consistently failed to elicit deciduomata in 70 ovariectomized mice not subjected to uterine traumatization.

The mouse is apparently capable of producing progesterone in amounts equivalent to 1.0 mg injected daily;[6] and the equivalent of the smaller injected amounts is assuredly produced normally by the ovaries of the mouse during pregnancy, pseudopregnancy, and lactation.[4, 6] Moreover, the long period after ovariectomy precludes any direct involvement of estrogen as such in these reactions, and presumably also rules out any preparatory action of this hormone.

Thus, without the recent or simultaneous action of estrogen, progesterone in physiologic amounts is capable of eliciting the development of placentomata in the suitably traumatized uterus of the mouse.

13012

Capsulation of *Neisseria gonorrhoeae.*

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We have succeeded in demonstrating and staining the capsule of *Neisseria gonorrhoeae* by a simple method. The presence of a capsule about the gonococcus has been reported only a few times and not substantiated, so that the Committee for Survey of Research on the Gonococcus and Gonococcal Infections[1] concluded, "The opinion of the majority is that the gonococcus does not form a capsule." Israeli[2] using a modified Hiss capsule stain found a clear or lightly stained zone, one to 3 times as wide as the organism; Szelvasi[3] claimed that in older cultures the capsule stained more intensively, although he did not describe the capsule. Recently Almaden[4] mordanted smears by Bailey's method, staining with carbolfuchsin, and stated that capsules could be found when the organisms were in the mucoid state.

In our work various technics have been tried, and finally Churchman and Emelianoff's[5] method, with modifications, was successful.

6 Pfeiffer, C. A., and Hooker, C. W., *Am. J. Physiol.*, 1940, **131**, 441.

1 Bayne-Jones, S., Chairman, *Am. J. Syph., Gon. and Ven. Dis.*, 1936, **20**, 5.

2 Israeli, C., *J. Am. Med. Assn.*, 1921, **76**, 1497.

3 Szelvasi, J., *Dermatolische Wschr.*, 1932, **94**, 204.

4 Almaden, P. J., *J. Infect. Dis.*, 1938, **62**, 36.

5 Churchman, J. H., and Emelianoff, N. V., *J. Exp. Med.*, 1933, **57**, 485.

Absolutely clean slides as used for flagella stains were employed; a drop of a slightly turbid aqueous suspension of the organism was placed at one end of the slide, the slide tilted and the drop allowed to run down and air dry. Then 10 drops of Wright's stain was placed on the slide and, when almost evaporated, washed off with tap water, distilled water, or buffered solution (Clark and Lubs, pH 6.4). To demonstrate capsules by this method a metallic sheen must surface the slide and provide a smooth pink-purple background and not a granular one.

The organisms used were 2 strains of gonococci isolated 2 months previously by Dr. R. D. Reid from clinical cases of gonorrhea. Dr. Reid kindly furnished 24-48-hour cultures grown on either dextrose starch agar or ascitic infusion agar; in the latter case the organisms were washed several times with physiological salt solution to free them of whatever protein material might have come from the medium. These cultures were rechecked for identity: they were Gram negative cocci seen most frequently in the typical biscuit shaped diplococcal form, fermented dextrose and not maltose, formed whitish-grey, translucent, raised colonies with smooth outline, one millimeter in diameter on chocolate-blood infusion agar, and these colonies gave a strong oxidase reaction with the paradimethylanalinemonohydrochloride reagent.

By the method described, preparations were obtained as shown in photomicrographs 1-5. The capsule is seen as a clear or very light pink halo, having a violet-pink outer membrane, and surrounds a blue stained soma, all against a smooth homogeneous background. The capsule may surround single organisms, diplococci, tetrads or groups of 8, the typical cleavage patterns as described by Neisser.[6] The cocci are .8 by 1 mu and the capsule (about diplococci) 2 by 3 mu.

In order to make certain that the clear zone was a true structural capsule and not an artifact, efforts were made to stain it more differentially; this was accomplished by flooding the dried smear with 5% acetic acid for 2 minutes, washing with water, allowing to dry and then staining as before. Photomicrograph 6 was taken of such a preparation and the capsule was seen as a red structure with a dark red outer membrane, surrounding the blue somatic body. Due to the chemical treatment the capsule is slightly smaller than that seen in photomicrograph 5, from the same suspension, but stained without preliminary fixation.

Moist India ink observations were also made to compare find-

[6] Neisser, A., *Deutsche Med. Wschr.*, 1882, **8**, 1852.

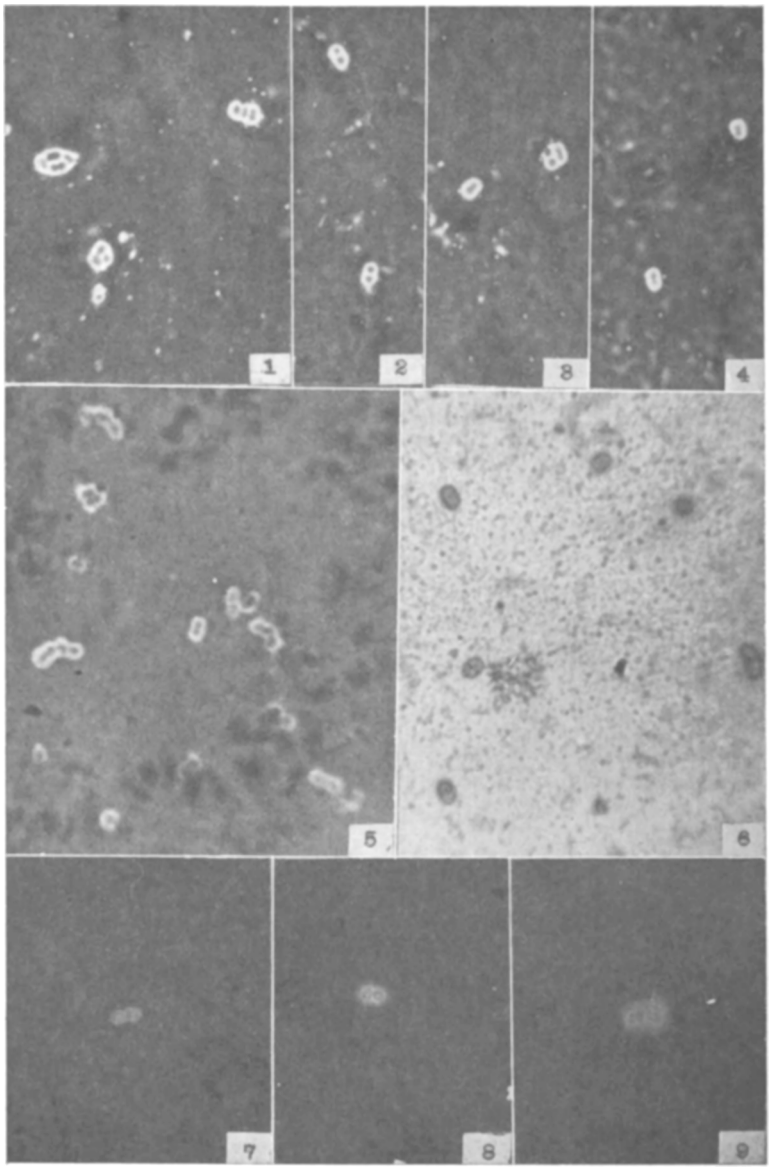

FIG. 1.

Photomicrographs 1000 ×.

1-5. *Neisseria gonorrhoeae*, Wright's stain.

6. *Neisseria gonorrhoeae*, fixed with acetic acid, then Wright's stain.

7-9. *Neisseria gonorrhoeae*, moist India ink preparations.

ings. The same appearances as in the stained slides were evident; photomicrographs 7-9 are of moist India ink preparations and show

the light, more refractile capsule about the darker somatic body of the gonococcus. The capsule possessed structural rigidity and appeared to have a definite elastic outer membrane, as the India ink particles were seen to cause indentations when they hit up against it. The ink particles then rebounded, as though caused to do so by the contraction again of the membrane.

13013

Effects of Increased Metabolism on Ketosis

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In animals in a state of ketosis exercise has been reported not to change significantly the rate of excretion of ketone bodies.[1, 2] Increased activity of isolated diabetic muscles causes marked increase in rate of disappearance of these substances.[3] Elsewhere we have suggested that these apparently contradictory findings can be reconciled if the liver increases its production of ketone bodies as a result of exercise.[4] To test this hypothesis, the ketone excretion of the human subject on a constant ketogenic diet was determined during several days. The urine was divided into day and night portions at 8 A.M. and 8 P.M. In several different experiments each lasting several days, the amount of muscular activity during the daytime period was increased on certain days by strenuous exercise (tennis) for at least 2 hours and on some days it was cut down to a minimum by rest in bed. The ordinary activity was light laboratory work. Regular hours for sleep were maintained during the night periods throughout. From Table I it is obvious that except for days of complete inactivity the nocturnal excretion of ketone bodies is definitely higher than the diurnal. There is also a tendency for the daytime output to be less, the greater the activity. At the same time the night periods which follow days of high activity tend to have more ketonuria than those which follow days

1 Barker, S. B., PROC. SOC. EXP. BIOL. AND MED., 1936, **34**, 893; *J. Physiol.*, 1939-40, **97**, 394.

2 Drury, D. R., *California and Western Medicine*, 1936, **45**, 1.

3 Blixenkrone-Moller, N., *Hoppe-Seyler's Z. f. Physiol. Chemie.*, 1938, **253**, 423.

4 Barnes, R. H., Drury, D. R., Greeley, P. O., and Wick, A. N., *Am. J. Physiol.*, 1940. **130**, 144.