

measure of the quantity of an inhibitory substance given off by the cell suspension. This loss, however, does not depend on the temperature at which the suspension was hemolysed.

In the second experiment, there was essentially no difference in the hemolysis time between the experimental and the control tubes.

These results show that the resistance of a red cell suspension increases, but as no appreciable inhibitory substance can be recovered from the saline of a suspension which has been standing, this increased resistance is probably due to a change in the cell membrane. Such a change, possibly of the nature of disorientation in a highly oriented cell membrane, is not unlikely to occur.

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Effect on Body Growth of Small Doses of Testosterone Propionate Administered at Different Seasons.

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Since the observation that castration in the male albino rat was followed by growth inhibition,¹⁻⁶ studies have been carried out to determine whether growth could be stimulated by the injection of male sex hormones. The first attempts made with daily injection of 1 mg of testosterone propionate resulted, surprisingly enough, in growth depression.⁷ This seeming paradox was considered to have resulted from the inhibition of the pituitary growth function induced by excessive hormonal dosage.⁷⁻¹⁴

¹ Rubinstein, H. S., Abarbanel, A. R., and Kurland, A. A., *Endocrinol.*, 1939, **25**, 397.

² Donaldson, H. H., and Hatai, S., *J. Comp. Neurol.*, 1911, **21**, 155.

³ Evans, H. H., and Simpson, M. E., *Anat. Record*, 1937, **35**, 36.

⁴ Korenchevsky, V., *J. Path. and Bact.*, 1930, **33**, 607.

⁵ Commings, W. D., *J. Exp. Zool.*, 1932, **63**, 573.

⁶ Lawless, J. J., *Anat. Rec.*, 1936, **66**, 455.

⁷ Rubinstein, H. S., Kurland, A. A., and Goodwin, M., *Endocrinol.*, 1939, **25**, 724.

⁸ Hamilton, J. B., and Wolfe, J. M., *Endocrinol.*, 1938, **22**, 360.

⁹ Steinach, E., and Holzknacht, G., *Arch. f. Entwicklungsmech. d. Organ*, 1917, **42**, 490.

¹⁰ Bugbee, E. P., and Simmond, A. E., *Endocrinol.*, 1926, **10**, 360.

¹¹ Spencer, J., D'Amour, F. E., and Gustavson, R. G., *Endocrinol.*, 1932, **16**, 647.

Experiments were then begun with smaller quantities of this same substance and it soon appeared that for treatment beginning at 26 days of age, the optimal dosage for growth stimulation was 0.05 mg daily.¹⁵

During the past year, 4 different groups of animals were subjected to this treatment, first in order to substantiate our previous findings, then in order to determine the effect of climatic conditions on the growth response.

During this period of time a total of 102 animals were observed. Fifty-three of these were treated while the remaining 49 served as uninjected litter mate brother controls. Treated animals received daily subcutaneous injections of 0.05 mg testosterone propionate (Perandren) from 26-80 days. With the exception of injections, all animals were kept under similar conditions. Injections were begun in successive groups in October 1939, January 1940, April 1940 and July 1940 so that respective batches were observed during each of the 4 seasons.

Body weights were taken at weekly intervals and body lengths were observed at the end of the experiment. Data were considered statistically¹⁶ and a "significance ratio" of 3.0 was arbitrarily chosen as an indication of "probable significance" in observed differences.

Results. The mean initial weight of all control animals was 40.6 ± 0.7 g. This was not significantly different from the mean initial weight of the treated group (41.1 ± 0.7 g). The final body weight of the treated animals (188.2 ± 2.3 g) was 15.8 ± 3.3 g heavier than the final body weight of the controls (172.4 ± 2.4 g). This difference with its "significance ratio" of 4.8 is "probably significant." The mean final body length of the treated animals ($196. \pm 1.2$ mm) was only 4.8 ± 1.4 mm more than that of the controls (191.2 ± 1.1 mm) but the "significance ratio" here was 3.4. The difference is, therefore, "probably significant."

Because of the significant difference between initial weights of the controls and treated animals of the January 1940 group (test animals, 39.3 ± 1.6 g; controls, 33.2 ± 0.9 g; difference,

¹² Spencer, J., Gustavson, R. G., and D'Amour, F. E., *PROC. SOC. EXP. BIOL. AND MED.*, 1931, **28**, 500.

¹³ Spencer, J., D'Amour, F. E., and Gustavson, R. G., *Am. J. Anat.*, 1932, **50**, 129.

¹⁴ McCullagh, D. R., and Walsh, E. L., *Endocrinol.*, 1935, **19**, 466.

¹⁵ Rubinstein, H. S., and Solomon, M. L., *PROC. SOC. EXP. BIOL. AND MED.*, 1940, **44**, 442.

¹⁶ Pearl, R., *Medical Biometry and Statistics*, Saunders, Philadelphia, 1930.

TABLE I.

Differences in Weight Gains Between Respective Control and Treated Groups Whose Treatments began at Different Times of the Year. S.R. indicates "significance ratio."

Groups	Controls		Treated animals		Diff. in gain	S.R.
	No. animals	Gain (g)	No. animals	Gain (g)		
Oct.	12	128.4 $\pm$ 3.7	12	148.6 $\pm$ 4.1	20.2 $\pm$ 5.3	3.8
Jan.	10	139.3 $\pm$ 3.5	6	163.9 $\pm$ 4.4	24.6 $\pm$ 5.7	4.3
Apr.	17	118.8 $\pm$ 6.3	17	126.8 $\pm$ 6.2	8.0 $\pm$ 8.0	1.0
July	14	130.6 $\pm$ 5.4	14	148.9 $\pm$ 3.9	18.3 $\pm$ 6.4	2.9
Totals	53		49		71.1 $\pm$ 12.9	
Mean difference in gain					17.8 $\pm$ 3.2	5.5

6.1 $\pm$ 1.73 g; significance ratio, 4) comparisons were made between the weight gains of the all-treated and control groups.

From Table I it may be seen that all 4 treated groups gained more weight during the period of treatment than their respective controls. Taken as a whole the treated animals had gained 17.8 $\pm$ 3.2 g more than their controls. This leads to a "significance ratio" of 5.5 which is statistically quite "significant." However, it will be noted that the gain observed in the April animals (significance ratio of 1) was not significant.

Discussion. From these results on experiments carried out throughout the year, it appears that testosterone propionate, in suitably low dosage definitely stimulates body weight and length. It is interesting to note, however, that although all animals were taken from the same strain, their initial weights varied tremendously with the season. For example, the initial weight of the October control group was 45.7 $\pm$ 2.3 g (test=45.2 $\pm$ 1.6 g); for the January controls it was 33.2 $\pm$ 0.9 g (test=39.3 $\pm$ 1.6); for the April controls it was 52.5 $\pm$ 1.4 g (test=48.8 $\pm$ 1.7 g), and for the July group it was 31.2 $\pm$ 0.2 g (test=31.2 $\pm$ 0.6 g). From this it appears that the growth up to 26 days was similar for the October and April groups and likewise similar for the January and July groups. On the other hand the October and April animals were markedly heavier initially than the January and July groups. It appears that the animals born in Autumn and Spring (October and April groups) had a naturally more rapid early growth period than the animals born in Winter and Summer (January and July groups).

This initial weight difference in control groups is perhaps the most important consideration prompting the comparison of weight

gains rather than merely the final weights between control and test groups.

Those animals treated from April to July failed to show a definite growth response, remaining at the end in the same weight class as their controls. This seems to indicate that the result of treatment, like natural growth, is definitely influenced by climatic conditions. It appears from this study that the optimal season to begin growth studies of this nature in the albino rat is the Winter (January).

Summary and Conclusions. Testosterone propionate (Perandren) was subcutaneously injected through the 4 seasons into 4 respective groups of male albino rats in daily doses of 50 γ from 26 to 80 days. Final body weight in the treated animals was observed to be 188.2 ± 2.3 g; of the controls 172.4 ± 2.4 g. The difference of 15.8 ± 3.3 g in favor of the treated animals (S.R.=4.8) was "probably significant." Final body length of the treated group (196 ± 1.2 mm) was 4.8 ± 1.4 mm longer than that of controls (191.2 ± 1.1 mm). This was also "probably significant" (S.R.=3.4). Because of initial weight differences among the controls for the different seasons and because of a significant difference between the initial body weights of one treated group and their respective controls, comparisons were also made of "weight-gains" during the periods of observation. It was found that the treated animals gained 17.8 ± 3.2 g more than their controls. This difference in weight-gain was considered to be statistically "probably significant" (S.R.=5.5). Although animals whose treatment began in April failed to show abnormally rapid growth those groups receiving injections during the Fall, Winter and Summer did become significantly larger than their respective controls.

It is therefore concluded that while animals generally respond to appropriately small doses of testosterone propionate with increased body weight and length, seasonal variation is a potent factor in the results obtained.

The authors gratefully acknowledge the coöperation of the Ciba Pharmaceutical Products Company, Inc., for partially defraying the expense of this study and for furnishing the testosterone propionate (Perandren) used.