Iron Metabolism in Patients Undergoing Regular Dialysis Therapy

B. BROZOVICH, W. R. CATTELL, M. F. COTTRALL, M. M. GWYTHYER, J. M. McMILLAN
J. S. MALPAS, A. SALSURY, N. G. TROTT

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Summary

Thirteen patients with chronic renal failure maintained on regular renal dialysis were studied. Seven proved to have iron deficiency on the basis of marrow iron studies, reticulocyte iron uptake, and saturation of the serum iron-binding capacity. They absorbed iron when given it by mouth and were able to utilize it for haemoglobin formation. Iron-deficient patients given 600 mg of ferrous sulphate daily for three months showed an increase in haemoglobin, but the failure to replace stores of iron is probably related to their relatively limited ability to absorb iron and the variable but sometimes considerable blood loss occurring with each dialysis.

The loss may be occult, and prolonged iron therapy may be required. This is most safely achieved by giving iron by mouth.

Introduction

The anaemia of chronic renal failure is commonly normocytic and normochromic. Iron-deficiency anaemia may occur and when transfusion is avoided is not uncommon in patients undergoing regular dialysis therapy. Controversy exists regarding the effectiveness of oral iron therapy in such patients

resulting in the advocacy of prolonged parenteral iron therapy (Crockett *et al.*, 1967). Such treatment can, however, result in iron overload (Curtis *et al.*, 1969).

The present study was undertaken to re-examine both the absorption of oral iron and the utilization of oral and intravenous iron in patients undergoing membrane dialysis who were carefully studied with respect to intercurrent iron loss.

Methods and Patients

Thirteen patients on regular dialysis therapy for 3 to 34 months were studied. All were dialysed for a minimum of 28 hours weekly with a Kiil dialyser. They were in good health and received no haematinics at the time of the isotope studies. Two patients (Cases 6 and 13) had had 750 mg and 1,000 mg of iron intravenously 18 and 15 months before the investigation. Case 13 had had 12 pints (6.8 litres) of blood, Case 4 5 pints (2.8 litres), Cases 1, 2, 6 and 11 each 2 pints (1.1 litres) previously. The details of diagnosis are given in Table I.

The haemoglobin and packed cell volume (P.C.V.) were measured at the beginning of the study, after each dialysis, and before and after iron therapy by methods described by Dacie and Lewis (1968); serum iron was measured by an automated micromethod (Garry and Owen, 1967) and iron-binding capacity by a semiautomated micromethod (Brozovich and Copestake, 1969). Serum and whole blood folate levels were measured by an automated method described by Millbank *et al.* (1970). Reticulocyte uptake of iron in vitro has been shown to be a sensitive guide to iron deficiency and was carried out by the method of Brozovich (1971). The use of automated micromethods for all of these determinations reduced the amount of blood taken from these patients to a minimum.

Bone marrow specimens were obtained before the study began and again after iron therapy. They were stained for iron by the method of Rath and Finch (1948). Each was independently examined and graded for its iron content by two of us (J.S.M. and A.S.).

The absorption studies were carried out by means of a whole body counting technique using four NaI (Tl) crystals in a shielded room (Cottrall *et al.*, 1968). Pulse height analysis

St. Bartholomew's Hospital, London E.C.1

B. BROZOVICH, M.D., PH.D., Lecturer, Department of Haematology

W. R. CATTELL, M.D., F.R.C.P.E.D., Consultant Physician

J. S. MALPAS, D.PHIL., M.R.C.P., Senior Lecturer in Medicine

Institute of Cancer Research, Royal Marsden Hospital, Sutton, Surrey

M. F. COTTRALL, B.A., M.SC., Member of Scientific Staff, Physics Department

M. M. GWYTHYER, B.SC., Member of Scientific Staff, Radiotherapy Research Unit

N. G. TROTT, PH.D., F.INST.P., Reader in Physics Applied to Medicine

Regional Renal Dialysis Unit, St. Leonard's Hospital, London N.1

J. M. McMILLAN, M.B., M.R.C.P., Senior Registrar

Brompton Hospital, London S.W.3

A. SALSURY, M.D., Consultant Haematologist

TABLE I—Details of Cases

Case No.	Age	Sex	Diagnosis	Hb (g/100 ml)	P.C.V. (%)	Serum Fe (μg/100 ml)	T.I.B.C. (μg/100 ml)	Saturation of T.I.B.C. (%)	Marrow Iron		Reticulocyte Iron Uptake (μg/ml)
									Grade	Granules (%)	
1	23	M.	Triad synd.	5.2	20	25	542	4.5	0	2.2	0.723
2	37	M.	G.N.	6.7	22	22	314	7.0	0	0.8	0.532
3	47	M.	Uric acid nephropathy	8.6	28	30	329	9.0	0	0.8	0.376
4	48	F.	C.R.F.	8.36	24	—	—	—	0	0.4	—
5	33	F.	G.N.	6.7	18	27	477	5.6	1	3.0	0.005
6*	27	M.	P.N.	8.3	24	65	292	22	1	1.8	0.120
7	35	F.	P.N.	8.1	24	137	377	36	1	1.0	0.363
8	31	M.	Papillary necrosis	7.4	25	41	347	12	2	3.6	0.350
9	40	M.	P.N.	7.4	20	80	328	24	2	2.8	0.551
10*	37	M.	G.N.	8.7	25	14	256	6	3	17.4	0.305
11	26	F.	G.N.	11.0	33	—	—	—	3	11.8	—
12*	42	M.	G.N.	8.6	24	13	284	5	4	5.8	0.206
13	29	M.	G.N.	7.0	19	99	246	40	6	21.0	0.244

G.N. = Glomerulonephritis. P.N. = Pyelonephritis. C.R.F. = Chronic renal failure—cause unknown. T.I.B.C. = Total iron-binding capacity.

*Treated with analgesics.

was performed with a multichannel analyser. The fasting subject received a drink containing 1 μ Ci of ^{59}Fe in the ferrous state; the dose was prepared by adding 1 μ Ci of ^{59}Fe ferrous chloride to a mixture of 50 mg of ascorbic acid and 5 mg of elemental iron as ferrous sulphate. After the dose had been administered the initial count rate for the subject was measured within an hour to obtain the initial 100% value. Retention of the dose was estimated by repeating the measurement on the fourteenth day. A blood sample was taken at that time and measured under low background conditions in a 3-in (7.5-cm) diameter NaI (T1) crystal well counter to provide a value for the utilization of absorbed ^{59}Fe for haemoglobin formation.

A second method of measuring absorption was also used (Cottrill and Trott, 1971). A non-absorbable ^{51}Cr chromic chloride marker was given half an hour after the ^{59}Fe . Faecal collection was carried out over the next few days, and the percentage of iron and chromium measured. The amount of iron absorbed was then calculated, assuming that both tracers were intimately mixed, and passed through the gut at the same rate. A similar method for measuring iron absorption with ^{131}Ba as the insert marker has been reported (Najean and Ardaillou, 1963; Boender and Verloop, 1969).

The blood clearance half-time of intravenously administered ^{59}Fe was then measured. ^{51}Cr -labelled red cells, given at the same time as the ^{59}Fe , enabled the red cell mass to be determined simultaneously and reduced the amount of blood taken—8 μ Ci of ^{59}Fe and 50 μ Ci of ^{51}Cr were used. Studies of the loss of iron from these patients were started two weeks later, after the completion of the utilization of the intravenous iron estimation, and were carried out by a method similar to that of Holt *et al.* (1967, 1968). Whole body counting was done at intervals of 14 and then 21 days up to a total of not less than 98 days. In seven of the patients all dialysis lines and membranes, dressings used at the time of attention to the shunts, and blood clots were carefully collected and incinerated; the radioactivity in the ash that had accumulated during the 21-day period was measured, two opposed NaI (T1) crystals in a shielded enclosure being used.

Haematological Studies

All patients were anaemic with haemoglobin levels ranging from 5.2 to 11 g/100 ml (Table I). P.C.V.s ranged from 18 to 33%. The red cell masses were low, ranging from 507 to 1,230 ml. Good correlation was seen between the red cell mass and the P.C.V. (Fig. 1) but was less satisfactory when compared on a volume per unit weight basis. The mean reticulocyte count was 1.1% (range 0.7 to 1.8%) though examination of most of the marrow smears showed active normoblastic erythropoiesis. Two marrows showed early megaloblastic changes though vitamin B₁₂ and serum and red cell folate were normal. Ring sideroblasts were not seen in the marrows in this series. The iron stores were assessed as zero in four patients, grade 1 in three, normal (grade 2 or 3) in four, and excessive (grade 4 or 6) in two (Table I). Patients were assessed as iron deficient with marrow iron of grade 0 or 1. Because of fluctuation in haemoglobin levels and P.C.V.s the

FIG. 1—Relation of P.C.V. to red cell mass.

mean of three to five estimations before and after treatment were taken (Table II). Marrow irons were graded before and after three months' treatment with ferrous sulphate 600 mg daily in six patients. Three iron-deficient patients showed a rise in the level of haemoglobin though one failed to show a rise in P.C.V. This change was not observed in the patients with normal marrow iron. None of the iron-deficient patients, however, showed any replenishing of their marrow iron stores.

Biochemical Studies

The serum iron, iron-binding capacity, and percentage saturation proved to be unsatisfactory guides to the state of the iron stores as indicated by the bone marrow studies. Low levels of saturation were seen in patients with adequate stores (Cases 8 and 10) and in one patient (Case 12) who had increased marrow iron. The reticulocyte uptake of ^{59}Fe showed better correlation with bone marrow grading and marrow iron granule counts. A previous study had shown normal levels of serum vitamin B₁₂ in these patients. In 11 patients the serum and red cell folate levels were normal at the time of this study.

Radioisotope Studies

ABSORPTION AND UTILIZATION

The results of the radioisotope studies are given in Table III. The retention of iron at 14 days, the most reliable indication of absorption, is represented in Fig. 2. Retention of iron in these patients has been compared with the retention seen in a control group of iron-deficient anaemic patients studied previously. Iron deficiency had been due to inadequate diet or chronic blood loss in the control group. Retention in this control group was 22 to 95% of the dose. Measurements of iron retention determined by whole body counting in normal subjects given ferrous salts have shown mean values of about 8% (Holt *et al.*, 1968).

TABLE II—Mean Estimations

Case No.	Mean Hb and P.C.V.				Marrow Iron			
	Pretreatment		Posttreatment		Pretreatment		Posttreatment	
	Hb	P.C.V.	Hb	P.C.V.	Grade	Granules %	Grade	Granules %
2	6.4	22	8	25	0	0.8	0	1.6
5	7.8	26	8.6	24	1	3.0	0	1.4
6	7.7	23	8.7	26	1	1.8	0	0.8
8	6.9	23	7.0	22	2	3.6	2	1.8
10	8.7	24	8.5	26	3	17.4	3	8.8
12	8.5	22	6.1	18	4	5.8	3	7.4

TABLE III—Radioisotope Studies

Case No.	Red Cell Mass (ml)	⁵⁹ Fe Absorption %*	⁵⁹ Fe Retention %*	Utilization of Oral Dose %†	Utilization of Intravenous Dose %*
1	658	23	28	71 ± 3	74
2	950	41	12	78 ± 7	74
3	1,230	48	43	69 ± 3	75
4	—	43	40	77 ± 4	—
5	507	59	65	66 ± 2	51
6	917	4	5	79 ± 16	69
7	880	30	26	67 ± 3	73
8	930	40	8	63 ± 9	70
9	825	13	16	45 ± 4	63
10	1,070	27	10	46 ± 7	84
11	—	28	10	64 ± 13	—
12	965	16	8	45 ± 8	65
13	735	29	8	19 ± 7	58

Standard deviations due to counting statistics: * < 1.0; † as indicated

The mean retention of iron in the patients on renal dialysis who were anaemic but not iron deficient was 11.3%. The mean retention of iron in the seven iron-deficient patients was 31.2%, and five out of seven showed levels of retention seen in the iron-deficient control subjects though the values tended to the lower part of the range. Retention of iron diminished as the marrow iron stores increased (Fig. 2).

FIG. 2—Retention of ⁵⁹Fe in iron-deficient control subjects and patients on regular dialysis.

The absorption of iron measured by the chromium chloride marker technique showed poor correlation with the retention values even in the iron-deficient patients where agreement had been good in previously reported studies. Reproducibility of results from successive daily samples was also poor. It is possible that there is poor mixing of the iron and chromium in these patients who take an unusual diet.

The utilization of the orally ingested iron for haemoglobin formation was greater than 45% in all patients except the iron-laden patient (Case 13). Though the utilization of ⁵⁹Fe was less than that sometimes seen in iron-deficient non-uraemic subjects, nevertheless 66 to 78% was available in the iron-deficient group for haemoglobin formation. The discrepancy between the utilization of the oral ⁵⁹Fe and intravenous ⁵⁹Fe was notable in Cases 5 and 6 where the latter was less well utilized. In four patients (Cases 9, 10, 12, and 13) the reverse was true. Since these studies were carried out at different times other factors such as menstruation or episodes of infection could have been responsible. In half the patients studied there was equally good utilization of the oral and intravenous iron.

STUDIES ON IRON LOSS

The mean daily loss of iron given as a percentage of the mean total body count was found to be in the range 0.02 to 1.3% per day, with a standard deviation due to counting statistics usually less than 0.01% per day. The loss of iron in control subjects using this method is 0.03% per day (Holt *et al.*, 1967). In 11 patients who completed these studies two (Cases 6 and 10) showed normal daily iron loss (0.02 to 0.04% per day), five (Cases 3, 7, 8, 12, and 13) showed intermediate losses (0.14 to 0.25% per day), and the other four (Cases 1, 2, 5, and 9) showed heavy iron loss (0.3 to 1.3% per day). This loss was not uniform over the period of the study (Fig. 3).

FIG. 3—Loss of iron on serial whole body counting in 11 patients on regular dialysis. Standard deviation due to counting statistics for each point was < 0.5% of administered dose. For a detailed discussion of the various errors of the measurement see Cottrill *et al.* (1968).

The steep fall in the activity in Cases 2 and 5 between the second and fifth week was related to clotting of the shunt and menorrhagia in Case 5 and a bleed into the dialyser in Case 2. The variation in the volume of blood lost into the membranes and lines in seven patients is shown in Table IV. The very big losses relate to accidents, but losses of between 5 and 20 ml were common and indicate how easily iron deficiency can develop insidiously in these patients.

TABLE IV—Blood Loss in Dialysis Membranes and Lines

Case No.	No. of Dialyses in Period	Volume of Blood left in Membranes and Lines per Dialysis (ml)		
		Min.*	Max.†	Mean
2	36	5.5	634	68
3	18	0.4	140	20
5	31	0.7	299	22
6	28	1.5	36	7.0
8	24	3.8	36	10
10	28	0.6	35	5.8
12	26	1.9	16	5.8

Standard deviation due to counting statistics: * 0.2 to 0.6; † 0.7 to 8.0

The results of the studies on the half-life of iron and chromium are similar to results previously reported. The ⁵⁹Fe half-life was in the range 39 to 156 minutes and that for ⁵¹Cr 15 to 24 days. Shortening of the red cell life is seen in some patients with chronic renal failure and may contribute to the anaemia in this condition. Patients on regular dialysis show this feature. In two patients (Cases 2 and 5) this apparent shortening was also related to the excessive blood loss occurring at the time.

Discussion

These studies show that patients on regular dialysis who are iron deficient will respond to oral iron therapy, and also that in these patients the state of the body's iron stores still influences absorption. The marrow iron stores are the most valuable guide to the need for treatment with iron and this confirms the observations of Edwards *et al.* (1970). The level of absorption in our iron-deficient patients did not approach those reported by Escbach *et al.* (1970). The two groups of patients may not be comparable, but their finding that iron absorption was entirely compatible with their patients' requirements was not confirmed in this study. The failure of the patients in the present study to replenish their marrow iron stores suggests that prolonged therapy may be required by those on regular renal dialysis.

Utilization of iron for haemoglobin formation occurred at the rate seen in the studies on intravenous iron by Carter *et al.* (1969), but there is no evidence that parenteral iron only was capable of being utilized. Parenteral iron is not more effective, and unless the requirement is carefully calculated and the result of treatment assessed from the marrow iron serious overload may develop.

The rate of blood loss varied considerably in these patients, and individuals could show pronounced variation in mean loss at dialysis in successive periods. The patient with the insidious loss of from 5 to 30 ml of blood per dialysis is most at risk as this may not be appreciated. It is in this group that prolonged oral iron therapy would be beneficial.

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Requests for reprints should be sent to Dr. J. S. Malpas.

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Effect of Measles Vaccination on Incidence of Measles in the Community

IAN SUTHERLAND, P. M. FAYERS

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Summary

A study of the effect of measles vaccination on the incidence of the disease in eight separate areas of England and Wales was begun in 1966. It showed an inverse association between the proportion of children vaccinated and the incidence of measles in the area in the following year, but measles epidemics occurred in several of the areas in subsequent years, despite continuing vaccinations.

Measles vaccination was introduced on a large scale in Britain in 1968. Analysis of the notification and vaccination statistics shows that the vaccination of about 10% of the child population (under 15 years) in 1968 sufficed to "replace" the measles epidemic which had been expected in the period October 1968 to September 1969 by a low incidence of the disease, typical of that in previous "inter-epidemic" years. Further, the effect of the vaccinations was to prevent the develop-

ment of natural measles in susceptible unvaccinated children as well as in the vaccinated subjects. Thus the number of immune subjects in the community was increased by the vaccinations, but as a result there was a reduction in the number of subjects who acquired immunity from natural measles. These opposed results can therefore explain why vaccination may be effective in the community for only a year or two, though vaccination protects the individual for much longer.

It is estimated that a continuing vaccination rate of 40 to 50% of the children born each year would be necessary to replace the regular biennial measles epidemics in Britain by a continuous endemic incidence, and might perhaps lead to the disappearance of the disease without a further major epidemic, but that a continuing vaccination rate of 80 to 90% of children born each year would then be necessary to prevent its reintroduction. The long-term control of measles by vaccination will thus probably prove more difficult than for any other infectious disease.

Introduction

At the instigation of the M.R.C. Measles Vaccines Committee a study was begun in 1966 of the effect of measles vaccina-

M.R.C. Statistical Research and Services Unit, London WC1E 6AS
IAN SUTHERLAND, M.A., D.PHIL., Director
P. M. FAYERS, B.Sc., Scientific Staff Member