

THE IRON CONTENT OF THE WHOLE BLOOD OF NORMAL INDIVIDUALS

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During the course of an investigation involving the determination of blood iron values in normal subjects, it became evident that the results were consistently higher than the figures recently reported for human blood by Murphy, Lynch, and Howard (1) and Reich and Tiedemann (2). Furthermore, hemoglobin values calculated on the basis of their iron determinations differed considerably from values obtained by them, with the Sahli and Newcomer methods. Oxygen capacity figures calculated from their iron determinations were also lower than those generally considered normal in the literature. Since the oxygen capacity method is the standard method of determining the hemoglobin content of blood, simultaneous determinations of oxygen capacity by the Van Slyke and Neill method (3) and iron determinations by the Kennedy method (4) were made on the blood of a series of normal men and women. Since the completion of this work, Sachs, Levine, and Appelsis (5), using the Wong method (6), have reported iron values for the blood of normal men that are in accord with the data recorded in this paper, although their figures for normal women are definitely lower.

In order to determine whether the discrepancies in the iron values quoted in the papers above might be explained by the methods used for estimating the iron content, a comparison was also made between the Kennedy and the Wong methods.

Methods

The subjects used for these experiments—doctors, nurses, students, and technicians—were all apparently normal individuals

between the ages of 20 and 40 years. Samples were obtained for analysis by withdrawing approximately 10 cc. of venous blood from the large arm vein of each individual and transferring the blood to a bottle containing iron-free sodium oxalate. All collections were made between the hours of 8.30 a.m. and 10 a.m. during the months of July and August.

TABLE I

Results of Analyses for Iron Content and Oxygen Capacity on Blood of Ten Normal Men and Ten Normal Women with Oxygen Capacity Also Calculated from Total Iron Content and Protein Iron Content

Case No.	Sex	Red blood cells		O ₂ capacity			Sex	Red blood cells		O ₂ capacity				
				Iron	Calculated from total Fe	Calculated from protein Fe				Experimental	Iron	Calculated from total Fe	Calculated from protein Fe	Experimental
1	M.	6.03	57.1	22.8	22.4	22.6	F.	5.03	43.9	17.6	17.2	16.5		
2	"	5.91	53.5	21.4	21.0	21.0	"	5.23	45.9	18.4	18.0	18.5		
3	"	5.69	51.3	20.5	20.1	20.4	"	4.70	42.0	16.8	16.4	17.4		
4	"	5.51	50.5	20.2	19.8	19.4	"	4.85	45.5	18.2	17.8	19.0		
5	"	5.22	49.8	19.9	19.5	20.1	"	4.32	48.2	19.3	18.9	18.4		
6	"	5.45	48.1	19.2	18.8	19.4	"	4.81	44.0	17.6	17.2	17.1		
7	"	5.66	50.5	20.2	19.8	20.7	"	5.12	46.5	18.6	18.2	18.7		
8	"	5.61	53.0	21.2	20.8	20.9	"	5.46	49.8	19.9	19.9	19.8		
9	"	5.50	52.3	20.9	20.5	20.5	"	5.29	45.7	18.3	17.9	16.7		
10	"	5.24	49.3	19.7	19.3	20.1	"	5.27	46.5	18.6	18.2	18.4		
Average...		5.58	51.5	20.6	20.3	20.5		5.01	45.8	18.3	18.0	17.7		

The oxygen capacity was then determined by the method of Van Slyke and Neill (3). Iron was determined by the Kennedy method (4). The blood was digested without difficulty in 25 × 200 mm. Pyrex glass test-tubes instead of the Kjeldahl flasks recommended by Kennedy.

In comparing the Kennedy and Wong methods, the same pipette was used to measure both blood samples, 1 cc. samples being used for both methods. We found better results could be obtained in the Wong method when 4 cc. of distilled water were added to the blood before the concentrated sulfuric acid was added.

The red blood cell counts were made on the samples obtained from the arm vein. Standard pipettes and counting chambers were used.

Results

The results of the analyses are shown in Table I. The iron values are converted into oxygen capacity figures by multiplying

TABLE II
Results of Determination of Iron in Whole Blood of Normal Individuals by Kennedy and Wong Methods

Case No.	Fe per 100 cc. blood		Red blood cells <i>millions per c.mm.</i>
	Kennedy method <i>mg.</i>	Wong method <i>mg.</i>	
1	51.5	50.5	5.05
2	52.5	47.6	5.30
3	55.3	52.7	5.47
4	53.8	50.5	4.80
5	50.8	47.2	4.79
6	51.0	49.3	4.85
7	54.5	53.2	5.16
8	55.3	54.4	5.77
9	53.8	49.5	5.31
10	57.2	55.6	5.57
11	51.6	45.4	6.16
12	52.1	50.3	5.08
13	51.3	49.8	4.67
14	52.7	48.8	5.48
15	56.2	52.1	5.80
Average.....	53.31 ± 1.21* ± 1.94†	50.26 ± 1.83* ± 2.72†	5.284

* Probable error.

† Standard deviation.

the mg. per cent of iron by 0.400. (Since 1 mole of oxygen occupies 22,400 cc. at 0°, 760 mm., the molal ratio Fe:O₂ = 1:1 corresponds to a ratio of gm. of Fe to cc. of O₂ = 56:22,400 = 1:400, or mg. of Fe to cc. of O₂ = 1:0.400.) McIntosh (7) has shown that normal blood contains 1.02 mg. of non-protein iron per 100 cc. Therefore we have also converted the iron values to oxygen capacity after subtracting this figure from the total iron values.

In Table II are shown the mg. of iron in 100 cc. of the same blood analyzed by the Kennedy and Wong methods.

DISCUSSION

The data presented in this paper show that there is a close agreement between the hemoglobin content of normal human blood as determined by the oxygen capacity method and the Kennedy iron method. Therefore, the determination of iron offers an easy means of estimating the hemoglobin content of blood or of standardizing colorimetric methods of estimating hemoglobin. If we add the figures of eight of the cases from Table II, which were not included in Table I, the average iron content of the blood of eighteen normal men, determined by the Kennedy method, varied from 49.3 to 57.2 mg. per 100 cc., with an average of 52.5 mg. per 100 cc. of blood. For the ten normal women the iron content varied from 42.0 to 49.8 mg. per 100 cc. of blood, with an average of 45.8 mg.

In Table III the results of the recent iron determinations in human blood and the oxygen capacity and hemoglobin figures calculated from the iron content are compared to the hemoglobin figures recorded in the literature for normal men and women. The data of Reich and Tiedemann are not included in Table III because their normals can hardly be called that in the strict sense of the word. The iron values reported in this paper agree with the values of hemoglobin reported on larger series of cases by Haden (8), Osgood (9), and Wintrobe and Miller (10). The results of Murphy and coworkers are definitely lower than would be expected for blood with normal hemoglobin content.

As shown in Table II, the Kennedy method gave distinctly higher results than the Wong method. Although the simplicity of the Wong method recommends its use, in our experience the Kennedy method proved to be more satisfactory.

SUMMARY

1. There is a close agreement between the hemoglobin content of blood as determined by its iron content and oxygen capacity.
2. The blood iron content of eighteen normal men, determined by the Kennedy method, varied from 49.3 to 57.2 mg. per 100 cc., with an average of 52.5 mg.

TABLE III

Results of Recent Iron Determinations in Human Blood, and Oxygen Capacity and Hemoglobin Values, Calculated from Iron Content, and Normal Values of Hemoglobin and Oxygen Capacity As Recorded in Recent Literature

Authors	Men				Women			
	Red blood cells	Iron	O ₂ capacity	Hb	Red blood cells	Iron	O ₂ capacity	Hb
	mil- lions per c.mm.	mg. per 100 cc.	vol. per cent	gm. per 100 cc.	mil- lions per c.mm.	mg. per 100 cc.	vol. per cent	gm. per 100 cc.
Helmer and Emerson.....	5.51	52.50	21.0	15.66	5.01	45.75	18.3	13.68
Sachs <i>et al.</i> (5).....	5.00	50.01	20.0	14.93	4.46	42.67	17.0	12.74
Murphy <i>et al.</i> (1).....	5.35	44.84	17.9	13.38	4.92	42.48	17.0	12.68
Sackett (11).....	5.09	56.02	22.4	16.71	4.68	51.20	20.5	15.28
Haden (8).....	5.08		21.2	15.83	4.26		17.3	13.34
Osgood (9).....	5.40			15.80	4.80			13.70
Wintrobe and Miller (10)....	5.85			15.87				

3. The blood iron content of ten normal women varied from 42.0 to 49.8 mg. per 100 cc., with an average of 45.8 mg.

4. Higher iron values were obtained with the Kennedy method than with the Wong method.

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