Observations on the Early Development of the White Rat (Rattus Norvegicus Albinus)

by

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I. INTRODUCTION

Although there is considerable literature on the early development of the albino rat (Rattus Norvegicus Albinus), ova which have not yet entered the oviduct and certain maturation stages have not been described, as far as the author can ascertain. The short space of time that elapses between some of the maturation stages makes it difficult to isolate them. The interval between the pronuclear stages and the two-cell stage, apparently is a comparatively short one. Added to this difficulty the exact time of ovulation is never exactly known.

An effort was made to record exact times of insemination in order to make as accurate a comparison as possible in the development of the ova at particular ages. At the present time the author has not enough material to record the exact time of beginning and ending of all the maturation and segmentation stages in the rat. This investigation will have to be left for a future time.

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2. MATERIALS AND METHODS

Materials for this work were obtained from albino rats of the colony belonging to the anatomy department of the University of Kansas. A female rat to be mated is placed in a cage with one or two males. If the female is in heat, copulation usually takes place within a few minutes. If she is not in heat, it soon becomes evident and the procedure is again tried in 24 hours. The male rat is not prostrated by the sexual act; the same male may serve for many copulations which may take place during a period of several hours. The female will accept the male many times before she begins to fight him away. According to Widakowich (1910),"a female rat permits many males to copulate in the course of many hours, receiving males 30 times or oftener, when suddenly she drives them away".

Sobotta and Burkhard (1910) paired female rats that were a few hours post-partem, since it has been observed that many mammals ovulate soon after parturition. Huber (1915) paired female rats that had borne at least one litter and in the majority of females used, about thirty days elapsed after the birth of a litter before pairing as the rat will then be free from "domestic cares". Female

rats used in this work were paired at random but none were paired unless at least thirty days had elapsed after parturition.

In the work described in this paper the male rat was placed in the cage with the female in heat; copulation began in a few minutes and occurred 15 or 20 times within the next 40 minutes. This time, i.e., 40 minutes after the rats were put together in the cage, was noted on a card which was attached to the cage and is used as the date of reference in estimating ages of ova and embryoes. It is referred to in this paper as "the time of completed insemination". This time as the reference point was chosen rather than the time of the first copulation which Huber (1915) used, since in many cases the first copulation is not complete, as evidenced by an examination of the vagina. After several copulations, the chances are much greater that sufficient sperm have been introduced into the female genital tract to insure fertilization; it is probably unnecessary to permit further copulation.

The term 'insemination' is that used by Long and Mark (1911) to indicate "the introduction of male sexual elements into the genital tracts of the female by act of coitus or otherwise".

The inseminated females were killed with cholorform. The ovaries and oviducts were care-

fully removed while being bathed with warm Ringer's solution to prevent as much alteration in the tissues as possible during the operation. The ovary and oviduct are almost completely surrounded by a mass of fat. As much fat as possible was dissected away without disturbing the periovarian capsule. The uterine horn was first tied off to preserve the contents of the tube, then divided in most cases three or four centimeters from the oviduct. Sections of ovaries and oviducts were made ranging in time from 10 to 87 hours after the time of completed insemination. Some of the material was fixed in Carnoy's solution and some in Bouin's. In using Carnoy's solution it is necessary to wash in absolute alcohol. Carnoy's sulution was found not as satisfactory as Bouin's as tissues were too hard for proper sectioning. The sections were cut at a thickness of seven microns and stained with Delafield's hematoxylin and eosin.

3. OVARIAN OVA

Description of the later development of ovarian ova from the rat and mouse has been very well done by Sobotta (1895) and Sobotta and Burkhard (1910). According to these investigators, the first polar body is formed before the ovum leaves the ovary. This polar body is lost during the passage of the ovum to the oviduct, but in the

mouse it may still persist with the ovum after it reaches the oviduct. These investigators describe the spindle for the second polar body as following that for the first without a resting stage in the rat and mouse and forms while the ovum is still in the follicle. The metaphase for the second polar body starts after penetration of the spermium into the ovum. Therefore, the monaster for the division of the second polar body persists for a considerable period, probably from 10 to 15 hours, since ovulation occurs near the time of copulation and it takes from 10 to 12 hours for sperm to reach the ampullar portion of the oviduct.

Ovulation in the rat and mouse occurs spontaneously regardless of copulation. Sobotta and Burkhard (1910) record in the rat five cases without copulation in which ova were seen in the oviduct.

This description by Sobotta and Burkhard corresponds with stages found in my work on the rat. An ovarian ovum found in an ovary of a rat of my series which had been inseminated 27 hours, shows an early anaphase stage in the formation of the first polar body. Sobotta and Burkhard (1910) record seeing the disaster stage for the first polar body relatively few times in the rat. Other

ovarian ova in my series, also, show chromosomes arranged in a monaster for the formation of the first polar body; others show the monaster for the second polar body while the first polar body is still present.

The monaster for the first polar body division is easily distinguished from the monaster for the second polar body division. Sobotta and Burkhard (1910) have computed measurements for the two spindles and found that the first is shorter and broader than the second. The length of the second spindle was found to be 27 microns and only 15 microns for the first. The second spindle lies more superficial in the ovum as compared with the first spindle.

Many ovaries in my material present disintegrating ova. These ova appear broken up into numerous small spherical masses, some of which have what appears to be a nucleus with prominent chromotoid bodies.

4. EXTRA-TUBAL OVA

(Plates 1 and 2, Figures 1, 2, 3, 4)

Two extra-tubal ova were found near the ovary and within the periovarian capsule in sections of the right ovary and oviduct of a rat which had been inseminated 12 hours. One ovum is definitely

in a stage of disintegration while the other appears normal. The disintegrating ovum (Plate 2, Fig. 3 and 4), has a large ovoid nucleus with chromatin clumped into chromotoid bodies. The cytoplasm suggest vacuolation. There are no discus cells or coagulated mucous substance around the ovum. The ovum is elongated and at one extremity appears a globule like body, apparently segmented from the ovum, suggesting that the cytoplasm is breaking up, as seen frequently in disintegrating ovarian and tubal ova. The vertical diameter of the ovum including the segmented portion, is 72 microns and the horizontal diameter is 28 microns. The ovum has no oolemma. The vitelline membrane is not pycnotic. The normal ovum (Plate 1, Fig. 1 and 2) found on the other side of the same ovary apparently is in the monaster stage for the second polar body formation. The first polar body apparently is not present. The spindle was sectioned transversely giving a polar view. All the chromosomes appear in one section of seven microns in thickness, indicating the chromosomes are all in about the same plane. Beyond the plane of section of the chromosomes, what appears to be cross sections of linin threads in the spindle can be seen. This mitotic figure is close to the periphery of the ovum, and is

oriented tangenitally. The length of this spindle is placed roughly at 25 microns. This ovum is surrounded by many discus cells. Those nearest the ovum are arranged in two or three rows like those enclosing the ovarian ova. These discus cells show no mitotic figures as do those in the ovary, suggesting that the formation of new discus cells has ceased. The cytoplasm of many show vacuoles and beginning fragmentation. The cells towards the periphery of this mass of discus cells, show more pronounced disintegration as compared with those nearer the ovum. Spaces exist between the more peripheral cells, which contain a fibrillar-like substance, suggesting coagulated mucous. The zona pellucida is much reduced and in some places the discus cells are in contact with the ovum. This ovum appears slightly pycnotic. This condition is seen in some ovarian and tubal ova and may suggest beginning disintegration. However, the cytoplasm appears normal in all these ova. The diameter of the ovum is 66 microns by 70 microns. A tubal ovum from the same specimen measures 62.9 microns by 59.2 microns in diameter. An

apparently mature ovarian ovum from the same ovary measures 74.5 microns by 62.9 microns in diameter. Sobotta and Burkhard (1910) give as the diameter of ovarian ova 60-65 microns and tubal ova 55-60 microns. Diameters of ova measured by Huber (1915) are a little more than those measured by Sobotta and Burkhard, possibly due to differences in fixation technique. These investigators show that tubal ova are smaller than ovarian ova. Diameters of ovarian and tubal ova in my series show this, also, but my measurements run a little higher.

Three ova, the chromosomes in the second monaster, were found near an opening in the third or fourth division of the right oviduct of a rat which had been inseminated 27 hours. This opening in the oviduct may be due to rupture of the tube during manipulation incident to the removal of the periovarian capsule. What appears to be similar openings were found in sections of tubes of three other rats of my series in which the periovarian capsule had unintentionally been removed. It is possible these ova were squeezed out through this

opening in the oviduct. In such a case they could not, of course, be considered as truly extra-tubal ova. Large clumps of discus cells are near the ova. The ova are surrounded by scattered discus cells, leucocytes and coagulated mucous. The ovary does not show any recently ruptured follicles. One follicle is much distended and appears about ready to rupture. It contains an ovum enclosed in a small mass of follicle cells attached to the tunica near the point where apparently the follicle is about to rupture, as suggested by the thinness of the wall.

Ova in the process of escaping from the ovary have been observed by Sobotta (1895) in the mouse, Longley (1911) in the cat, and O. Van der Strict (1909) in the bat. The ovum described by Longley in the cat, which he observed at first macroscopically, shows a mass of coagulum around the ovum, which he considers to be coagulated follicular fluid. Upon microscopic examination the ovum has the first polar body and the chromosomes arranged eccentrically in a monaster. Many follicle cells enclose the ovum.

5. TUBAL OVA

Development of tubal ova in my series range from the second monaster to and including the eight cell stage. The age and development of these ova correspond closely to those described by Huber (1915) and Sobotta and Burkhard (1910).

A. 10 Hours

The earliest ova in the series is that from a rat 10 hours after completed insemination. The oviduct of the right side was damaged in sectioning and could not be used. Many follicles in both left and right ovaries contain degenerating ova. These follicles appear about ready to rupture. Other follicles have normal ova which are about ready to ovulate. Other follicles show evidence of having recently ruptured.

The oviduct of the left side in the 10 hour specimen contains four ova in the ampullar portion. All are situated in one mass of discus cells. They are in the second monaster and none have yet been fertilized. There are no polar bodies. There are no sperm near the ova, but there are a few in the lower part of the oviduct.

The uterine horn contains a great amount of sperm between the folds of the mucosa. Very few sperm are seen in the lower portion of the oviduct but they increase in number towards the distal end of the oviduct. The oviduct opens into the side of the uterine horn by a small opening. The lower part of the oviduct runs in the muscular and mucosal layers of the uterine horn, enters the lumen from the side and protrudes down into the lumen of the horn. By comparison of the number of sperm around that part of the oviduct which protrudes into the uterine horn, and the number which have gained entrance to the lower part of the oviduct, it appears that this anatomical arrangement may act as a barrier to the number of sperm which enter the oviduct. By forceful injection of india ink into the uterine horn of two different specimens, the amount of ink that entered the oviduct in one case was small and in the other it was impossible to force ink past this portion of the horn. This arrangement acts as a valve against the entrance of the ink into the oviduct. According to Sobotta and Burkhard

(1910) the number of sperm that enters the oviduct is relatively small and the life of sperm in the genital tract of the rat is between 10 and 14 hours. They showed that at 10 hours most of the sperm were dead and at 14 hours all were dead.

An orderly arrangement of discus cells around the ova in the 10 hour rat is still apparent, although their cytoplasm shows more disintegration than that of the discus cells enclosing the extratubal ova. Many cells resembling mononuclear leucocytes are present, especially around the periphery of the mass of discus cells. Intermingled with the cells are many fibrils, suggesting as is noted above, a coagulated mucous. The ova are slightly pycnotic but show no internal evidences of disintegration.

The epithelium of the ampullar portion of the oviduct from this 10 hour specimen, shows evidences of secretion. A mucous-like substance appears in the lumen of the tube in close apposition to the epithelium. Secretory and ciliated cells were observed and their characteristics correspond to those described by Schaffer

(Monatssch. f. Geb. u. Gyn., 28(on the pregnant squirrel. Schaffer's plates illustrating the eipthelium of the oviduct of a pregnant squirrel, show ciliated cells with a pale homogeneous cytoplasm. The cilia are quite apparent ending at the "basalknotchensaum" or basal granulation. The secretory cells do not show a highly granular cytoplasm towards the lumen as is seen in the cells of the eipthelium from the oviduct in the non-pregnant squirrel, but secretory material is seen escaping into the lumen of the tube. The cellular arrangement, according to Schaffer, appears to be an alternate one in general, a secretory cell then a ciliated cell. According to Sobotta and Burkhard (1910), this mucous-like substance secreted by the cells of the oviduct may play an important part in directing the ova into the oviduct.

B. 12 Hours

The rat which had been inseminated 12 hours has in the ampullar portion of the right oviduct five ova, besides the two ova which have not yet

entered the tube. These two latter ova are described above under extra-tubal ova. Of those ova in the tube all but two have been fertilized and these two are in the second monaster. Two ova possess an anaphase spindle in process of formation of the second polar body. Both have been penetrated by sperm, one showing the spermium head entering the ovum. One possesses a spermium but its chromosomes are still in the second monaster. A few sperm are seen in the folds of the mucosa of the oviduct near the ampulla.

In the tubes on the left side of the same rat, no sperm were observed in the upper or middle segments. The ampulla contains seven ova. Only one has been fertilized and this one shows the formation of the second polar body. The rest of the ova are in the second monaster. All the tubal ova from both the right and left tubes are contained in one mass of discus cells. These cells appear similar and in about the same stage of disintegration as those observed in the 10 hour specimen. The ovary of the right side has recently ruptured follicles. One follicle in particular

has a small opening through the epithelium and a few follicle cells between the periovarian capsule and the ovary. Two other follicles have no definite opening but the epithelium has not yet regenerated over the plug of follicle cells. The ovary of the left side has recently ruptured follicles but none as recent as are found in the ovary of the right side, suggesting that the two extratubal ova had just recently been ovulated. The uterine horns of both sides are packed with sperm with relatively few in the oviducts.

C. 16 and 24 Hours.

Four ova in the pronuclear stage were observed in the right oviduct of the rat which had been inseminated 16 hours. Tubes of the left side were not sectioned. Three of the ova show the second polar body extruded. The pronuclei are quite near each other, towards the center of the ovum. The pronuclei contain a pale staining nucleoplasm with a few scattered granules. One nucleus is smaller than the other and according to Huber (1915), "this one is regarded as the male pronucleus,

since near it the 'sperm centrum' was now and then observed. Hill and Tribe (1924), in their work on the early development of the cat, believe that the smaller pronucleus is the female pronucleus and the larger the male pronucleus. In describing one of the eggs of their series they say, "of the two, one is smaller and situated nearer the polar bodies; we regard it as the female pronucleus; the other is larger and rather more superficially situated. This is in agreement with the conclusions of Lams (1913) for the pronuclei in Cavia, and of O. van der Stricht (1909) for those of Vesperugo". The pronuclear stages described by Huber (1915) were taken from rats which had been inseminated 24 hours. According to Huber, the pronuclear stages in his series seem to fall in about the middle of the series of the pronuclear stages described by Sobotta and Burkhard (1910). Ova in my series taken from the right oviduct of a rat inseminated 24 hours all show the pronuclear stage. This tube contains eight ova; one of these appears to be disintegrating. Two of the ova possess the second polar

body within the oolemma. The pronuclei in all these ova contain large dark staining chromotoid nucleolar bodies arranged on a linin network. The pronuclei from ova of the 16 hour rat show a pale-blue staining nucleoplasm with a few small darkly-staining granules in contrast to the characteristics above described of the 24 hour pronuclear stages. There are fewer discus cells around these ova than in the earlier specimens. Five ova are contained within the left oviduct. These ova are all in the pronuclear stage and similar to those of the right side. Therefore, it seems the pronuclear stage is of considerable duration. Ova from my 30 hour specimen show the two-cell stage and two ova in my 27 hour specimen show what is thought to be an early and late anaphase stage for the first segmentation division, to be described below with ova from the 27 hour specimen. From my data it would appear the pronuclear stage extends over a period of about 12 hours, considering fertilization takes place about 12 hours after copulation.

These ova in the 24 hour pronuclear stage have migrated past the ampullar portion of the oviduct.

Huber (1915) in his reconstructions of the oviduct, illustrating the position of the ova of this age, showed that these ova had migrated about eight millimeters from the fimbriated end or at the end of the first day had travelled about one-fourth the length of the oviduct. Huber describes the oviduct of the rat as containing eight or ten major folds; the middle group of folds is closely applied to the ovarian capsule, the upper or distal folds pierce the capsule, ending in the fimbriated end found within the capsule, while the lower or proximal folds, proximal with reference to the uterine horn, effect connection with the uterine horn. Sobotta (1895) in his description of the oviduct of the mouse recognizes four segments, characterized by epithelial lining, nature and extent of folding of the mucosa, and thickness of the musculature. Huber classifies the oviduct of the rat in respect to that of the mouse in that he puts the four distal segments beginning with the fimbriated end, with segments one and two of Sobotta's designation, which have a wide lumen and folded mucosa. In the third and fourth segments of the mouse oviduct the musculature is well developed, epithelium is simple

columnar and folds nearly absent. These correspond generally with the remaining folds or segments of the rat oviduct.

D. 27 Hours.

The oviducts from the rat inseminated 27 hours, present four ova in the right and six in the left. The three ova found outside of the tube on the right are described above under extra-tubal ova.

Three ova in the right oviduct have their chromosomes arranged in the second monaster. The stage of the fourth ovum is uncertain as the mitotic figure is cut transversely. The chromosomes extend through three sections, each seven microns in thickness. Therefore, its length is roughly placed around 21 or 25 microns. It is apparently in an early anaphase stage, probably for the first segmentation division. This mitotic figure is more centrally placed in the ovum in contrast to eccentrically placed mitotic figures for the polar bodies.

The mitotic figures of two ova in the left oviduct (see Plate 3, Fig. 5 and 6) present characteristics of an early and late anaphase stage for the formation of the first segmentation division.

However, since these two ova are the only ones of this particular stage that I have, they will deserve comparison with other ova of this age to be left for further investigation. It is this spindle for the first segmentation division that neither Sobatta and Burkhard (1910), Kirkham and Burr (1913), or Melissinos (1907), were able to find in their work on the rat, although all of the maturation stages are described by Sobotta (1895) on the mouse. Because of the somewhat eccentric position of the mitotic figures, some doubt appeared whether these two ova in my specimens were preparing to segment or whether the spindles belong to the second polar body. However, these mitotic figures are not placed as near the periphery of the ovum as the radially placed spindle for the second polar body. The width of the spindles is 11.2 microns compared with 6.4 microns for a second monaster spindle of an ovum in the same tube and 7.4 microns for a first monaster spindle of an ovum in the ovary of the same rat. The length of each spindle is about 25.9 microns, which is about the same length of the monaster spindle for the

second polar body division.

Very few discus cells are present around the ova and leucocytes predominate. There is some coagulated mucous present. No sperm are present in the tubes.

The first segmentation spindle has been observed in other animals. Sobotta (1895(describes this particular stage in the mouse ovum and Hill and Tribe (1924) describe an ovum of the cat which is incompletely divided into two halves by a deep circular groove extending in more than half-way towards the center. Each half contains a small nucleus. In the center, aboud mid-way between the two nuclei, remains of the spindle-fibers are distinctly visible.

E. 59 Hours.

Four ova in the two-cell stage were found in the right oviduct of the rat inseminated 30 hours. All possess a resting nucleus. Two have a polar body within the oolemma. One two-cell ovum possesses a spermium in one of the cells. The spermium is near the periphery of the cell and has not undergone development. Two of the two-cell

stages have a polar body within the oolemma. There are about the same number of discus cells around these ova as are found near the 27 hour ova. Leucocytes predominate. There is a large clump of discus cells showing some disintegration near one of the ova. These discus cells apparently have no relation with the ova. Very little coagulated mucous is evident. There are no sperm in the oviduct or uterine horn. The left oviduct was not sectioned.

F. 35 Hours.

Five ova in the two-cell stage exist in the right oviduct of the rat inseminated 35 hours. These ova are all in the same segment of the oviduct and near each other. There are no discus cells but a number of leucocytes are present. There is no coagulated mucous. The cells of the ova have a resting nucleus. The left oviduct was not sectioned.

G. 45 Hours.

From the right oviduct of the rat inseminated 45 hours five ova were found in the two-cell stage. All cells have a resting nucleus. No discus cells and only a few leucocytes are found near the ova.

A large clump of discus cells are present at some distance from the ova and show no apparent relation with the ova. A spermium was found in a cell of one of the two-cell ova, and shows no development. There is no coagulated mucous around the ova.

The two-cell stage with a resting nucleus extends through a relatively long period. Sobotta (1895) found in the mouse, two-cell stages present through a period ranging from 25 hours to 48 hours after copulation. Huber (1915) observed in the rat the two-cell stage during a period extending from 42 hours to 70 hours after the beginning of insemination.

In my work the two-cell stage is found as early as 30 hours after insemination. Many two-cell stages in my series show no oolemma. Huber (1915) says the oolemma was clearly observed in certain of his two-cell stages, but not in the four-cell nor eight-cell stages. Widakowich (1910), reports that he observed in the albino rat, a loss of the oolemma even in the two-cell stages, and Sobotta (1895), found that the oolemma disappears in the eight-cell stages of mice ova. Huber says that the early dis-

appearance of the oolemma may account for the fact that the egg mass during segmentation and transit through the oviduct does not, as a rule, present a spherical form but appears compressed and mouled to fit the form of the lumen.

H. 3 Day, 15 Hours.

An eight-cell stage was found in the lower segments of the right ov iduct of a rat which had been inseminated three days and 15 hours. Of five segmented ova in this tube only one shows definitely the eight-cell stage. All nuclei are in the resting stage. There are no discus cells, coagulated mucous, or leucocytes around any of the ova. No oolemma is present. This eight-cell ovum rests against a fold in the mucosa of the oviduct. Huber (1915) found in the oviduct, the eight-cell stage three days and 17 hours after insemination and the 12 and 16 cell stages at four days after insemination. At the beginning of the fifth day he found all segmented ova in the uterine horn. Therefore, he concludes from his work, the segmenting ova pass from the oviduct to the uterine horn at the end of the fourth day after the beginning of insemination, probably in the 12-cell to the 16-cell stages.

CONCLUSIONS

The albino rat is a particularly good mammal for the study of early development. The spontaneity of ovulation at intervals of five days or slightly less, during about the time the animal is manifesting signs of heat, makes the conditions favorable in securing developmental stages at certain ages, reckoning from the time of completed copulation.

Extra-tubal ova were observed in the 12-hour rat, suggesting that copulation took place quite early during the time the female was manifesting outward signs of heat. The normal extra-tubal ova are encircled with an abundance of discus or follicular cells.

The discus cells show a progressive disintegration during the developmental stages of the ova. Leucocytes predominate in the later stages of development of the ova, suggesting their function of assisting in the removal of discus cells. In later stages of the segmented ova the leucocytes also have disappeared. The process involved in the disappearance of the discus cells remains for future investigation.

Fertilization was observed 12 hours after

insemination. Sperm were observed in the oviduct of the 10-hour rat but no ova have as yet been fertilized. Fertilization occurs during the early part of the second half of the first day.

Pronuclear stages in my series were observed as early as 16 hours and as late as 24 hours after insemination.

What is thought to be an early stage of the first segmentation spindle was seen in three ova, 27 hours after insemination.

At 30 hours the two-cell ovum has developed, and still exists at 45 hours. Huber (1915) found the two-cell stage in the rat as late as 70 hours after insemination.

The eight-cell stage was found at three days and 15 hours, which corresponds with the eight-cell stage that Huber described, found three days and 17 hours after insemination. I observed no four-cell ova.

The epithelium of one of the oviducts observed, shows a secretory function and corresponds with the description given by Schaffer of the

epithelium from the oviduct of a pregnant squirrel. It is thought by Sobotta and Burkhard, that this mucous-like substance secreted by the cells of the epithelium in the fimbria of the oviduct, may exert an influence in directing the ova into the ampulla of the oviduct. Schaffer showed that secretory cells in the epithelium of the oviduct of a non-pregnant squirrel are secreting material towards the lumen but no secretory products are escaping, suggesting that the advent of ovulation and pregnancy causes a heightened activity of the secretory cells of the oviduct. The function of this mucous-like substance is as yet not definitely known.

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8. DESCRIPTION OF FIGURES

Plate 1.

Figures 1 and 2. An extra-tubal ovum from a rat inseminated 12 hours, surrounded by follicle or discus cells and coagulated follicular fluid. Disintegration of follicular cells is shown in figure 2. The mitotic figure is that of a monaster, preparatory for the second polar body division. Z.p.-Zona pellucida. P.c.-Periovarian capsule. (Fig. 1, Mag. about X93, Fig. 2, Mag. about X440).

Plate 2.

Figures 3 and 4. A disintegrating extra-tubal ovum from a rat inseminated 12 hours, showing vacuolation of the cytoplasm. Part of the cytoplasm is shown segmented from the ovum proper. Pc.E.-Epithelium of periovarian capsule. O.E.-Ovarian epithelium. (Fig. 3, Mag. about X114, Fig. 4, Mag. about X486).

Plate 3.

Figures 5 and 6. Two tubal ova from a rat inseminated 27 hours. The mitotic figures are thought to be spindles for the first segmentation division. L.-Mononuclear leucocytes within coagulated mucous substance. D.C.-A large clump of discus cells. Z.p.-Zona pellucida. (Fig. 5, Mag. about X120, Fig 6, Mag about X528).

Plate I

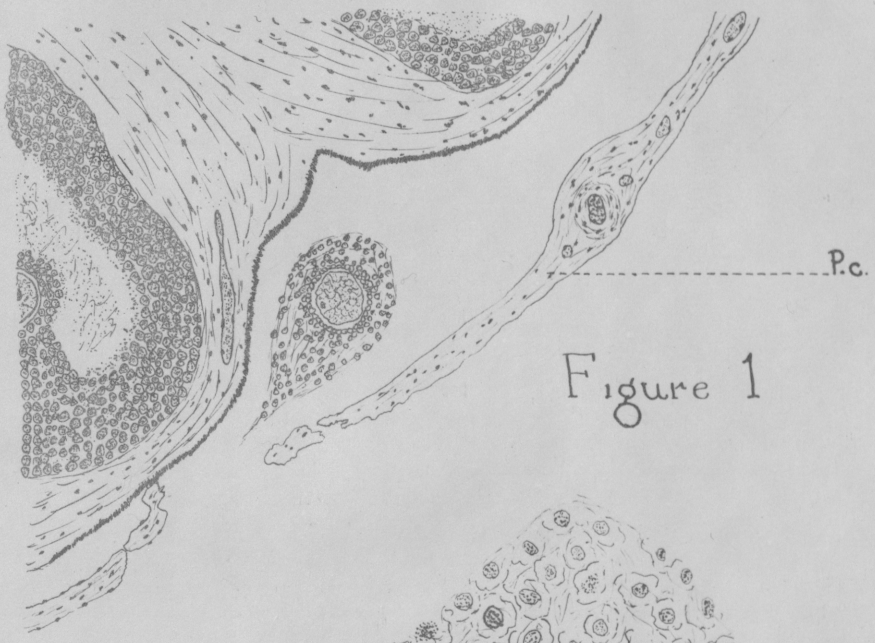

Figure 1

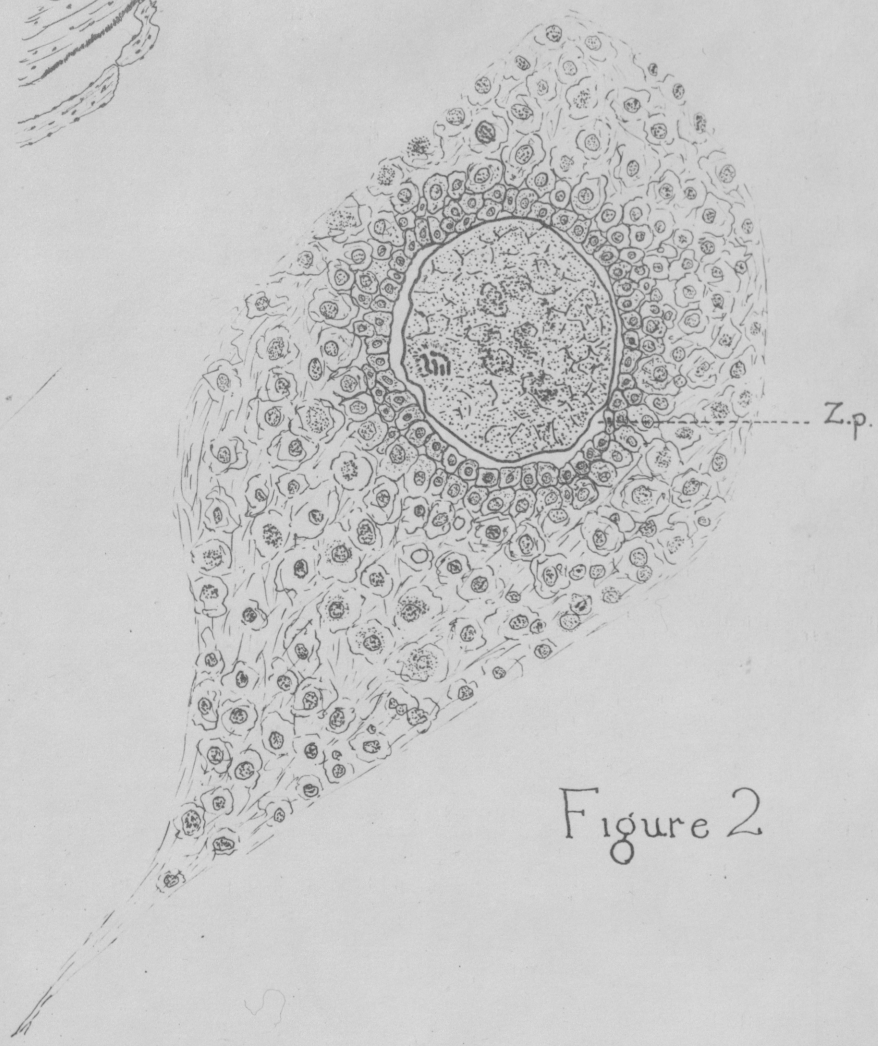

Figure 2

Plate II

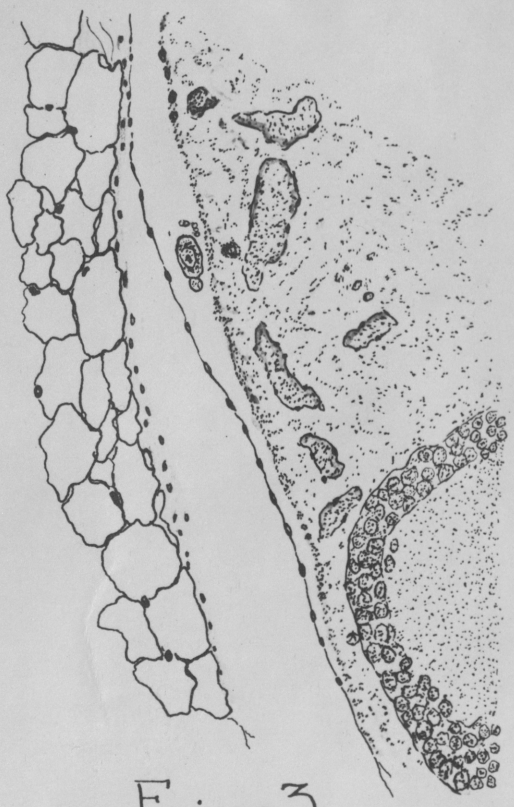

Figure 3

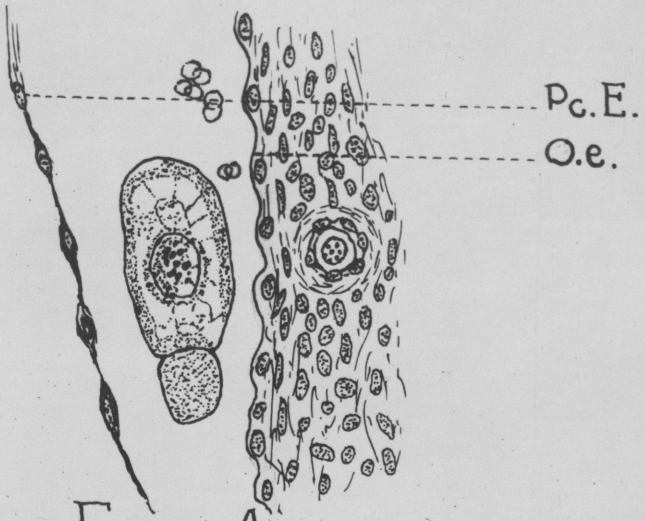

Figure 4

Plate III

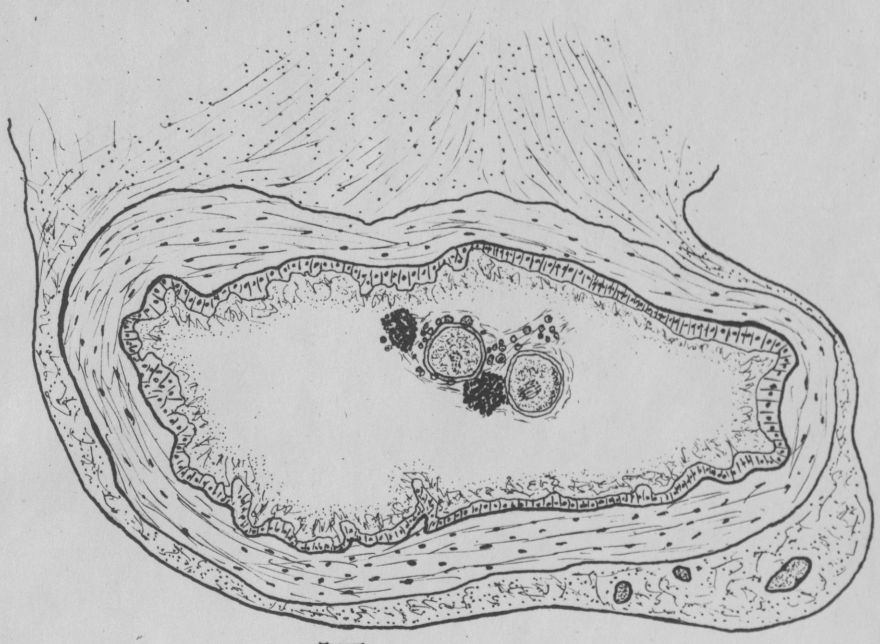

Figure 5

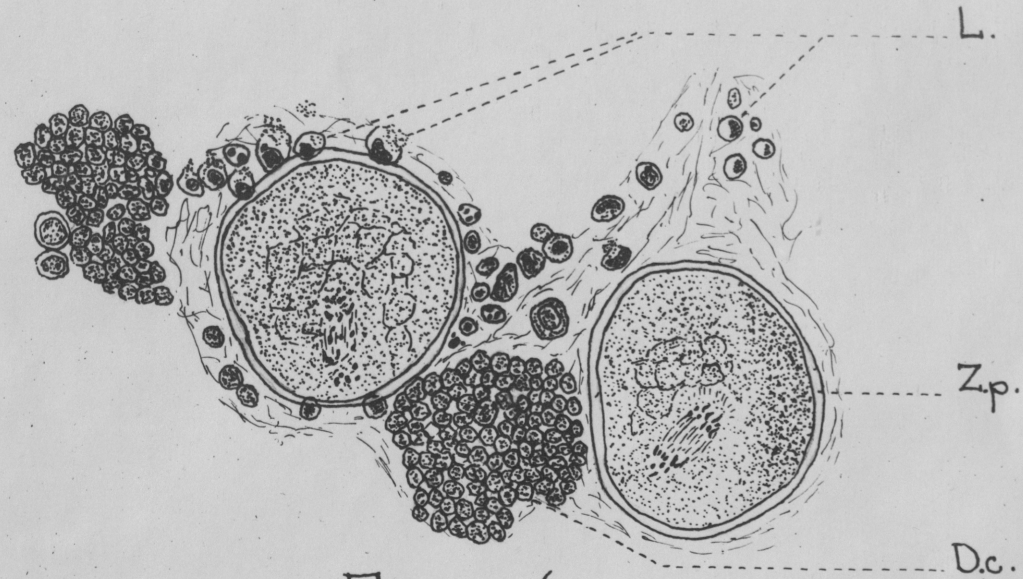

Figure 6