TABLE I. Numerical Data, Averages and Ranges (in Parentheses) for Groups.

	Sham operated, fed by stomach tube	Hypophysectomized, fed by stomach tube	Sham operated, fed *ad lib.*	Hypophysectomized, fed *ad lib.*
No. of animals	7	5	7	9
Body wt, g				
Day of operation	270(258–283)	272(262–287)	268(261–282)	269(260–285)
Beginning of fast	297(282–314)	287(280–293)	332(298–353)	251(237–266)
16th day of fast	209(193–215)	190(182–197)	223(199–234)	174(161–187)
Day animal died	155(148–172)	146(138–153)	156(150–174)	152(135–170)
Blood sugar, mg %				
Beginning of fast	108 (94–116)	124(101–147)	117(101–134)	107 (93–114)
2nd day " "	78 (72–84)	87 (78–96)	79 (71–95)	73 (62–84)
16th " " "	78 (65–102)	65 (46–72)	76 (64–98)	66 (54–84)
Survival after beginning of fast in days	24 (19–28)	29 (27–31)	28 (23–31)	23 (18–29)
Organ wt at death, g				
Testes	1.96(1.76–2.20)	.35(.28– .42)	1.93(1.60–2.38)	.37(.29– .44)
Liver	2.1 (1.8 –2.3)	3.3 (3 –3.6)	2.2 (1.9 –2.6)	3.6 (3 –5)

during fasting which were comparable to those of their controls.

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Spermicidal Effect of Certain Physiologically Active Substances. (19632)

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A great number of compounds have been tested to determine their action on spermatozoa. Among those which have been found to be toxic are alkyl hydrocupreidine derivatives(1), metabolites such as indole, maleate, and glyceraldehyde(2), and long chain compounds such as sulfo-dioctylsuccinate(3). Ketone reagents have also been reported to be toxic(4), as have known enzyme inhibitors like fluoride and iodoacetate(5). Heavy metal salts, like phenylmercuric acetate(6,7), wetting agents, such as Duponol(8), and quinones(9) have been found to have toxic properties.

It is apparent that a large number of compounds of widely varying chemical structure have in common the property of toxicity toward spermatozoa. For this reason the possibility is relatively high that a given compound may have some spermatocidal effect. The ever increasing number and variety of chemical compounds in clinical use suggested that it might be of value to determine whether such substances would have a deleterious effect on spermatozoa, since this might affect their utility as therapeutic agents. In addition it was felt that the possibility of discovering one or more spermatocidal agents in types of

compounds not hitherto extensively studied warranted the investigation reported here.

Experimental. Spermatocidal activity was determined by a modification of the method of Lardy and Phillips(10). Rat sperm were used. The rats were killed with ether and the testes removed immediately and cut in half. Testicular contents were removed with a platinum loop and flushed in 0.08 ml of Locke-Ringer solution on a glass slide. A cover glass was then placed over the solution and the slide was examined microscopically at 10-minute intervals. One testicle of each rat served as the control; the other testicle was treated with Locke-Ringer solution in which was dissolved the compound being tested. Under these conditions the control sperm were usually motile for a period of 30 minutes. Any shortening of this period was taken as indicating spermatocidal activity by the compound being tested. Compounds were run at serial dilutions. The highest concentration used was 1.0 mg/ml. Several series of compounds were tested. These included a group of wetting agents and a number of flavonoid compounds; the latter were run because they are known to form quinones in biological media(11), and quinones are known to have spermatocidal properties(9). Also investigated were a number of surface-active agents and a group of metabolite analogs. Particular attention was paid within this last group to analogs of pteroylglutamic acid, purines and pyrimidines, because of the high concentration of nucleic acids known to occur in spermatozoa. Finally a number of therapeutic agents were tested. An attempt was made in this group to cover as wide a range of therapy as possible, so that the group includes representatives of such pharmaceutical classes as antihistamines, antibiotics, etc. A number of standard spermicides were tested to provide a basis for comparison.

Results and discussion. Standard spermicides tested were lactic acid, oxyquinoline sulfate and phenylmercuric nitrate. Their minimum effective concentrations (M.E.C.) were, respectively, 1.0, 0.1 and 0.01 mg/cc.

Members of each of the other groups tested were found to show activity. In the flavonoid compounds, activity appeared to depend on ability for quinone formation. The most active compounds of the group were quercetin, dihydroquercetin, rufigallic acid and 3,3′,4,4′ tetrahydroxy chalcone, all of which had an M.E.C. of 0.1 mg/cc. A reduction in the number of possible quinone-forming groups reduced the activity; thus rutin and 3,4-dihydroxy chalcone had an M.E.C. of 1.0 mg/cc. Compounds which structurally cannot form quinones, such as hesperidin methyl chalcone and esculin, were inactive.

The results obtained with surface-active agents were not consistent. Although all the materials tested function in the same manner, some had a spermatocidal effect and some did not. A number of Spans and Tweens tested, for example, were inactive. Three samples of Triton had M.E.C.'s ranging from 1.0-0.1 mg/cc. This behavior would lead to the conclusion that spermicidal action by these agents is a function of their chemical structure rather than of their surface-active properties.

The activity of the metabolite analogs tested appears to be capable of rough classification. No appreciable effect was shown by analogs of phenylalanine, threonine, glycine, alanine or tyrosine. Analogs of thiamine, pantothenic acid and pyridoxine were likewise inactive. 2-Methoxy-1,4-naphthoquinone was active, having an M.E.C. of 0.1 mg/cc, but whether this is a manifestation of anti-vitamin properties is doubtful, since the quinone structure as such is known to have activity.

Pteroylglutamic acid analogs, however, showed spermatocidal activity as a group. 4-(((2-amino-4-hydroxy-6 pteridyl)-ethyl)-amino)-benzoic acid was particularly active among these compounds, giving an M.E.C. of 0.1 mg/cc. Activity at 1.0 mg/cc was shown by 12 other analogs tested. These represented structural modifications including replacement of the 6-pteridyl grouping with a 7-pteridyl, substitution of an aspartic acid grouping for the glutamic, inclusion of a methyl group on the pteridine nucleus, and replacement of the 2-amino group with a hydroxyl. The activity shown by these compounds appeared to depend on the pteridine nucleus, since replacement of this with a quinazoline grouping caused an inactivation. There is furthermore a correlation, although not a perfect one, be-

tween the anti-folic acid activity of these compounds and their spermatocidal effect(12). The possibility is thus raised that the spermatocidal effect of folic acid antagonists is due to a displacement of folic acid in the sperm cell. The importance of nucleic acids in the sperm cell and the function of folic acid in purine and pyrimidine synthesis lend more interest to this possibility. The purine and pyrimidine analogs tested were generally ineffective, although a few, particularly 2,4,5-triamino-6-hydroxy-pyrimidine, had appreciable activity. This had an M.E.C. of 0.1 mg/cc. Hitherto studies on the effect of inhibitors on spermatazoan metabolism have been concerned mainly with glycolytic mechanisms; the results discussed above suggest the utility of further study on purine and pyrimidine metabolism.

The group of therapeutic substances tested included representatives of the classes of diuretics, antihistamines, analgesics, and adrenolytics. The results obtained presented no coherent picture, and it would appear that spermatocidal activity in this group is a function of chemical structure rather than of therapeutic effect.

It is worthy of note that, among a number of compounds tested which do not fit into any of the above classifications, coumalic acid was particularly effective, having an M.E.C. of 0.01 mg/cc.

Summary. A number of compounds of different chemical structure and physiological effects were tested to determine their spermatocidal properties. Several were found to be active, including a group of pteroylglutamic acid analogs.

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Renotrophic-Androgenic Properties of Orally Administered Androgens.* (19633)

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Previous studies(1,2) have indicated that among the androgenic steroids the 17β-hydroxyl group is essential for maximum renotrophic activity and the 3-ketone group for androgenic activity in the castrated mouse. On the basis of these observations an attempt(3) was made to obtain further separation of these two properties of the androgens by studying compounds with a functional group in only one of these two positions. Such compounds administered parenterally, however, proved to be very insoluble in the tissue fluids. It seemed that another route of administration might prove more efficacious. Therefore, since one of these compounds, 17-methylandrostan-17β-ol, was still available, it was studied by oral administration and its properties compared with

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