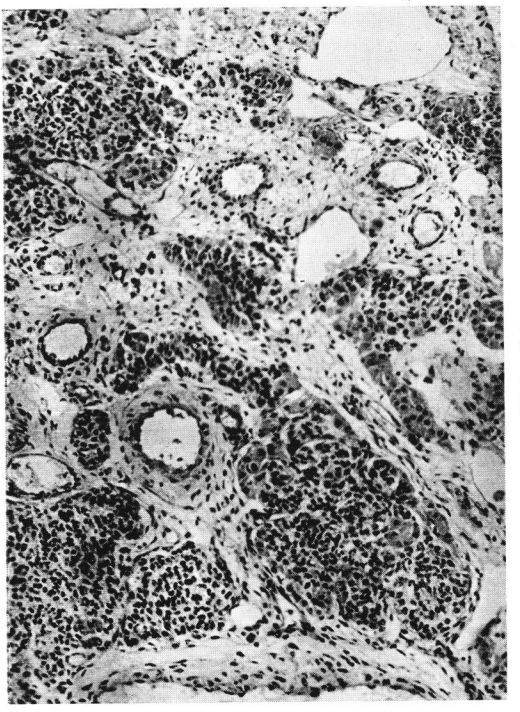

FIG. 6

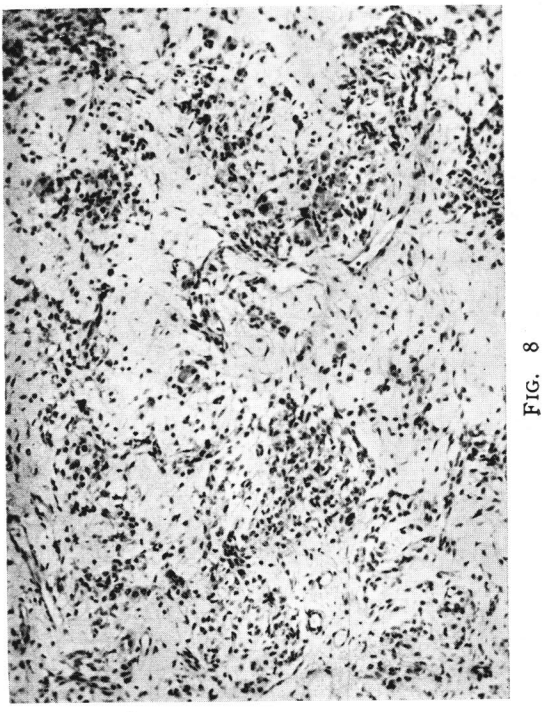

FIG. 8

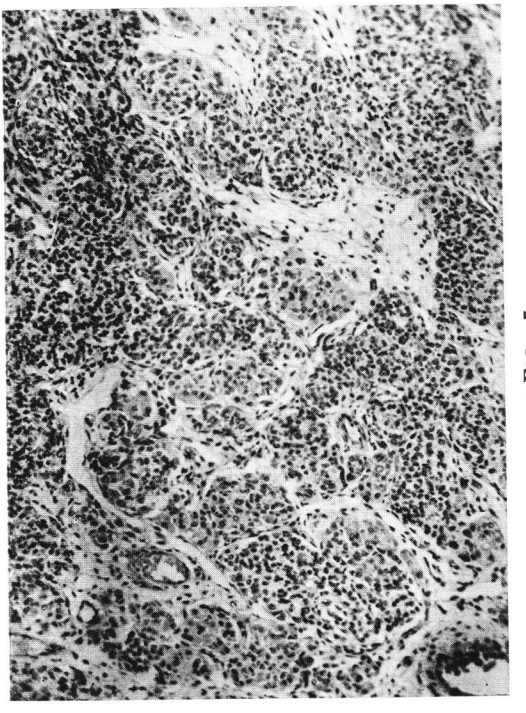

FIG. 5

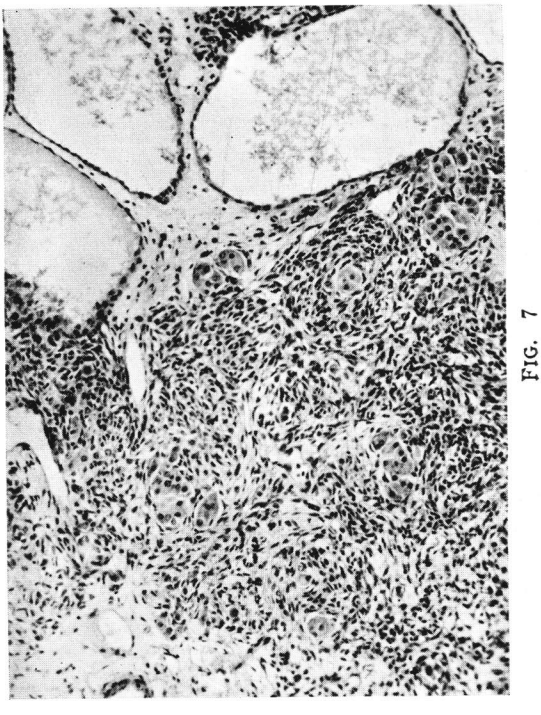

FIG. 7

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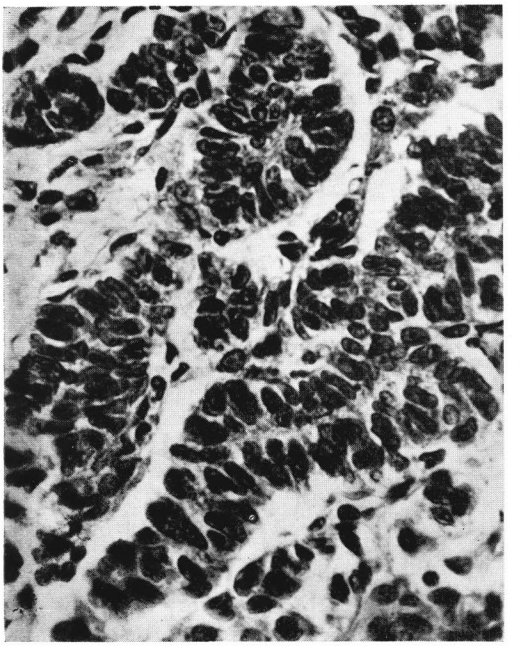

FIG. 9

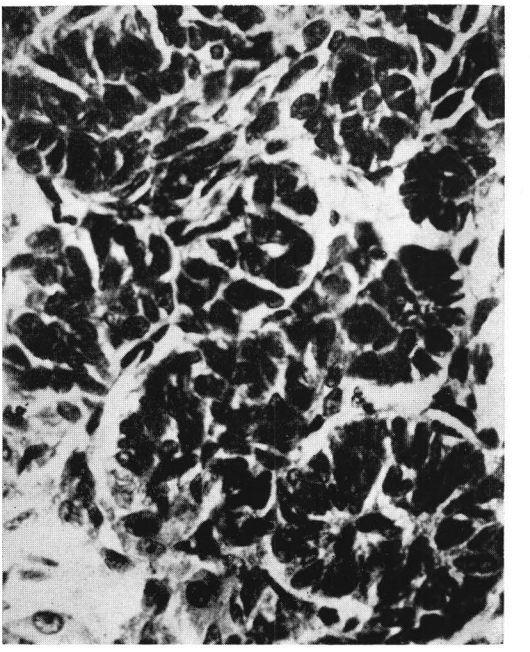

FIG. 10

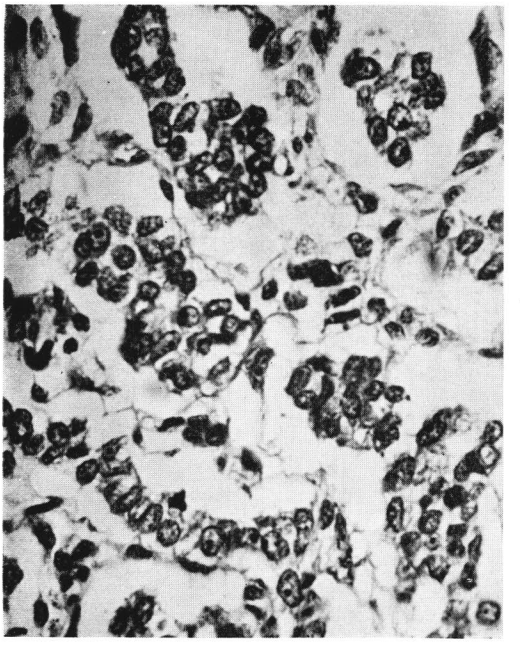

FIG. 11

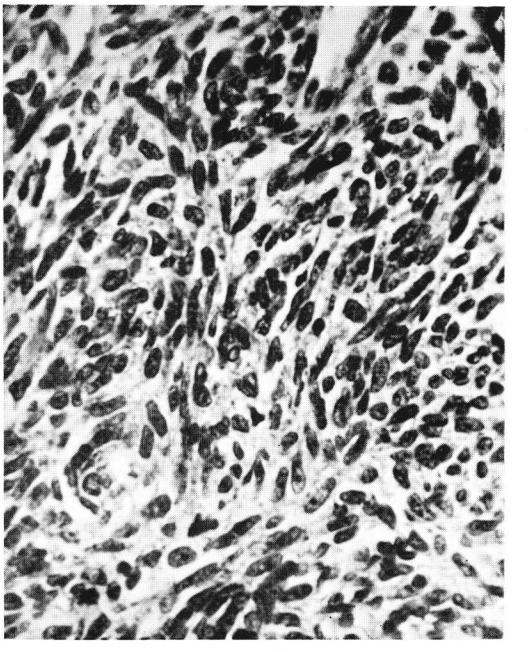

FIG. 12

Hirosi Usizima: *Masculinizing Ovarian Tumor; an Arrhenoblastoma*

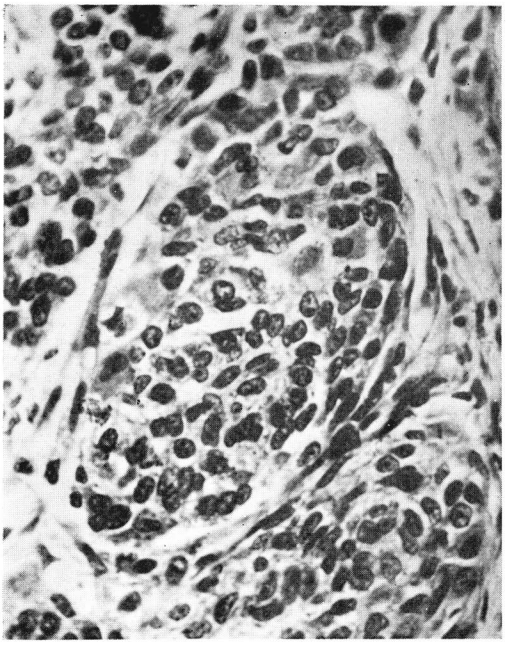

FIG. 13

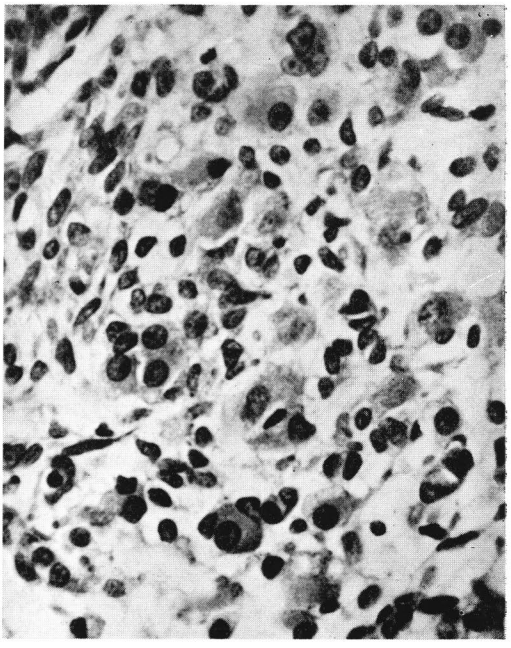

FIG. 14

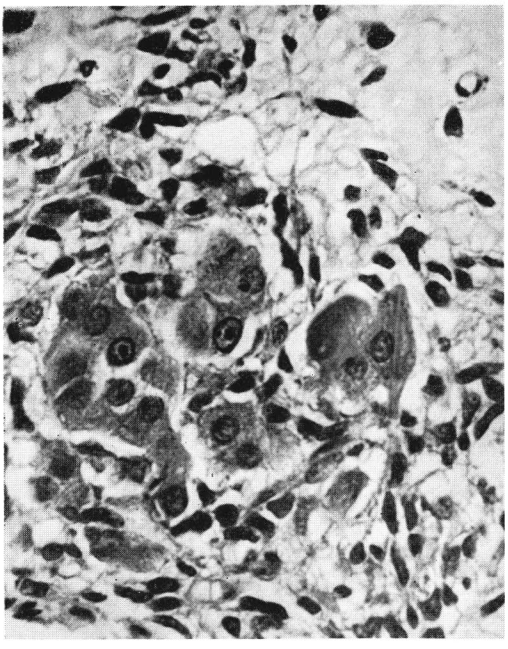

FIG. 15

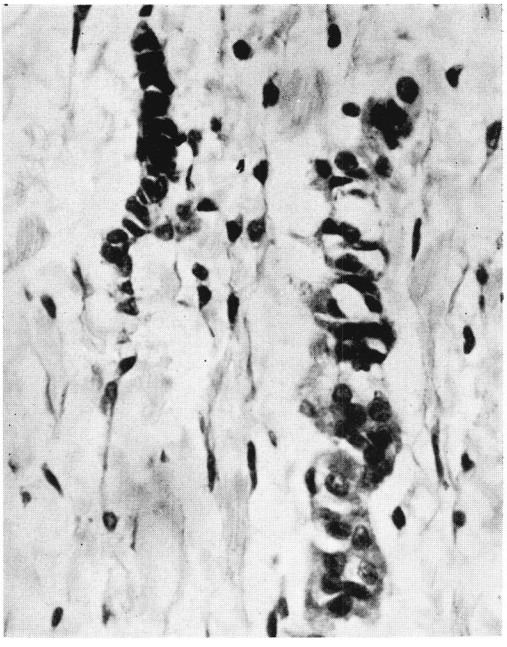

FIG. 16