It would appear that in part the disturbance in resistance following suprarenalectomy may be due to the disturbance in electrolytes resulting from withdrawal of the cortical hormone. However, this is only one factor, for administration of salt to suprarenalectomized rats will raise the resistance slightly but not to a degree comparable to that obtained with injections of suprarenal cortical hormone. Furthermore, suprarenalectomized rats are killed by an amount of histamine (100 to 200 mg per kilo of body weight) approximately one-sixth to one-tenth the M.L.D. for normal rats.

It is of interest to compare these results with the abnormalities which develop in humans suffering from similar electrolyte disturbances, as occurs in Addison's disease, and in heat cramps.[5]

Summary. The production in normal rats of an electrolyte disturbance analogous to that observed after suprarenalectomy is followed by a marked drop in resistance to histamine.

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Adult Phosphatase Levels in Prepubertal Rhesus Prostate Tissue after Testosterone Propionate.

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An "acid" phosphatase with optimum activity at about pH 5 is present in *adult* human prostate gland and seminal fluid[1] in concentrations greater (500-2,000 units/g fresh prostate tissue, in our series[2, 3]) than the phosphatase activity of any other human tissue. *Prepubertal* prostate gland, on the other hand, contains less than 5 units of "acid" phosphatase activity per g fresh tissue.[3, 4] An intermediate value of 73 units was found in the prostate gland of a 13-year-old boy.[3]

This correlation in man between sexual maturity and the "acid" phosphatase activity of prostate tissue suggested the possibility that stimulation of the prepubertal prostate gland by injection of testo-

[5] Dill, D. B., *Life, Heat, and Altitude*, Harvard Press, 1938.

[1] Kutscher, W., and Wolbergs, H., *Z. f. physiol. Chem.*, 1935, **236**, 237.

[2] Gutman, E. B., Sproul, E. E., and Gutman, A. B., *Am. J. Cancer*, 1936, **28**, 485.

[3] Gutman, A. B., and Gutman, E. B., PROC. SOC. EXP. BIOL. AND MED., 1938, **39**, 529.

[4] Moore, R. A., and Hanzel, R. F., *Arch. Path.*, 1936, **22**, 41.

TABLE I.

Phosphatase Activity of Prostate Tissue of the Sexually Mature and Immature Rhesus Monkey; and of Immature Animals After Injection of Testosterone Propionate or of Estradiol Benzoate.

pH 4.9: M/200 monophenylphosphate substrate; M/10 citrate buffer; 37°C; 1 hr.
pH 9.0; M/200 " " M/20 Na veronal buffer; 37°C; 1 hr.

No.	Status	Wt, kg	Treatment	Wt both lobes prostate, g	Phosphatase Activity at pH 4.9 (units/g fresh prostate tissue)	Phosphatase Activity at pH 9.0 (units/g fresh prostate tissue)	Phosphatase Activity at pH 4.9 (units/whole prostate gland)	Phosphatase Activity at pH 9.0 (units/whole prostate gland)
1	Mature	10	Control	3.6	1,134	102	4,080	367
2	"	9.4	"	3.5	573	60	2,001	210
3	"	*6.0	"	3.2	356	22.5	1,140	72
4	Immature	3.5	"	0.7	1.2	1.2	0.8	0.8
5	"	3.5	"	0.6	2.8	—	1.5	—
6	"	4.0	"	0.3	4.8	0.4	1.4	0.1
7	"	4.0	Testosterone propionate (185 mg in 18 days)	2.0	975	30.5	1,950	61
8	"	2.9	Testosterone propionate (150 mg in 13 days)	1.1	779	14.8	858	16.3
9	"	2.1	Estradiol benzoate (6,000 rat units)	0.6	11.5	2.4	6.9	1.4

*Tuberculous animal.

sterone propionate would result in the precocious development of adult "acid" phosphatase levels. Rhesus monkeys were employed to investigate this possibility since, as indicated by Wolbergs,[5, 3] only in the monkey has the "acid" phosphatase activity of the prostate gland been found to be of the same order of magnitude as in man.

Methods. Experimental animals No. 7 and No. 8 (Table I) were injected daily (except Sunday) with 12.5 mg testosterone propionate, sacrificed after 18 and 13 days respectively; No. 9 received 500 Rat Units of estradiol benzoate in oil daily for 12 days.* Typical external evidences of the effects of these agents[6, 7] developed within the period of treatment. While the number of experimental animals is, of course, too small for definitive conclusions, the striking changes observed are thought to be significant.

Aqueous tissue extracts were prepared in the dilutions indicated elsewhere.[3] Tissue phosphatase activity at pH 9.0 was determined by the King and Armstrong method[8] and, in optimal dilutions, at pH 4.9 by the adaptation of that method previously outlined.[9] The results of tissue analyses are expressed in units, a unit being that degree of phosphatase activity which under the stated conditions of hydrolysis (Table I), will liberate one mg of phenol in one hour from the specified buffer—monophenylphosphate substrate solution. Serum phosphatase activity was determined by the Bodansky method.[10]

Results (Table I). In confirmation of Wolbergs,[5] at pH 4.9 we find marked phosphatase activity of prostate tissue of the *adult* Rhesus monkey, values of the same order of magnitude as in man. Unlike man, adult monkey prostate tissue exhibits appreciable phosphatase activity at pH 9.0, as in the rat.[3] In the *prepubertal* monkey, both "acid" and "alkaline" phosphatase activity of prostate tissue are negligible. Following treatment with testosterone propionate, the phosphatase activity of immature monkey prostate gland increases strikingly at pH 4.9 and at pH 9.0, reaching adult levels at the former and possibly also at the latter pH. Following treatment with estra-

[5] Quoted by Kutscher, W., and Pany, J., *Z. f. physiol. Chem.*, 1938, **255**, 169.

* We are indebted to Ciba Corporation for testosterone propionate (Perandren) and to Dr. Schwenk of Schering Corporation for estradiol benzoate (Progynon B). We are further indebted to Dr. E. T. Engle for guidance and for histological sections of the prostates of the experimental animals.

[6] Parkes, A. S., and Zuckerman, S., *Lancet*, 1935, **1**, 925.

[7] Zuckerman, S., and Parkes, A. S., *Lancet*, 1936, **1**, 242.

[8] King, E. J., and Armstrong, A. R., *Canad. M. A. J.*, 1934, **31**, 376.

[9] Gutman, A. B., and Gutman, E. B., *J. Clin. Invest.*, 1938, **17**, 473.

[10] Bodansky, A., *J. Biol. Chem.*, 1933, **101**, 93.

diol benzoate an equivocal increase in phosphatase activity was noted which cannot be interpreted as significant without further study.

The serum phosphatase activity at pH 9.0 was found to vary between 20 and 30 Bodansky units per 100 cc in immature Rhesus monkeys and was not significantly affected by injection of steroids; nor was the "acid" phosphatase activity of the serum,[11] which ranged between 2.5 and 3.6 units per 100 cc, affected thereby.

The phosphatase activity at pH 4.9 was found to be slight in the seminal vesicles (2.0-8.0 units/g fresh tissue), testis (4.0-5.6 units) and Cowper's glands (0.8 unit) of immature and mature Rhesus monkeys, with no significant difference in steroid-treated animals. The values obtained at pH 9.0 (0.6-3.9 units), (2.3-2.8 units) and (0.3 units) respectively, were also not different in the steroid-treated animals.

Discussion. The extraordinarily high "acid" phosphatase activity of prostate gland tissue in sexually-mature man and the monkey implies some as yet unknown prostatic function, effected by the enzyme. Within broad limits, the level of "acid" phosphatase activity of prostate tissue would appear to afford some measure of the capacity to exercise that function. In this sense, we interpret the effect of testosterone propionate in elevating the "acid" phosphatase activity of the prepubertal monkey prostate gland to adult levels as physiological evidence of a transformation in the direction of sexual maturity. This evidence is in harmony with the similar interpretation of concurrent morphological changes.[7] It will be noted (Table I) that estradiol benzoate, which acts chiefly upon the fibromuscular elements of the immature monkey prostate gland,[6] did not evoke a significant rise in "acid" phosphatase activity of prostate tissue; whereas a marked increase followed injection of testosterone propionate, which stimulates the development of the glandular epithelium.[7]

In our calculations (Table I), we conformed to common usage in regarding the prostate gland of the Rhesus monkey as composed of 2 lobes. The caudal lobe of the mature or stimulated gland, however, contains much more "acid" phosphatase than the cranial lobe: the ratio, *"acid" phosphatase activity per g caudal lobe/"acid" phosphatase activity per g cranial lobe* being 704/33, 2,270/124 and 1,357/86 in animals Nos. 3, 7, and 8, respectively.† In fact, such phosphatase

[11] Gutman, A. B., and Gutman, E. B., PROC. SOC. EXP. BIOL. AND MED., 1938, **38**, 470.

† The respective ratios at pH 9.0 were: 294/15.8, 53/10.3, and 22/6. In animal No. 9, however, the ratio at pH 4.9 was 9.6/19.9, at pH 9.0 2.9/2.5. Because of the small size of the cranial lobe in this animal, and the single observation, we are unable to say whether or not this deviation is significant.

activity as was found in the cranial lobe may well be due to reflux of caudal lobe secretion. The unequal enzyme distribution in the Rhesus gland suggests that the cranial lobe, if it is part of the true prostate gland, differs in function from the caudal lobe; as indicated also by differences in morphology and by van Wagenen's observations on the "coagulating" power of the cranial but not of the caudal lobe.[12]

While the function of the "acid" phosphatase of the mature prostate gland in man and in the monkey is not known, it would appear that suitable substrates are present in the seminal fluid (Ivanov,[13] and others) and that the slightly acid reaction of the vaginal secretion affords a favorable medium for its activity.

Summary. As in man, the prostate gland of the mature Rhesus monkey contains high concentrations of an "acid" phosphatase, whereas the prepubertal monkey prostate is virtually devoid of this enzyme. It is shown that testosterone propionate causes a several hundred-fold increase in "acid" phosphatase activity of the prepubertal monkey prostate gland to adult levels. It is inferred that in prepubertal man, so treated, an analogous increase occurs. In the monkey, "acid" prostate phosphatase is elaborated chiefly or solely in the caudal lobe.

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Combined Immuno and Chemotherapy of Pneumococcus Rat Infections.

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Fleming[1] has shown that type specific antipneumococcus serum increases the antibacterial action of sulfapyridine *in vitro* against pneumococci. McIntosh and Whitby[2] have shown, moreover, that sulfapyridine does not stimulate phagocytosis or antibody production against pneumococci, but suppresses their growth to a point that biological defense can become effective. Subsequently, we[3] showed that a small dose of broadly acting non-type-specific antipneumococ-

12 van Wagenen, G., *Anat. Rec.*, 1936, **66**, 411.

13 Ivanov, I. I., *Chem. Abstr.*, 1938, **32**, 2584.

1 Fleming, A., *Lancet*, 1939, **2**, 74.

2 McIntosh, J., and Whitby, L. E. H., *Lancet*, 1939, **1**, 431.

3 Powell, H. M., and Jamieson, W. A., *J. Immunology*, 1939, **36**, 459.