

It will be noted that 155 failed to utilize citric acid and are therefore considered as of the genus *Escherichia*. The remaining 246 strains are tentatively considered as *Aerobacter* because they did utilize citric acid. Of these, 80 failed to attack glycerol and are considered *Aerobacter cloacae*. Of the remaining 166 cultures, 123 were typical for the *Aerobacter aerogenes* group as respects the citric acid, V.P., M.R. and glycerol reactions. The 43 remaining cultures differed from the latter in that the V.P. and M.R. reactions were non-correlating with growth on citric acid. These were practically the only strains which produced H<sub>2</sub>S, a characteristic which clearly differentiated them from all of the other groups under consideration. Furthermore, this group of 43 strains seems to be clearly differentiated from each of the other groups by a number of characters, as may be readily seen from the accompanying table. Generic or specific allocation is withheld pending determination of ability to produce trimethylene-glycol from glycerol.

Determinations of H<sub>2</sub>S were made with a number of different media and indicators, including iron (ferric chloride), nickel, lead, and manganese salts. Difco proteose peptone (2%) 0.1% K<sub>2</sub>HPO<sub>4</sub> in agar with iron citrate (0.05%) as the indicator was found to be very sensitive, definite tests for H<sub>2</sub>S, as indicated by blacking along the line of inoculation, being observed in as short a time as 12 hours, and very strong tests in 24 to 48 hours at 37°C.

## 6200

### Endocrine Reactions of X-Ray Sterilized Males.\*

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Investigations in the field of testicular physiology have borne out that the germinative and the endocrine systems are highly independent of each other, despite the obvious parallelism in their embryonic development. Cryptorchidism and X-ray sterilization result in many cases in complete degeneration of the germ cells, without affecting, however, the endocrine system, as evidenced by the normal development and persistence of the secondary sex characters

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\* This investigation was supported by grants from the Committee for Research in Problems of Sex and by the Committee on Effects of Radiation, National Research Council.

(seminal vesicle and prostate tests). On the other hand, Moore and Samuels<sup>1</sup> recently have shown that vitamin B deficiency inhibits selectively the endocrine system so that the male rats assume the castrate type while spermatogenesis proceeds normally.

The close time correlation in endocrine and spermatogenetic development is simply due to the fact that both are under the control of the hypophysis. The hypophysis stimulates the testicular development and answers with an increased hormone production if the testes are ablated. This reaction is most strikingly demonstrated in parabiosis experiments. Hill<sup>2</sup> has shown that the normalcy of the oestrus cycles of a rat female is undisturbed by a male parabiont. After castration of the male, however, the female falls into anoestrus for a period and then goes into permanent oestrus. These reactions are apparently due to the influx of excessive amounts of hypophysis hormone from the castrate, which initially cause luteinization of the ripe follicles and later the formation of cystic follicles.

One has generally been inclined to assume that the hyperactivity of the hypophysis is an answer to the deficiency or the complete absence of the endocrine component of the testis. It appeared of interest, therefore, to determine whether the hypophysis responds also to the absence of the germinal component only.

A series of 9 rat males, 2 months old, were sterilized, by the administration of 2400 r units of X-rays in 3 treatments. One month later they were united in parabiosis with normal females. Following this operation all the females exhibit irregularities in their oestral cycles; the oestrus periods are prolonged and eventually become permanent. The reaction differs only quantitatively from that observed in females united with totally castrated males. Hence it follows that X-ray sterilization causes a hyperactivity of the hypophysis. Pair 119 was selected for a thorough investigation. A colorimetric test proved that the blood exchange between the X-rayed male and the female was of a high average value. The testicles measured 11 x 6 x 6 mm., their weight and volume being about 25% that of normal controls. The epididymes and Cowper's glands were of normal size, the prostate and seminal vesicles distinctly enlarged. Cross sections through the testes give the usual picture of X-rayed testes with the relatively abundant interstitium and absence of germ cells.

There remains to consider whether the decrease in size of the

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<sup>1</sup> Moore, C. R., and Samuels, L. T., *Am. J. Physiol.*, 1931, **96**, 278.

<sup>2</sup> Hill, J. *Exp. Zool.*, 1932, in press.

testis might include a reduction of its endocrine component, which could possibly be the cause of the increased hypophyseal activity. Facts are not in favor of this interpretation. The interstitial cells appear normal in quality and quantity and the fact that the seminal vesicles and the prostates of our irradiated male are rather above the normal size clearly indicates that the endocrine system remained unimpaired by the amount of X-rays administered. We conclude, therefore, that the lack of spermatogenetic activity alone is responsible for the increase in hypophyseal activity in the X-ray sterilized male rat.

## 6201

### Biological Assay of Pregnandiol.

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Through the kindness of Dr. A. Butenandt of Göttingen who supplied us with 500 mg. of crystalline Pregnandiol<sup>1</sup> obtained from pregnancy urine, we were able to subject the substance to physiological tests. Pregnandiol, of which from 0.1 to 0.2 gm. can be obtained from 100 liters of human pregnancy urine, is, according to Butenandt, a saturated di-secondary alcohol of the formula  $C_{21}H_{34}(OH)_2$  containing 4 hydrated rings in the molecule.

The tests performed were as follows:

a. Injected (in oil) into castrated mice up to the dosage of 10 mg. it shows no female sex hormone effects (as already determined by Butenandt).<sup>1</sup>

b. Injected into immature rats up to the dose of 30 mg. it produces no ovarian changes characteristic of the prepituitary hormone.

c. Injected for 5 days into castrated rabbits primed for 5 days with female sex hormone, it produces no progestin (corpus luteum) effect on the uterus with a total dosage of 150 mg.

d. Injected into the virgin immature castrated guinea pig primed with female sex hormone up to a dosage of 100 mg., it fails to cause relaxation.

e. Injected into the castrate mouse brought to estrus by means of female sex hormone, it shows no mucifying effect on the vaginal epithelium.

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<sup>1</sup> Butenandt, A., *Berichte d. deut. chem. Gesellschaft*, 1930, **63**, 659; 1931, **64**, 2529. Marrian, G. F., *Biochem. J.*, 1929, **23**, 1090.