

# GLYCOLYSIS IN THE BLOOD OF PATIENTS WITH PERNICIOUS ANEMIA

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Following the discovery by Warburg that tumor cells have quantitative and qualitative differences from those of normal tissues with regard to their carbohydrate metabolism, many investigators have demonstrated that the method of glycolysis was most satisfactory for the study of the metabolism of the blood cells. Maclean and Weir (1) pointed out the role played by the different blood elements in glycolysis, and showed that the erythrocytes were a vital factor in the sugar consumption of normal blood. Inasmuch as the red blood cells play an important part in the glycolytic activity of normal blood, and since pernicious anemia is a disease primarily involving the red cells (both number and type), a study of the glycolysis of the blood in this malady has been undertaken for further investigation of the metabolism of the red cells.

## METHODS

Blood sugar determinations were made from the blood of nine patients with pernicious anemia before treatment and at the height of the reticulocyte response, following ventriculin therapy. Red cell, white cell and reticulocyte counts were made on corresponding days.

The blood sugar values were calculated according to the micro-method of Folin and Wu (2). All the estimations were made from blood samples obtained before breakfast at 8 A.M. The usual procedure was to withdraw 10 to 15 cc. of blood from the arm vein under sterile precautions, no tourniquet being used in order to avoid stasis. The blood was discharged into a large test tube, mixed with heparin, and kept in a warm room at a temperature of 37° C. Heparin was used as an anti-coagulant as it was shown by Falcon-Lesses (3) to have no appreciable effect on the rate of glycolysis.

The blood sugar content was determined immediately after the removal of the specimen from the patient, and then at hourly intervals for 5 hours to estimate the glycolytic activity. The red blood cell and white blood cell counts were made with U. S. Bureau of Standards pipettes and Neubauer-Levy counting chambers. Reticulocyte estimations were made from brilliant cresyl-blue film preparations.

TABLE I  
*Rate of glycolysis in pernicious anemia during relapse*

Case number	Red blood cells <i>millions per cu. mm.</i>	White blood cell count	Reticulo-cyte <i>per cent</i>	Mgm. glucose per 100 cc. blood						Rate of glycolysis in mgm. glucose per 100 cc. blood per hour					
				Initial values	Values after					1st hour	2d hour	3d hour	4th hour	5th hour	Average per hour
					1 hour	2 hours	3 hours	4 hours	5 hours						
1	1.98	8400	2.0	137	120	109	95	79	70	17	11	14	16	9	13.4
2	2.90	5500	0.5	94	87	80	74	68	58	7	7	6	6	10	7.2
3	1.80	7450	3.1	78	67	62	58	55	47	9	5	4	3	8	5.8
4	2.27	5600	0.7	86	75	65	60	55	53	11	10	5	5	2	6.6
5	1.20	5000	3.7	104	92	91	83	80	79	12	1	8	3	1	5.0
6	1.38	2550	1.8	107	95	90	83	79	75	12	5	7	4	4	6.4
7	1.81	5600	0.8	130	121	108	100	100	93	9	13	8	0	7	7.4
8	.83	4900	1.2	120	120	117	115	111	107	0	3	2	4	4	2.8
9	1.27	3650	1.6	115	113	109	107	104	101	2	4	2	3	3	2.8
Average	1.71	5405	1.7	108	99	92	86	81	76	9	7	6	5	5	6.4

## DATA

In Table I are presented the results of the glucose determinations of the blood from 9 cases with pernicious anemia during relapse. The average initial glucose value is 108.0 mgm. per 100 cc. blood. There is a uniform rate of glycolysis averaging about 6.4 mgm. per hour, the limits being 5 to 9 mgm. The average rate of glycolysis per hour per million red cells is about 3.7 mgm., which is approximately the same as in normal blood.

In Table II are listed the results of the blood sugar determinations of 9 patients with pernicious anemia at the height of the reticulocyte response following ventriculin therapy. The initial average glucose value is 100 mgm. per 100 cc. blood. The average rate of glycolysis per hour is 10.2 mgm., the extremes being 8 to 13 mgm. The average rate of glycolysis per hour per million red cells is 5.4 mgm. These averages are about 65 per cent more than the glycolytic rate before treatment.

Compared to the standard rate of glycolysis of normal blood as established by Schmitz and Glover (4), and Falcon-Lesses (3), the glycolytic activity of the blood in pernicious anemia during relapse or early remission is markedly retarded. In Chart 1 is a comparison of the average rate of glycolysis of 9 cases with pernicious anemia during relapse and in early remission with normal glycolytic activity as determined by Schmitz and Glover, and Falcon-Lesses.

Although the initial average glucose values in the cases of pernicious anemia were slightly elevated, they remained within the upper limits of normal. Many investigators have pointed out the fact that glycolysis proceeds at a faster rate with a high initial glucose concentration. In view of this, then, the rate of glycolysis in pernicious anemia would be even slower by comparison, since the initial glucose values are higher than those of normal blood. It is further noted that the initial values of the blood sugar determinations of patients with pernicious anemia in early remission are less than those of the patients in relapse, yet the average rate of glycolysis in the former cases is about 65 per cent more. In all instances, glycolysis appears to progress at a fairly uniform rate, averaging 15 to 17 mgm. per hour in the normal; 5 to 9 mgm. per hour in pernicious anemia during relapse (red blood cell counts from 0.83 to 2.90 millions per cu. mm.); and 8 to 13 mgm. per hour in pernicious anemia during early remission (red blood cell counts from 0.80 to 2.90 millions per cu. mm.).

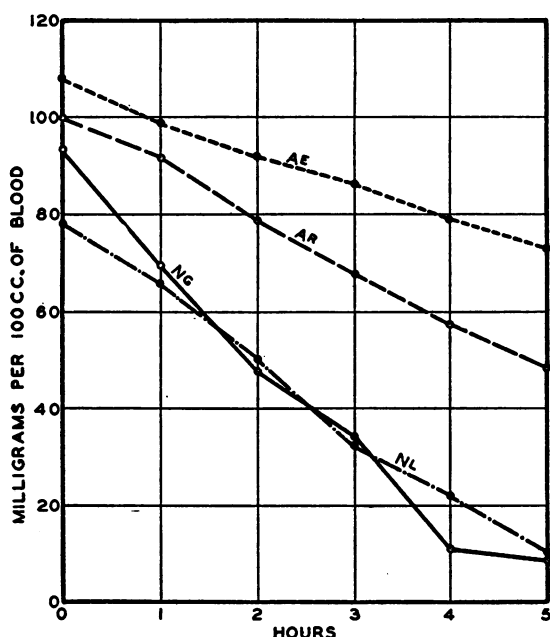
Calculated from figures for normal, the average rate of glycolysis per hour per million red blood cells is about 3.5 mgm. In pernicious anemia, the average rate of glycolysis per hour per million red blood cells is 3.7 mgm. The decreased rate of glycolysis noted in the blood in anemia as compared to that in the normal blood was definitely proportionate to the decreased number of red blood cells. In the cases of pernicious ane-

TABLE II  
*Rate of glycolysis in pernicious anemia in early remission following ventriculin therapy*

Case number	Red blood cells	White blood cell count	Reticulo- cyte	Mgm. glucose per 100 cc. blood					Rate of glycolysis in mgm. glucose per 100 cc. blood per hour						
				Initial values	Values after				1st hour	2d hour	3d hour	4th hour	5th hour	Average per hour	
					1 hour	2 hours	3 hours	4 hours							5 hours
1	2.13	12500	15.5	111	104	72	61	42	31	7	32	11	19	11	16
2	2.90	5000	6.0	85	76	68	59	50	44	9	8	9	9	6	8.2
3	2.25	6950	15.7	81	72	63	55	45	37	9	9	8	10	8	8.8
4			16.5	92	87	71	51	43	30*	5	16	20	8	13	11.8
5	1.80	5500	22.0	108	89	70	52	40	30*	19	19	18	12	10	15.6
6	1.30	4650	20.0	102	88	76	76	71	66	14	6	8	5	5	8.0
7	2.10	2750	12.5	102	93	87	79	68	58	9	6	8	11	10	8.8
8	.80	5500	42.0	121	120	108	99	89	80	1	12	9	10	9	8.2
9	1.60	9500	15.7	102	97	91	83	74	66	5	6	8	9	8	7.2
Average	1.86	6544	18.4	100	92	79	68	58	49	8	13	11	10	9	10.2

\* Less than 30 mgm.

mia in early remission, the blood presents, in addition to a decreased number of red cells, a marked increase in the number and percentage of immature red cells or reticulocytes. The average rate of glycolysis per hour per million red blood cells in this instance is 5.4 mgm. Compared to the rate of glycolysis in normal blood there is a decrease proportional to the decreased number of red cells present. However, in comparison to the rate of glycolysis per hour per million red blood cells, in pernicious



GLYCOLYTIC RATE IN VITRO OF NORMAL AND PERNICIOUS ANEMIA BLOOD.

NG = NORMAL OF SCHMITZ - GLOVER ----- 17 CASES  
 NL = NORMAL OF FALCON - LESSES ----- 6 CASES  
 AE = PERNICIOUS ANEMIA BLOOD BEFORE TREATMENT - 9 CASES  
 AR = PERNICIOUS ANEMIA BLOOD AT HEIGHT OF RETICULOCYTE RESPONSE - 9 CASES

CHART I

anemia in early remission the glycolytic activity is more rapid than in normal blood. Per unit number of red blood cells (with all factors constant), the only difference between the two is the increased number of reticulocytes. This one factor probably accounts for the accelerated rate of glycolysis which is present in the blood in early remission. As suggested by Barer, Needles and Baldrige (5), this increase in the glycolytic activity may indicate that immature red blood cells have a more active metabolism than adult red blood cells.

Five of the patients had higher red cell counts a week after therapy

was instituted, two the same, and one less, yet in all instances the rate of glycolysis was faster with the presence of increased numbers of reticulocytes. In Case 6, the rate of glycolysis was determined before therapy, at the height of the reticulocyte response, and after the immature red cells returned to normal numbers. The glycolytic activity was accelerated when the reticulocytes were present in increased numbers, although the red cell count in this instance was the lowest. This is added evidence that the type of cell present is as important as the number of cells.

According to Glover, Daland, and Schmitz (6), there is a difference of 0.004 mgm. per hour in the rate of glycolysis with white cell counts showing a variation of 10,000 per cu. mm. In the 9 cases of pernicious anemia, the greatest difference in white cells was 6000 per cu. mm. It would seem that the effect of the white blood cells on the rate of glycolysis in pernicious anemia is extremely small; the glycolytic activity being primarily correlated with the number and type of red cells which were present.

#### CONCLUSIONS

1. The average rate of glycolysis per hour in vitro in the blood of 9 cases of pernicious anemia in relapse was 6.4 mgm., the range being 5 to 9 mgm. at 37° C.
2. The average rate of glycolysis per hour in the blood of 9 cases of pernicious anemia in early remission was 10.2 mgm., the range being 8 to 13 mgm. at 37° C.
3. The average rate of glycolysis per hour per million red blood cells in blood of normal individuals is 3.5 mgm.; in pernicious anemia in relapse 3.7 mgm.; in pernicious anemia in early remission 5.4 mgm. at 37° C.
4. The retarded rate of glycolysis in pernicious anemia is proportional to the red cell decrease.
5. With all other factors constant, the increase in the rate of glycolysis per hour per million red cells in pernicious anemia in early remission is associated with the increased number of reticulocytes present.

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