

TABLE III. Effect of Restricted Access to Food on Serum and Liver Lipids of 18 Pigs Fed for 173 Days.

Variable	Ad libitum	One 2-hr feeding	Two 1-hr feedings
Serum Lipids (mg/100 ml)			
Total cholesterol	104.5 $\pm$ 2.3*	100.6 $\pm$ 2.3	102.2 $\pm$ 2.3
Triglyceride	30.5 $\pm$ 4.1	48.2 $\pm$ 4.1†	41.8 $\pm$ 4.1
Phospholipid	100.6 $\pm$ 7.6	127.3 $\pm$ 7.6	106.3 $\pm$ 7.6
C/P ratio	1.04 $\pm$.06	.79 $\pm$.06†	.96 $\pm$.06
Liver lipids (mg/g)			
Total cholesterol	2.99 $\pm$.09†	2.38 $\pm$.09	2.20 $\pm$.09
Triglyceride	9.62 $\pm$ 1.59†	3.56 $\pm$ 1.59	2.57 $\pm$ 1.59
Phospholipid	3.34 $\pm$.13	3.09 $\pm$.13	2.95 $\pm$.13
C/P ratio	.90 $\pm$.03†	.75 $\pm$.03	.77 $\pm$.03

* Means with standard errors.

† P <.05.

‡ P <.01.

than the *ad libitum*-fed animals. Since the restricted-fed chickens used in the work of Cohn *et al*(1) ate one-third less feed and gained one-third less body weight than *ad libitum*-fed controls it may be that species differences exist between chickens, pigs and rats in the adjustment to restricted feeding and in the way that limited access to food influences the manner in which the food is metabolized.

Summary. Serum cholesterol and phospholipid levels were significantly elevated ($P < 0.05$) in *ad libitum*-fed pigs compared with the restricted-fed animals at the end of 84 days. After 173 days of restricted feeding pigs fed *ad libitum* had significantly higher liver cholesterol, triglyceride and C/P ratios than pigs with limited access to feed. The serum C/P ratio was significantly higher but serum triglyceride significantly lower ($P < 0.05$) in the *ad libitum*-fed pigs when compared with the group receiving one 2-

hour feeding. Marked variations in the serum lipid levels between different breeds of pigs and between sexes were observed.

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Effect of Triiodothyronine Administration on Erythrocyte Radioiron Incorporation in Rats. (28925)

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Thyroxine or triiodothyronine has produced an augmentation of erythropoiesis with a resultant erythrocythemia in both dogs and rats(1,2,3). An increase in oxygen require-

ment as a result of thyroid hormone administration reportedly acts as the stimulus to the enhanced red blood cell production. This hypothesis is consistent with the concept that the red cell mass is related to total tissue

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oxygen requirement. In the present study, L-triiodothyronine (LT^3) alone, in combination with reserpine, or D-triiodothyronine (DT^3) alone was administered to rats and the red cell radioiron incorporation determined. Oxygen utilization was assessed in rats given LT^3 and DT^3 . L-triiodothyronine produced an increased erythrocyte radioiron uptake which was not altered by the concomitant administration of reserpine. LT^3 and DT^3 produced comparable increases in iron incorporation; however, LT^3 produced a greater increase in oxygen utilization.

Methods. Female Sprague-Dawley rats weighing 180-200 g were starved for 96 hours. Water was allowed *ad libitum*. Intramuscular injections were made at 0-24-48 and 72 hours following initiation of starvation. Twenty-four control rats were given 1.0 ml of physiologic saline. Comparable groups of animals were given 250 μg of L-triiodothyronine[†] (LT^3) alone or in combination with 100 μg of reserpine.[‡] An additional group was given 100 μg of reserpine alone. Eighteen hours prior to termination of the experiment, one μc /one μg Fe^{59} citrate was injected I.V. into the tail vein. Eighteen hours later, the animals were lightly anesthetized with ether and a blood sample was removed by cardiac puncture. The radioactivity of one ml of whole blood was determined in a well counter, and red blood cell Fe^{59} utilization calculated on the basis of radioactivity of Fe^{59} recovered as related to amount of radioactivity injected (4). Total blood volume was assumed to be 5% of body weight(5).

Other groups of Sprague-Dawley rats weighing 180-200 g were starved for 120 hours. Water was allowed *ad libitum*. Prior to the initiation of starvation and at 96 hours, oxygen uptake was determined by means of a minute oxygen spirometer§ using 100% oxygen, and anhydrous potassium hydroxide as a CO_2 absorber. The results were expressed as the difference between the initial

TABLE I. Effect of LT^3 and Reserpine on RBC Fe^{59} Incorporation.

Material injected	No. rats	18 hr RBC Fe^{59} inc. % $\pm$ S.D.
Saline	24	5.42 $\pm$ 2.59
LT^3	24	19.32 $\pm$ 7.75
LT^3 & reserpine	24	18.99 $\pm$ 7.55
Reserpine	24	10.64 $\pm$ 2.75

TABLE II. Effect of DT^3 and LT^3 on RBC Fe^{59} Incorporation and Oxygen Consumption.

Material injected	No. rats	18 hr RBC Fe^{59} inc. % $\pm$ S.D.	Change in O_2 util. (ml/100 g/hr $\pm$ S.D.)
Saline	12	4.85 $\pm$ 3.54	- 28 $\pm$ 32
LT^3	11	14.95 $\pm$ 4.46	+130 $\pm$ 20
DT^3	12	16.07 $\pm$ 4.93	+ 37 $\pm$ 26

and the 96 hour oxygen utilization in ml/100 g/hr. One ml physiologic saline, 250 μg L-triiodothyronine or 250 μg D-triiodothyronine^{||} (D-T^3) was given intraperitoneally at 0-24-48 and 72 hours. At 96 hours after completion of the oxygen uptake, one μc /one μg Fe^{59} citrate was injected I.V. Eighteen hours later Fe^{59} incorporation was determined as above.

Results. The effect of LT^3 and reserpine on the 18 hour red blood cell Fe^{59} incorporation is shown in Table I. LT^3 administration increased RBC incorporation of Fe^{59} . This increase in Fe^{59} incorporation was not modified by addition of reserpine. The difference in red cell Fe^{59} incorporation of LT^3 treated rats as recorded in Table I and Table II results from alteration of the period of starvation. Animals utilized for the experiments recorded in Table I were injected intramuscularly and fasted for 96 hours, while those in experiments recorded in Table II were injected intraperitoneally and fasted for 120 hours. The effect of LT^3 and DT^3 on Fe^{59} incorporation and oxygen utilization is presented in Table II. Oxygen utilization decreased by 28 $\pm$ 32 ml/100 g/hr in saline controls, increased by 130 $\pm$ 20 ml/100 g/hr in LT^3 treated animals and increased by 37 $\pm$ 26 ml/100 g/hr in DT^3 treated rats. The Fe^{59} RBC incorporation was comparable in DT^3 and LT^3 treated rats.

|| Procured from Sigma Chemical Co., St. Louis, Mo. (Lot #T33B-073).

[†] Supplied as Cytomel® sodium (Lot #BS-0207), Smith Kline and French Research Laboratories, Phila., Pa.

[‡] Procured as SANDRIL®, Eli Lilly & Co., Indianapolis, Ind. (Lot #5134-805850FR).

[§] Aloe No. V-68808.

Comment. Triiodothyronine acts to augment erythropoiesis in the rat and dog. This accelerated erythropoiesis has been attributed to the calorogenic properties of the hormone and, thus, the imbalance between total tissue oxygen requirements and red blood cell mass has been postulated as the mechanism by which increased erythropoiesis occurs. The present results demonstrate that triiodothyronine produces increased erythrocyte Fe^{59} incorporation which is not diminished by administration of reserpine, and therefore, presumably is not dependent upon the autonomic hyperactivity produced by thyroid hormone. In the animals given DT^3 and LT^3 the increase in Fe^{59} RBC incorporation was comparable; although oxygen utilization of the LT^3 treated animals was markedly increased in comparison to the DT^3 treated animals (Table II). These results suggest that the mechanism by which triiodothyronine produces increased erythropoiesis is not dependent upon the autonomic effects of the

hormone and is not completely dependent upon changes in oxygen utilization.

Summary. LT^3 administration produced an increase in erythrocyte radioiron incorporation in rats. This response was not modified by administration of reserpine. Animals given LT^3 or DT^3 showed comparable increases in erythrocyte radioiron incorporation, however rats given LT^3 demonstrated a marked increase in oxygen utilization when compared to DT^3 treated rats.

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Influence of Excess Dietary Phenylalanine on Pregnant Rats and Their Fetuses.* (28926)

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Our earlier publication(1) provided evidence that female rats fed excess quantities of phenylalanine during pregnancy and lactation had normal litter size with no gross abnormalities. Some of the suckling rats died early and this was found to be due to decreased milk flow. The surviving offspring showed slightly elevated plasma phenylalanine levels but no phenylketonuria. This report will provide data on plasma phenylalanine and tyrosine levels of the fetuses and the role of maternal phenylalanine hydroxylase in regulating these plasma values.

Experimental. Pregnant albino rats, 3-5 months old (Holtzman Rat Co.) were fed 5

or 7% L-phenylalanine from about the tenth day of timed gestation.[†] Comparable pregnant females fed the unsupplemented diet served as controls. The animals were housed in individual cages in a dark-light room previously described(2). On the eighteenth to twenty-first day of pregnancy the rats were anesthetized with ether and blood was obtained directly from the heart. The fetal young were removed by cesarean section and then bled directly from the chest into heparinized tubes. The maternal livers were excised, weighed, and homogenized and a soluble fraction was prepared and assayed for phenylalanine hydroxylase activity according

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[†] Onset of pregnancy determined by presence of sperm in vaginal smear.