

The effect of parasitism on the crab seems to be limited to the destruction of the epithelial and cuticular lining of the gut by the mechanical action of the parasite. The epithelial lining may be thinned out beneath the point of attachment of the parasite, or in other cases may be sloughed off, especially in heavy infections. Since the gregarine does not appear to pierce the cells, it must obtain its food supply from the lumen of the digestive tract. Although an exceedingly heavy infection might destroy sufficient tissue to inconvenience the host, it is doubtful if this would occur normally since, even in severe infections, comparatively few of the host's epithelial cells seem to have been materially injured.

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Reticulocyte Counts in *Bartonella Muris* Anemia.

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The striking stimulation of the bone marrow in splenectomized rats suffering with *Bartonella muris* anemia is reflected in the reticulocyte count of the peripheral blood. In the course of extensive studies in this disease¹⁻⁷ the appearance of large numbers of reticulocytes during the height of the anemia was noted.* A detailed correlation of the degree of anemia as reflected in the blood count and hemoglobin determination with the reticulocyte count and the appearance of Bartonella bodies has yielded certain interesting observations. For the study and demonstration of reticulocytes in

¹ Marmorston-Gottesman, J., and Perla, D., *J. Exp. Med.*, 1930, **52**, 121.

² Perla, D., and Marmorston-Gottesman, J., *J. Exp. Med.*, 1930, **52**, 131.

³ Perla, D., and Marmorston-Gottesman, J., *J. Exp. Med.*, 1931, **53**, 869.

⁴ Marmorston-Gottesman, J., and Perla, D., *J. Exp. Med.*, 1931, **53**, 877.

⁵ Perla, D., and Marmorston-Gottesman, J., *J. Exp. Med.*, 1932, **56**, 777.

⁶ Perla, D., and Marmorston-Gottesman, J., *J. Exp. Med.*, 1932, **56**, 783.

⁷ Sandberg, M., Perla, D., and Marmorston-Gottesman, J., *J. Exp. Med.*, 1933, **57**, 81.

* For a review of *Bartonella muris* anemia see Lauda⁸ and the papers of Perla and Marmorston-Gottesman, and the recent review of Kikuth.⁹

⁸ Lauda, E., In Kolle, W., and von Wassermann, A., *Handbuch der pathogenen Mikroorganismen*, Jena, Gustav Fischer, 3rd ed. (Kolle, W., Kraus, R., u. Uhlenhuth, P.) 1930, **8**, Liefg. 20, 1073.

⁹ Kikuth, W., *Ergebn. d. Hyg. Bakt. Immunitätsforsch. u. exp. Therap.*, 1932, **13**, 559.

large numbers, *Bartonella muris* anemia offers a readily obtained source.

Estimations of the hemoglobin, red blood cells and reticulocytes were made daily on a group of 6 splenectomized male adult rats of *Bartonella muris* carrier stock, from the day of splenectomy to the death of the rats. Hemoglobin was determined by the Dare colorimeter. For the preparations of the smears for the reticulocyte count a slight modification of the method recommended by Key¹⁰ and Thoro¹¹ was used. The dye was diluted as follows: 4 drops of a saturated aqueous solution of brilliant cresyl blue were added to 5 cc. of physiological salt solution to which had been added 2 drops of a 2% potassium oxalate solution. This was made up fresh every few days. One drop of the dye solution was placed on the slide and near it one drop of blood. The blood and dye were intimately mixed with the edge of a second slide and smears drawn. When dry the smears were counterstained with Wright's stain. The reticulocytes were clearly defined and readily counted. Smears prepared without vital staining were made for the study of Bartonella bodies.

Stucky and Brandt¹² recently reported the curve of the reticulocyte count in normal rats at different ages. They found that the drop in the hemoglobin level during the nursing period is associated with a progressive decrease in the number of reticulocytes. At birth 60% of the red cells are reticulocytes. There is a progressive decrease after this. At 21 days the reticulocytes have dropped to a 25% level. At 2-3 months normal rats show a constant 2-3% reticulocyte count. We have been able to confirm these observations. In our own stock at 3 months of age, the reticulocyte count of normal rats varied from 1.7 to 4.8%.

From the table it may be seen that with the development of anemia 4 to 5 days after splenectomy a rise in the reticulocytes occurred to a level as high as 23%. As the anemia progressed and the hemoglobin and red cell count fell, the percentage of reticulocytes progressively increased. By the 8th day after splenectomy the reticulocyte count was over 40%. By the 10th day the animals began to recover, the hemoglobin rose, the red cell count increased and the reticulocyte count progressively diminished so that on the 22nd day the count dropped in most of the rats to below 10%. Two days later they again rose as the anemia recurred to as high as 70% and

¹⁰ Key, J. A., *Arch. Int. Med.*, 1921, **28**, 5.

¹¹ Thoro, W. C., *Clinical Laboratory Methods*. Press Publishing Co., East Stroudsburg, Pa., 1926, 53.

¹² Stucky, C. J., and Brand, E., *Proc. Soc. Exp. Biol. and Med.*, 1933, **30**, 932.

TABLE I
Reticulocyte Counts in Bartonella muris Anemia of Splenectomized Adult Albino Rats

Rat No.	Before Oper.	Counts After Operation—No. Days															48		
		1	2	4	6	7	8	9	11	12	15	17	20	22	24	28		36	44
1	%Hbg. 1	110	105	100	95	20	20	30	35	60	55	80	90	115	60	80	80	90	70
	R.B.C. × 000	8500	8600	7700	6200	1300	2200	1500	2000	3200	4950	5400	6500	6400	3200	4200	5500	7000	4500
	% Retic. 3	3.2	3	6.4	4	15.6	29	50	33	30	10	8	6	2	7	22	8	1	2
	% Bart. m ⁴	0	0	0	0	30	8	R	R	0	R	R	R	2	25	0	0	0	R
2	% Hbg.	105	108	95	100	90	70	35	35	25	40	70	75	105	85	90	55	85	40
	R.B.C. × 000	8000	8200	8500	8100	7500	5000	1150	2500	3400	3200	4000	5400	5600	4450	4250	4550	6000	3200
	% Retic.	3.6	1.6	4	3	1.2	40	8	16	20	18	18	21	3	6	40	72	33	56
	% Bart. m.	0	0	0	0	0	0	50	3	2	R	R	0	R	12	15	1	0	0
3	% Hbg.	110	110	100	95	10													
	R.B.C. × 000	9000	8600	7200	5600	1400													
	% Retic.	2	1.6	3	3.4	3	Died												
	% Bart. m.	0	0	0	0	90													
4	% Hbg.	106	100	110	85	10	30	35	40	55	105	95	50	20	35	55	90	40	90
	R.B.C. × 000	8000	8600	6600	5400	2500	2800	2500	2900	3350	4650	5500	4050	2150	1000	2700	4600	2000	6200
	% Retic.	4.8	1	2.6	16	40	42	40	26.4	22	23	18	23	50	36	38	18	65	4
	% Bart. m.	0	0	0	88	7	R	R	R	0	0	2	30	2	0	0	20	10	6
5	% Hbg.	95	95	90	40	25	40	80	90	80	120	100	100	10					
	R.B.C. × 000	8500	8600	6700	5300	1800	3000	2550	4400	5450	6100	4500	5500	2700					
	% Retic.	3.7	4.4	2	23	39	47	43	33	25	22	15	5	3					
	% Bart. m.	0	0	0	75	R	R	0	R	0	R	R	R	10					
6	% Hbg.	105	115	100	85	40	25	50	45	110	100	90	70	100	105	85	45	50	40
	R.B.C. × 000	9000	8700	6500	6800	2500	2500	3000	3750	4850	5650	5150	4500	5550	5250	4850	4900	3500	3000
	% Retic.	1.7	5.4	1	12.8	38	25	41	50	52	56	52	55	23	10	6	50	36	12
	% Bart. m.	0	0	0	95	84	R	R	0	0	4	0	0	0	5	5	8	5	15

1 The Hemoglobin expressed in % as calculated from readings with the Dare Hemoglobinometer.

3 % of reticulocytes determined by counting 500 red blood cells.

4 % number of red cells containing Bartonella bodies indicated in this column.

R. Rare.

continued high till just prior to death when they dropped considerably. A persistently low reticulocyte count in the presence of continued anemia is a fatal prognostic sign.

Concomitantly with the rise in the reticulocyte count and the drop in hemoglobin percentage and the number of red cells, there was a marked decrease in the percentage of red cells and the number of red cells infected with *Bartonella* bodies. The height of the *Bartonella* infection of the red cells occurs just before the marked drop in hemoglobin percentage and the red cell count. With the development of a severe anemia, *Bartonella* bodies were found with difficulty, though when the anemia recurred the appearance of large numbers of *Bartonella* bodies again was manifest in the early phase of the relapse.

The resistance of young red cells (reticulocytes) to *Bartonella muris* is striking. In the vitally stained preparations *Bartonella* bodies were never observed within the reticulocytes. The complete disappearance of *Bartonella* bodies at the height of the anemia suggests a peculiar resistance of the new formed red cells to their invasion. This factor may account for temporary recovery from the infection that follows the anemia in most instances. In a large percentage of instances, however, particularly in males, relapses occur and death of anemia and infection result.

Conclusions. 1. Reticulocytes may constitute more than two-thirds of the red cells in the circulation at the height of the anemia in *Bartonella muris* anemia of splenectomized adult albino rats. 2. Reticulocytes are particularly resistant to invasion by *Bartonella muris*. 3. The reticulocyte response during *Bartonella muris* anemia may be used as a prognostic sign. A poor reticulocyte response is associated with death of the rat within a few hours. On the other hand a high reticulocyte count is followed by temporary recovery from the anemia though the rat may succumb during a subsequent relapse.