No. 2. Spleen. Died on 4th day. Inguinal glands enlarged. Spleen typical.

No. 3. Liver. Died on 5th day. Inguinal glands enlarged and spleen typical.

No. 4. Lung. Died on 5th day. Right inguinal and right axillary glands involved. Spleen typical of tularemia.

It would appear that the ruffed grouse succumbs to an experimental infection with *Bact. tularense* with the same regularity as the guinea pig and the rabbits. There appears to be little tendency for the formation of a local lesion. Although guinea pigs and rabbits usually develop macroscopic lesions by the 4th day, no lesions were visible in the grouse which died even as late as the 6th and 9th day. That the organism produces a true septicemia is indicated by its wide distribution in the body and its isolation from normal appearing tissues, such as lung. It would appear that the occurrence of tularemia in grouse as a natural disease is a probability. They are highly susceptible to the disease and subject to infestation by a natural insect carrier, common, according to Parker and Spencer, to grouse and rabbits. If the disease does occur as a natural infection in grouse, its presence might easily be overlooked at necropsy by the absense of gross lesions of the disease. It appears that no well defined human cases of tularemia have been reported as the result of cleaning grouse. This would not appear to invalidate the possibility of the natural infection of grouse, as few such birds are killed and cleaned compared with the number of rabbits. The indications justify a diligent search for tularemia in grouse dying from disease. *Bacterium tularense,* already of great importance because of its ability to produce infections in numerous species of mammals, becomes even more remarkable by its ability to produce at least experimental infection in birds.

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Significance of Reticulocyte as Index of Regeneration in Different Types of Experimental Anemias.

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A new emphasis has recently been placed on the reticulocyte count of the circulating blood through its introduction by Minot and Murphy as a convenient check of the effects of a liver diet in Pernicious

Anemia. In the untreated cases of Pernicious Anemia during the anemic stage the reticulocyte count is usually less than 1%, and sometimes *nil*. The behavior of the reticulocyte count during a spontaneous remission can hardly be studied now, since one does not feel justified in waiting for such a remission to occur. We have endeavored by the production of different types of anemias to use the reticulocyte count as a measure for the intensity of spontaneous regeneration of erythrocytes. One of the authors has recently pointed out the usefulness, in the consideration of the anemias, strictly to distinguish between the morphological and chemical phases of blood regeneration. In Phenylhydrazine anemia we encounter an anemia due chiefly to the destruction of the erythrocyte with but little loss of the hemoglobin from the body, and as far as we know, without any direct damage to the hemoglobin synthesis in the body.

The second type of anemia which we have employed—destroying erythrocytes by intravenous injections of distilled water—is similar to Phenylhydrazine Anemia in this respect that only a small amount of the liberated hemoglobin is lost through the kidney, the largest amount being disposed of within the organism.

In direct opposition to these 2 types stands the third type of anemia produced by repeated bleeding, in which form the cells are removed and no hemoglobin or hemoglobin products are left in the organism.

Before presenting our experimental data we offer these conclusions to which we have arrived in regard to the interpretation of the reticulocyte count: 3 factors are of importance in estimating the regenerative value of a given reticulocyte count. Neither the percentage of reticulocytes, nor the total number per unit volume is sufficient for a proper interpretation. The further factors for consideration are the level of the erythrocyte count and the direction in which the erythrocyte count is moving, that is, increasing or decreasing. For example, a reticulocyte count of 5% with a total erythrocyte count of 4,000,000 per cubic millimeter, means a rapid regeneration and a total reticulocyte count of 200,000 per cubic millimeter; while a 5% reticulocyte count with an erythrocyte count of 1,000,000 per cubic millimeter means a total reticulocyte count of 50,000 per cubic millimeter, and a comparatively low grade regeneration. This point is a fundamental one. The situation is further complicated as follows: A total erythrocyte count of 2,000,000 will show a certain percentage of reticulocytes, say 20% to 30%, while the blood is in the stage of rapid regeneration, but only about 2% to 5% while the blood is rapidly being destroyed.

Only when these points are taken into consideration is it possible properly to use the reticulocyte count as a measure of regeneration in the different types of anemia.

Our results, broadly speaking, show that in Phenylhydrazine Anemia there is an exceedingly rapid regeneration of the erythrocytes, with reticulocyte counts as high as 800,000 per cubic millimeter or 41.3%, a count similar to that obtained in cases of Pernicious Anemia during the period of maximal response to the feeding of raw liver. This regeneration is so intensive that it requires careful watching to produce a severe Phenylhydrazine Anemia, each injection being followed in 2 or 3 days by intensive regeneration. The danger involved is that as soon as the dosage is pushed hard, one is apt to lose the animal by a too severe anemia or by a consequent intercurrent infection. In some of our experiments, continued over 39 and 49 days respectively, we have not been able to convince ourselves of the possibility materially to slow down the regenerative process by means of a prolonged phenylhydrazine poisoning. The degree of anemia produced was such that the red count for single days was down as low as 680,000 per cubic millimeter, but usually the count was reduced to between 1,000,000 and 2,000,000 per cubic millimeter. The blood picture showed normoblasts reaching a maximum of 11,300 per cubic millimeter. Even in this high count the normoblast always represented a late stage of development with distinctly

TABLE I.

Anemia from intravenous injection of distilled water.

Day of experiment	Water intravenously	Urine		Hemoglobin	Erythrocyte	Reticulocytes	Leukocytes	Normoblasts
		In 24 hours	Remarks					
	cc.	cc.		%				
0	—	—	—	95	5,980,000	5,980=0.1%	12,700	0
1	300	—	—	94	5,870,000	11,740=0.2%	12,700	25
2	920	1200	Bloody	98	6,280,000	6,280=0.1%	13,800	28
5	950	1350	"	64	5,100,000	40,800=0.8%	28,600	29
7	940	1050	"	71	5,440,000	157,900=2.9%	46,600	93
9	675	830	"	59	4,340,000	60,700=1.4%	17,200	34
11	750	1120	"	68	4,640,000	222,600=4.8%	19,300	38
12	720	820	Not bloody	61	4,720,000	184,000=3.9%	28,300	28
13	1025	1190	Bloody	57	4,070,000	289,000=7.1%	29,400	29
14	940	600	Not bloody	52	3,650,000	157,000=4.3%	22,200	44
15	900	660	Bloody	44	3,610,000	122,700=3.4%	36,700	147
16	1230	1050	Bloody	42	2,260,000	149,200=6.6%	17,100	102
18	1520	1670	Very bloody	49	4,750,000	446,500=9.4%	42,600	766
19	—	—	—	50	4,280,000	453,500=10.6%	22,600	158

pycnotic nuclei, and only seldom did the cytoplasm show a marked degree of basophilia. On the whole the reticulocyte curve and the normoblast curve ran parallel. In all cases, the normoblasts were observed in the circulating blood for a long period of time after the anemia had fully disappeared and the reticulocyte count had returned to normal. However, we think it possible that a single normoblast may occasionally be found in the normal dog's circulating blood.

In the production of an anemia by the intravenous injection of distilled water a 16 kg. dog was used. Large amounts of distilled water were required. At first 5 injections of from 600 to 1000 cc. were made, one every 2nd or 3rd day, with a reduction of the erythrocytes from 6,000,000 to 5,000,000. Then a daily injection of from 750 to 1250 cc. of distilled water was given over a period of 6 days, which reduced the erythrocyte count to 2,300,000. The injections were usually followed by a hemoglobinuria and a diuresis. The diuresis must have produced a rather marked dehydration for the drop in the erythrocyte count and hemoglobin estimation did not occur until 1 to 2 days after the injection.

The response of the reticulocytes, erythrocytes and leukocytes was somewhat less intense than in the Phenylhydrazine Anemia. The normoblast response alone was markedly less intense than after phenylhydrazine.

A 15 kg. dog was used for the production of an anemia by repeated bleeding. A total of 1615 cc. of blood was removed in 15 days. This reduced the erythrocytes from 5,200,000 to 2,300,000. The hemoglobin was reduced 15% lower than corresponded to the same erythrocyte count in the other types of anemia. The leukocytes were only slightly affected, if at all. The response of the reticulocytes and normoblasts was much less lively than in the Phenylhydrazine Anemia, and slightly less than in the anemia from distilled water. The maximal reticulocyte count under the most rapid regeneration with 2,300,000 erythrocytes was 310,000 per cubic millimeter, or 13.2%. When compared with the Phenylhydrazine Anemia this is a very mild response; in the latter an erythrocyte count of 2,000,000 corresponded to a reticulocyte count of 476,000, or 23.8%, and an erythrocyte count of 1,100,000 to a reticulocyte count of 438,000 or 38.5%. Regeneration from the posthemorrhagic anemia was complete in 10 to 12 days, which is 1 to 2 days longer than was required for regeneration from a much lower level—980,000 erythrocytes per cubic millimeter in the Phenylhydrazine Anemia. When the anemia produced by distilled water is compared with the posthemorrhagic and with the Phenylhydrazine

Anemia it occupies an intermediate position in its degree of regenerative response.

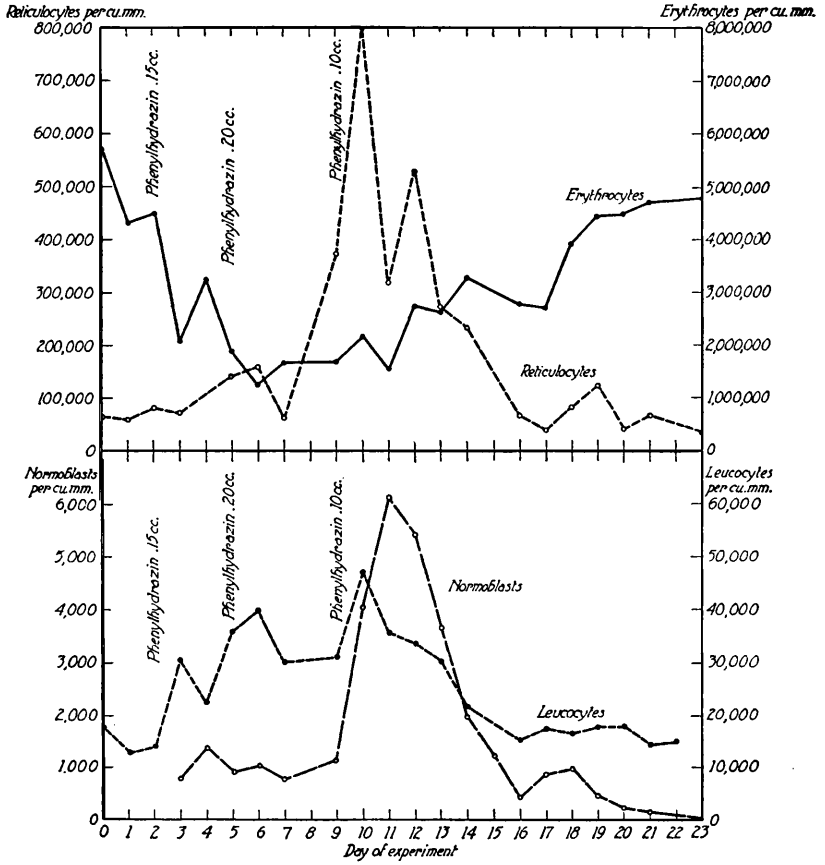

CHART 1. Blood response in Phenylhydrazine Anemia.

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Positive Effect of Tyrosine Feeding Upon Excretion of Reducing Urinary Compound in Myasthenia Gravis.

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In the course of the studies of the reducing urinary substance recently described by Medes, Berglund and Lohmann,[1] in a case of

[1] Medes, Grace, Berglund, Hilding, and Lohmann, Anne, PROC. SOC. EXP. BIOL. AND MED., 1927, XXV, 210.