previously were much over standard weight. Different patterns of post-operative weight loss are discussed.

Defective fat absorption follows gastrectomy, but the significance of this alone in relation to body weight is doubtful. Both dietary calorie intake and fat absorption are related to body weight, and failure to gain is greatest when both are impaired.

When loss of weight follows operation it occurs in the first few post-operative weeks, and thereafter the patient cannot regain it. The literature upon dietary requirements after severe weight loss and that dealing with the catabolic phase after injury or operation are reviewed.

Early post-operative drip feeding of a high-calorie high-protein mixture can prevent the loss of weight in the early post-operative period.

I wish to thank Mr. R. Marnham, who sponsored the work; the surgeons of St. George's Hospital for allowing me to investigate their patients; Professor Nicholas Martin for his advice and for affording me facilities in the Department of Biochemistry at St. George's; Professor T. Crawford and Dr. Hugh Gainsborough for much encouragement and help; and Dr. W. J. Martin for statistical advice. The weight tables were kindly lent by Dr. Malcolm Purvis.

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One of the subjects discussed at a recent meeting of the Fever Group of the Society of Medical Officers of Health at Fazakerley Hospital, Liverpool, was acute gastro-enteritis. Dr. E. BAILY (Park Hospital, London) summarized the bacteriological aspects, and drew attention to the importance of unmasking latent roughness in physically smooth colonies: the addition of a 1 in 500 solution of acriflavine to a saline suspension would at once cause auto-agglutination of serologically rough colonies and so prevent mistakes of interpretation. He showed slides illustrating finger contamination: a half-minute wash in antiseptic followed by a half-minute wash in running water was required for satisfactory cleansing. Dr. STANLEY BANKS (Park Hospital, London) dealt with the practical applications of bacteriological findings. Cubicles were no safeguard against cross-infection unless they were scrupulously clean: all unnecessary fittings must be removed, vacuum cleaning used, and small hand-towels, for use once only, provided. Nail brushes were often infected and were not necessary. Dr. R. B. MACKAY (Royal Children's Hospital, Manchester) and Dr. S. KEIDAN (University of Liverpool) gave brief reports on the use of polymixin and neomycin respectively in gastro-enteritis. Both antibiotics seemed to have an effect as least as good as other antibiotics on the disease: they appeared to have no toxic effects when given by mouth.

GAS TRANSPORT FUNCTION OF THE BLOOD IN CONGENITAL FAMILIAL METHAEMOGLOBINAEMIA

BY

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In normal blood only 1 to 2% of haemoglobin is present in the form of methaemoglobin, for, although methaemoglobin is constantly being formed by the oxidation of haemoglobin, there exists a very efficient mechanism within the red cell for the reduction of methaemoglobin to haemoglobin (Gibson, 1943). Methaemoglobin occurs clinically when drugs such as phenacetin or nitrites oxidize haemoglobin more rapidly than the normal reducing mechanism can function, or when there is an inherent defect in the reducing mechanism, as in congenital methaemoglobinaemia. Iron occurs in methaemoglobin in the oxidized or ferric form and the pigment is inert as an oxygen carrier.

It was first observed by Douglas *et al.* (1912) and by Barcroft *et al.* (1926) that the presence of abnormal derivatives of haemoglobin not only diminishes the oxygen capacity of the blood but also alters the readiness with which the blood releases oxygen to the tissues. The latter change is reflected in a shift of the oxygen dissociation curve. A shift of the curve to the left indicates an impaired release of oxygen to the tissues due to a greater affinity of haemoglobin for oxygen. Hitzenger (1932) and Gibson and Harrison (1947) each found in one case of congenital methaemoglobinaemia a shift to the left of the oxygen dissociation curve. On the other hand, Litarczek *et al.* (1930), Eder *et al.* (1949), and Waisman *et al.* (1952) reported no alteration of the oxygen dissociation curve in single cases of methaemoglobinaemia.

In view of the conflicting nature of these reports the opportunity was taken to study gas transport in two cases of familial methaemoglobinaemia.

Case 1

A married woman aged 44 was admitted to hospital in March, 1953, for an operation for hallux valgus and vari-

FIG. 1.—Family tree, with cyanosed subjects indicated by solid symbols, Case 1 and 2 having proved methaemoglobinaemia.

cose veins. She was cyanosed, and she said that she had been so throughout her life. There was a family history of cyanosis (Fig. 1) but some years previously, when in a maternity hospital, she had been told she had congenital heart disease. The cyanosis varied from day to day, was worse in winter, and was never entirely absent. There was no history of dyspnoea or undue fatigue.

On examination the patient was pale and cyanosed. The cyanosis had no special quality to distinguish it from the cyanosis of cardiac or respiratory disease, although it was more intense than the usual cyanosis due to cardiac or respiratory disease. There was no clubbing

of fingers. Apart from the cyanosis and the disabilities for which she sought surgical treatment, clinical examination was negative. The patient's two children (Fig. 1), aged 18 and 17, were examined, and neither was found to be cyanosed, nor was methaemoglobin detected.

Methaemoglobin was identified spectroscopically in the patient's venous blood, and the results of estimations before and during treatment with ascorbic acid and methylene blue, together with other laboratory findings, are shown in the Table and Figs. 2, 3, and 4.

FIG. 2.—Effect of treatment with methylene blue and ascorbic acid on methaemoglobin levels in Case 1. The times of oxygen dissociation curve studies are indicated.

FIG. 3.—Oxygen dissociation curve in Case 1 before treatment and after ascorbic acid (shifted to the left), and after treatment with methylene blue (normal).

FIG. 4.—Carbon dioxide dissociation curves of normal blood compared with values determined for Case 1.

Results of Laboratory Findings

Curve No.	Date	Time	R.B.C. Mill./c. mm.	P.C.V. %	Total Pigment G. %	O ₂ Carrying Hb		Met Hb		Pressure CO ₂ mm. Hg	(Total CO ₂) ₀ * Vol. %	(Total CO ₂) ₁ † Vol. %	(Total CO ₂) ₂ ‡ Vol. %	pH _§ †	PO ₂ for Hb=HbO ₂ §		HbO ₂ Vol. % at PO ₂ =20	I/K
						HbO ₂ Vol. %	G. %	G. %	Total Pigment %						pH _§	pH _§ =7.4		
Case 1																		
1	27/3/53	Before treatment	4.5	37	13.5	13	9.5	4	30	{ 20 40 60 40	38	43	45	7.61	23	22.5	55	1,500
2	31/3/53	After methylene blue i.v.	4.5	37	13.5	17	12.5	1	6		49	54	56	7.39			40	2,700
		After ascorbic acid orally									57	63	67.5	7.29			32	3,700
3	15/6/53	After methylene blue i.v.				16.5	12.5	1.5	10	40	48	55	57	7.40	23	23	40	2,700
4	26/6/53	After methylene blue orally	4.8	39	14	16	12.2	1.8	12	40	45	52	55	7.38	23	22	40	2,600
5	19/1/54	After methylene blue orally	4.8	40	14	17	12.7	1.3	9	40	47		56	7.39	27.5	27	32	3,800
6	19/1/54	After methylene blue i.v.				17.5	13	1	6	40	46		55.5	7.38	27	26	33	3,500
Case 2																		
1	22/9/53	Before treatment		52	18	19	13.5	4.5	25	{ 20 40 60	34	40	43	7.58	23.5	23	54	1,600
											45	52	55	7.38			39	2,900
											52	60	65	7.27			32	3,700
Mean values for normal blood						19.5			2	{ 20 40 60	37 48 56	43 55 64	44 57 69	7.60 7.40 7.30	27	27	46 33 26	2,100 3,600 5,200

* (Total CO₂)₀=Total carbon dioxide of oxygenated blood at a CO₂ partial pressure of 40 mm. Hg.

† (Total CO₂)_†=Total carbon dioxide of reduced blood at CO₂ partial pressure of 40 mm. Hg.

‡ pH_§=Plasma pH determined in true plasma from blood saturated with oxygen when PCO₂ equals 40 mm. Hg.

§ PO₂ for Hb=HbO₂=Partial pressure of oxygen at which haemoglobin saturation with oxygen is 50% at the plasma pH of the recipient and at standard plasma pH of 7.4.

K=Affinity constant of haemoglobin for oxygen.

Case 2

This patient, a man aged 32, is the only sibling (Fig. 1) of Case 1 with a history of cyanosis. Cyanosis was clinically more striking than in Case 1, but pallor was not present. There was no history of dyspnoea, fatigue, or headache or any evidence of cardiac or respiratory disease. As in Case 1, methaemoglobin was identified spectroscopically in the patient's blood: the results of laboratory findings are shown in the Table.

Methods

Methaemoglobin was identified by the presence of an absorption band at 630 m μ which disappeared on the addition of a few drops of 10% sodium cyanide. For this examination a Hartridge reversion spectroscope was used. Methaemoglobin was estimated by a modification of the method of Evelyn and Malloy (1938), using an S.P. 600 spectrophotometer, and by gas analysis (Peters and Van Slyke, 1931). Total haemoglobin was estimated after conversion to cyanmethaemoglobin as described by Evelyn and Malloy (1938). It is worthy of note that even when methaemoglobin accounted for 20–30% of the total haemoglobin the characteristic absorption band could not easily be seen in a 10% aqueous dilution of blood in a 1-cm. layer, using the reversion spectroscope.

Oxygen and carbon dioxide dissociation curves were obtained as described by Valtis and Kennedy (1954). The value I/K at different pH was determined from the Hill-Barcroft equation, taking $n=2.5$ (Adair, 1925), by equilibrating blood in tonometers at a standard oxygen pressure of 20 mm. Hg and at various CO₂ pressures (20, 40, and 60 mm. Hg). All the values for CO₂ in the serum of whole blood, whether reduced or oxygenated, were directly determined because of the unknown properties of methaemoglobin in this physico-chemical system.

Results

In both cases, before treatment, when methaemoglobin levels were 30% and 25% respectively, the oxygen dissociation curves showed a shift to the left. In Case 1 this was corrected after treatment with methylene blue, when, on three occasions with methaemoglobin values of 6, 9, and 6%, the oxygen dissociation curve was found to be normal. After treatment with ascorbic acid, with methaemoglobin values of 10 and 12%, the oxygen dissociation curve on two occasions was abnormal. These and other results are set forth in detail in the Table and in Figs. 2, 3, and 4. Conventional symbols, the meanings of which are explained in the footnotes to the Table, are used for the headings of several of the columns. The position of the oxygen dissociation curve is indicated by the partial pressure of oxygen at which 50% of the haemoglobin is oxygenated and by the affinity constant K.

In both cases the oxygen dissociation curve before treatment was shifted to the left both at the pH of the blood and at standard pH (see Table). The value of I/K was found to be 2,700, compared with mean values of 3,600 in six normal subjects. The values of I/K plotted against the partial pressure of CO₂ are in these cases of methaemoglobinaemia proportional to but much smaller than the normal.

When considering results for carbon dioxide due allowance must be made for the amount of total pigment present in reduced blood and for active haemoglobin in oxygenated blood. When this was done the position and slope of the carbon dioxide dissociation curves, the difference in the CO₂ content of reduced and oxygenated blood, and the difference between CO₂ content of true oxygenated plasma and oxygenated whole blood were all within normal limits.

In Case 1, after treatment with methylene blue intravenously, the oxygen dissociation curve and consequently

the relationship of I/K to the partial pressure of CO₂ both became normal. The position of the CO₂ dissociation curve of reduced and oxygenated blood, and the CO₂ difference between them, also became normal. Later, after treatment with ascorbic acid, the oxygen dissociation curve on two occasions was found to have the same abnormality as before treatment. The carbon dioxide dissociation curves for reduced and oxygenated blood were normal. Still later, after prolonged treatment with methylene blue, the oxygen dissociation curve was once more normal in shape and position.

Discussion

As has already been mentioned, Hitzenger (1932) found in a case of congenital methaemoglobinaemia a shift to the left of the oxygen dissociation curve. Gibson and Harrison (1947) also found a similar alteration in a single case of familial congenital methaemoglobinaemia. At an earlier date Litarczek *et al.* (1930) observed a slight shift of the oxygen dissociation curve to the left in a case of congenital methaemoglobinaemia, but it was attributed by these workers to an alteration in the alkalinity, buffering capacity, and the I/K to pH relationship of the patient's blood, rather than to the presence of methaemoglobin *per se*. Eder *et al.* (1949) reported an investigation at the Harvard Fatigue Laboratory of a case of congenital methaemoglobinaemia in which the oxygen dissociation curve was found to be normal in position; and Waisman *et al.* (1952) described a similar finding in another case.

In our two siblings with familial congenital methaemoglobinaemia with 25 to 30% methaemoglobin we found the oxygen dissociation curves to be shifted to the left, both at the actual pH of the blood and when corrected to standard pH. This shift was less in magnitude than that described by Hitzenger (1932) in his patient with methaemoglobinaemia, but was substantially the same as that reported by Gibson and Harrison (1947). When allowance was made for the inactive pigment present, the position and slope of the carbon dioxide dissociation curves were found to be normal. When considered in relation to the Hill-Barcroft equation

$$v\text{CO}_2 = c + b(c\text{H} \times 10^*)$$

this finding removes any suspicion that the alkalinity (value c) or the buffering capacity (value b) of the blood might be responsible for the altered oxygen dissociation curves. Similarly, changes in the value I/K plotted against the partial pressure of carbon dioxide, which indicates the relationship between the affinities of haemoglobin for oxygen at different pH, were proportional to changes occurring in normal blood.

Darling and Roughton (1942) reported observations on the oxygen dissociation curve of blood containing methaemoglobin prepared by the oxidation of haemoglobin *in vivo* and *in vitro*. By using solutions of laked blood in strong phosphate buffer they avoided interference by corpuscular factors and by pH changes due to varying degrees of haemoglobin saturation with oxygen. In addition, they studied the oxygen dissociation curve without adding carbon dioxide to the tonometers and were thus able to exclude its effect. They found a shift to the left of the oxygen dissociation curve proportional to the amount of methaemoglobin present and concluded that these findings were attributable to the presence of molecules of both haemoglobin and methaemoglobin containing the ferrous and ferric atoms in various atomic proportions.

Since methaemoglobin in sufficient concentration is itself thought to be responsible for the shift of the oxygen dissociation curve, it was natural in our case to expect this abnormality to be abolished by effective treatment. Three curves were performed after treatment with methylene blue, and all were found to have returned to normal. Gibson and Harrison (1947) have reported a similar return to normal after the addition of methylene blue to the blood of their patient *in vitro*. During and after treatment with ascorbic acid the oxygen dissociation curve was

studied on two occasions, and both curves showed a shift to the left of the same magnitude as before treatment.

The seeming contradiction between our findings on the one hand and those of Eder *et al.* (1949) and Waisman *et al.* (1952) on the other may be resolved by postulating the existence of two or more kinds of congenital methaemoglobinaemia not completely distinguished by methods at present available. On very different grounds Lutembacher (1947) and Hörlein and Weber (1948) have made similar suggestions. An alternative explanation is that it is not methaemoglobin itself but some other factor which is responsible in congenital methaemoglobinaemia for the abnormality of the oxygen dissociation curve. From our findings it would seem that this factor must be influenced differently by ascorbic acid and by methylene blue. It was found by Litarczek *et al.* (1931) that glutathione exerts an influence on the oxygen dissociation curve, and it seems to us that glutathione has been inadequately investigated in methaemoglobinaemia.

The mode of inheritance of familial methaemoglobinemia is still uncertain. A family tree suggestive of a Mendelian dominant character has been reported by Baltzan and Sugarman (1950). On the other hand, Gibson and Harrison (1947) and Codounis (1952) each described a family in which inheritance as a recessive character seems more likely. In our family, too (Fig. 1), inheritance as a recessive character seems the more probable.

Summary

The oxygen dissociation curves of two cases of familial congenital methaemoglobinaemia have been studied.

A shift of the curve to the left was found in both cases. This indicates impaired efficiency of oxygen delivery to the tissues in addition to what is attributable to loss of oxygen capacity due to the presence of an inert pigment.

Treatment with methylene blue restored the curve to normal. After treatment with ascorbic acid the curve remained abnormal.

These observations and published reports of similar studies are discussed. It is concluded either that there is more than one kind of congenital methaemoglobinaemia or that the alteration in oxygen transport in the condition is due to some factor other than methaemoglobin itself.

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BLOOD CHANGES ASSOCIATED WITH DISSEMINATED TUBERCULOSIS

REPORT OF FOUR FATAL CASES AND REVIEW

BY

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Although anaemia is common to all forms of tuberculosis it is rarely severe (Wintrobe, 1951), and usually normochromic or hypochromic. Chapman and Whorton (1946), in a clinico-pathological study of 63 cases of generalized miliary tuberculosis, noted a moderate normochromic anaemia in 84%. The lowest haemoglobin level was found to be 43%, the level in the majority ranging from 65% to 85%. The white-cell count was persistently low for two weeks or more in 11 out of a total of 49 patients in the above series, a relative neutrophilia being usually associated with leucopenia. The haematological picture in two cases resembled aplastic anaemia, and the presence of immature white cells and nucleated red cells in one case suggested a leuco-erythroblastic anaemia. Dyke (1924) recorded a case with generalized miliary tuberculosis associated with aplastic anaemia. Rosenthal (1928) has reported on aplastic anaemia as a complication of miliary tuberculosis, and referred to the haemorrhagic diathesis sometimes resulting from thrombocytopenia. Crail *et al.* (1948) described four cases of miliary tuberculosis associated with myelofibrosis and suggested that tuberculosis was the predisposing factor in the marrow changes. The same authors stated that the bone marrow in miliary tuberculosis shows, in the early stages, hyperplasia of all haemopoietic tissue, followed later by hypoplasia or aplasia.

Bone-marrow infiltration by disease in miliary tuberculosis seems to be not uncommon, Chapman and Whorton (1946) describing its presence in 33 out of 37 cases at necropsy. They believed that the presence of miliary tubercles in bone marrow did not necessarily predispose to depression of blood formation. Doan and Sabin (1927) produced extensive tuberculous lesions of the bone marrow of rabbits following the intravenous injection of virulent tubercle bacilli. There was an initial depression of marrow function, as judged by peripheral blood studies, followed, in animals that survived, by compensatory hyperactivity and finally by functional and morphological recovery.

In the rare condition of tuberculous splenomegaly, cytopenic states are recognized as being due to a hypersplenic mechanism, the bone marrow being normal or hyperplastic (Engelbreth-Holm, 1938). Polycythaemia has also been recorded in association with tuberculous splenomegaly (Guild and Robson, 1950).

Leukaemoid reactions accompanied by leucocytosis and the presence of immature white cells in the peripheral blood, making a differentiation from myeloid leukaemia difficult, is mentioned by Forkner (1938). Gardner and Mettier (1949) described a lymphocytic leukaemoid reaction of the blood associated with miliary tuberculosis.

Muller (1943) reviewed in detail the clinical significance of the blood changes associated with tuberculous infection.

The following four cases of generalized tuberculosis are reported in view of the profound haematological