sugar is tentatively explained as follows: the utilization of glucose by the rapidly multiplying trypanosomes exercises a progressively increasing demand upon the glycogenolytic function of the liver. The release of glycogen by the liver is probably facilitated under the existing condition of reduced alkali reserve. This may account for the occasional temporary hyperglycemia noted and for the increase in muscle glycogen (Bruynoghe, Dubois, and Bouckaert[3]). It seems probable that the central necrosis of the liver, caused by the anoxemia, interferes with the glycogenic function of that organ, so that it ultimately fails to supply sufficient glycogen to maintain the normal level of glycemia.

4991

The Passage of the Spermatozoa and Ova Through the Oviducts of the Rabbit.*

G. H. PARKER.

From the Zoological Laboratory, Harvard University.

In earlier papers[1, 2, 3] I pointed out that the oviducts of the turtle and of the pigeon possess narrow pro-ovarian tracts of cilia by which spermatozoa may be carried from the lower part of these ducts to the neighborhood of the ovary. It was a matter of interest to ascertain whether such tracts obtain in the mammals. For this purpose I have studied the oviducts of the rabbit.

In the female rabbit the vagina is a single median tube about 5 cm. long. This is followed by two independent uteri each about 7 cm. long, opening separately into the vagina. From each uterus a narrow Fallopian tube approximately 7 cm. long is continued forward to its termination in an infundibulum close to the ovary of its own side. After coition spermatozoa reach the anterior end of the uterus in about 2 hours and are found at the infundibulum in 2 hours more. Ovulation takes place about 10 hours after coition. Spermatozoa will live in the female ducts of the rabbit for about 30 hours, the eggs

[3] Bruynoghe, M., Dubois and Bouckaert, *Bull. L'Acad. Roy. de Med. de Bel.*, 1927, v, 7, 142.

* The expense of these investigations was borne in part by funds received from the National Research Council, Committee for Research Problems of Sex.

[1] Parker, G. H., PROC. SOC. EXP. BIOL. AND MED., 1928, xxvi, 52.

[2] Parker, G. H.,, *Am. J. Physiol.*, 1926, lxxxvii, 93.

[3] Parker, G. H., PROC. SOC. EXP. BIOL. AND MED., 1930, xxvii, 704.

after liberation from the ovary, for from 2 to 4 hours. It therefore follows that spermatozoa which can live about 30 hours take in all 4 hours to reach the spot where 6 hours later eggs that can live from 2 to 4 hours will arrive. The questions that I wish to consider are how these elements, male and female, transferred through the rabbit's ducts.

The spermatozoa are deposited by the male rabbit in the vagina of the female and if the female is killed and examined as quickly after coition as possible, living spermatozoa will be found in the lower part of the uterus. The period between coition and examination is so brief, 3 or 4 minutes, that it is impossible that the spermatozoa through their own activity could reach so high a position in the oviducts as the lower part of the uterus. It is, therefore, probable that these cells are transferred from the vagina to the uterus by some activity of the female. This region of the uterus is ciliated and the cilia beat toward the exterior. They, therefore, do not afford an easy means of transfer for the sperm cells. If artificial insemination is performed on a female rabbit slightly under the influence of ether and so guarded as not to suffer an orgasm, the spermatozoa will remain for over an hour in the vagina without making their way into the uterus. In consequence of these conditions it is concluded that the transfer of the relatively voluminous sperm from the vagina to the uterus takes place through a muscular response of the oviduct in the nature of an antiperistalsis and probably associated with coition.

The ascent of the spermatozoa through the uterus has been attributed in part to the muscular movements of this organ and in part to its scanty ciliary supply. If in a quiescent uterus, freshly removed from a rabbit and kept in Ringer's solution at body temperature, living spermatozoa are injected into the distal end of this organ, they will be found to have arrived at its proximal end in about one hour. Similar injections at the proximal end are followed by spermatozoa at the distal end in about the same time. As such uteri were quiescent it is concluded that the spermatozoa make their way through these ducts by their own powers of locomotion. The spermatozoa are known to swim forward at an approximate rate of 60 μ's per second. They might, therefore, cover the length of the uterus, 7 cm., in 20 minutes. Hence one hour is not an unreasonable time for this migration. The conclusion that the spermatozoa make their way through the uterus by their own powers of locomotion is in harmony with the observation of Lim and Chao[4]

[4] Lim, R. K. S., and Chao, C., PROC. SOC. EXP. BIOL. AND MED., 1926, xxiii, 668.

that the operational inversion of a section of the uterine duct does not prevent pregnancy.

A most thorough examination of the Fallopian tubes in the rabbit has failed to disclose any other form of ciliary action than that of the abovarian type. No tract of pro-ovarian cilia was anywhere discoverable and the tubes in this respect are unlike those in the turtle and in the pigeon. The cavity of the Fallopian tubes in the rabbit is much divided by folds, as in most mammals, and the cilia covering these folds appear in all cases to have an effective stroke away from the ovary. It has been customary to assume that the spermatozoa pass through these tubes by swimming against the ciliary current, thus exhibiting a true rheotaxis.

When a freshly prepared tube is cut open so that its ciliary surfaces can be inspected under the microscope and it is flooded with a mixture of Ringer's solution and spermatozoa, the movements of the sperm cells in relation to the cilia can be easily observed. Many spermatozoa attach themselves by their heads to the surface of the ciliated folds. Their tails trail out from the point of attachment in the direction of the ciliary current. They are, therefore, oriented with their heads up the tube. Should they swim further they would, at least in the beginning of their course, ascend the tube. Such preparations have been watched for a long time but in no instance have spermatozoa been seen to free themselves and swim against the current. In the current next the cilia spermatozoa, swimming indiscriminately in all directions, are carried down the tube. Still further away from the cilia it is usual to find a counter current in which spermatozoa, again swimming in all directions, are being carried up the tube. The cavity of the tube is so generally divided by the folds already mentioned that counter currents are as common as downward currents. This interplay of downward currents and counter currents in the tube results in the transfer of small bodies such as spermatozoa both up and down the tubes. If India ink mixed with Ringer's solution is injected in small amount into the middle of the length of a Fallopian tube, it will sooner or later find its way out of this tube both into the uterus and into the region of the ovary. When an especially thin-walled portion of the Fallopian tube without having been cut open is inspected under the microscope, currents and counter currents can be easily seen in its interior. It is, therefore, concluded that overlapping counter currents are responsible for the transfer of the spermatozoa toward the ovary. This does not imply that the spermatozoa may not move upward under their own locomotion, but it is probable that this method of ascent is much less important than that of the overlapping counter currents. These currents also offer an explanation for the transfer to the region of the ovary of other small bodies such as fragments of epithelium from the uterus, which may thus gain an appropriate position to produce the pathological state of chocolate cysts.

The eggs of the rabbit are discharged from its ovary about 10 hours after copulation. The extrusion of the egg from the follicle is a relatively slow process. While this is going on the infundibulum is, according to Westman,[5] closely approximated to the ovary and the newly liberated eggs are carried into the Fallopian tube in large part by peristaltic suction. As is well known, the eggs pass rather rapidly through the upper part of the tube but much more slowly through the lower part. Much difference of opinion exists on the question of the ciliary and the muscular efficiency of the tube in this transportation. That the Fallopian tube exhibits peristalsis is well established. That in most mammals the tube is lined with cilia that beat toward the uterus is also well known. In certain forms, as, for instance, the mouse, the cilia are partly deficient and in regions where there are no cilia the action of the tube must be purely peristaltic. If the egg of the rabbit is measured it will be found to have a maximum diameter of about 0.16 mm. Its size is such that it cannot pass down the spaces in the Fallopian tube without pressing rather firmly against the faces of the folds in the tube. Since these faces are ciliated it follows that the pressure of the egg against them must bring the cilia into most effective play for the downward passage of the egg. It, therefore, appears probable that in the rabbit the passage of the egg through the tube is dependent in part upon ciliary action and in part upon peristalsis. This conclusion agrees with the observations of Kue and Lim[6] to the effect that when a section of the tube in the pig is reversed sterility follows.

The eggs of the rabbit develop into embryos in the uterus. Observations on gravid females made by the window method show that the discharge of these embryos is accomplished by peristalsis in which the 2 uteri act more or less independently.

Conclusions: 1. Spermatozoa are transferred from the vagina of the rabbit to its uterus by the muscular activity of the duct; 2, spermatozoa pass up the uterus by their own locomotion; 3, they ascend the Fallopian tube possibly by their own locomotion but chiefly through transportation by counter currents induced by the

[5] Westman, A., *Skan. Arch. Physiol.*, 1926, xlix, 243.

[6] Kue, Y. P., and Lim, R. K. S., *Chinese J. Physiol.*, 1928, ii, 389.

cilia; 4, the egg, in consequence of its size, so presses upon the ciliary surfaces of the tube that the cilia become effective in the downward passage of this body and thus supplement peristalsis.

4992

A General Test for Carbohydrates.

VICTOR E. LEVINE.
(With the technical assistance of Stanley T. Kucera.)
From the Department of Biological Chemistry and Nutrition, School of Medicine, Creighton University, Omaha, Nebraska.

A number of tests for carbohydrates depend upon interaction in the chromogenic system, phenol-aldehyde-acid. The Molisch, the Seliwanoff, the Bial, and the Tollen tests belong to this reaction system. The Molisch reagent employes the phenol, α-naphthol, the Seliwanoff reagent resorcin, the Bial reagent orcinol, and the Tollen reagent phloroglucin. The aldehyde in the reaction system is either the carbohydrate itself or the furfural obtained by the decomposition of carbohydrate by means of more or less concentrated acid. Either the reagent or the final reacting mixture contains hydrochloric or sulphuric acid. The aldehyde and the phenol react to form a compound of chromogenic power. The Seliwanoff, the Bial and the Tollen reagent serve to detect certain types of carbohydrates, while the Molisch reagent is universal in its ability to detect carbohydrates of any number of carbon atoms, aldehydic or ketonic in nature, with or without an open carbonyl group, free or in combination with other compounds.

We have developed a test for carbohydrates, which like the Molisch is of general application, by introducing thymol as the phenol in the system, phenol-aldehyde-acid. We have found that a 5% thymol in 95% alcohol serves as a useful, sensitive and practical reagent for carbohydrates. The thymol reagent has the advantage over the Molisch reagent, since the former is a colorless solution and does not deteriorate on long standing. We have kept the alcoholic thymol solutions in ordinary bottles exposed to window light for over a year without the development of any color.

To make the test we proceed as follows:* 3 or 4 drops of the 5%

* Molisch described a test for sugar in the urine based upon a reagent containing 15% thymol in alcohol. At this concentration 3 to 4 drops of the solution alone will yield a colored ring with concentrated sulphuric acid. The thymol used in our experiments was obtained from Merck and was of reagent quality.