Dietary iron overload in southern African rural blacks

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Summary

A survey conducted in rural southern African black subjects indicated that dietary iron overload remains a major health problem. A full blood count, erythrocyte sedimentation rate, serum concentrations of iron, total iron-binding capacity, ferritin, C-reactive protein (CRP), γ-glutamyltransferase (GGT) and serological screening for hepatitis B and human immunodeficiency virus (HIV) infections were carried out in 370 subjects (214 inpatients and 156 ambulatory Mozambican refugees). The fact that the geometric mean (SD range) serum ferritin concentration was much higher in the male hospital patients than in subjects living in the community [1 581 μg/l (421 - 5 944 μg/l) and 448 μg/l (103 - 1 945 μg/l) respectively] suggested that dietary iron overload was not the only factor raising the serum ferritin concentration. The major additional factor appeared to be inflammation, since the geometric mean (SD range) serum CRP was significantly higher in male hospital patients [21 mg/l (8 - 53 mg/l)] than in subjects in the community [3 mg/l (1 - 5 mg)]. Alcohol ingestion, as judged by history and by serum GGT concentrations, was also associated with significantly raised serum ferritin concentrations. This finding was ascribed to the fact that traditional brews are not only associated with alcohol-induced hepatic damage but are also a very rich source of highly bio-available iron. The role of iron overload in the genesis of the raised serum ferritin concentrations are confirmed in the diagnostic liver biopsy study. The majority of biopsies showed heavy siderosis, with varying degrees of hepatic damage. No subject tested positive for HIV antibodies, while the hepatitis B infection rate was high (± 70%) in both hospital and community subjects, with a surface antigen carrier rate of roughly 10%.

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Dietary iron overload resulting from the consumption of alcoholic beverages contaminated with iron has been described in various sub-Saharan populations.[1] Pathological associations with the condition include: portal fibrosis and cirrhosis of the liver;[2,3] glucose intolerance;[4] scurvy and osteoporosis;[5] and possibly oesophageal carcinoma.[6] There is also evidence to suggest that iron overload from any cause may predispose to infection[7] and there are data to show that hepatic amoebiasis may occur more frequently in blacks with iron overload.[8]

Both the prevalence and severity of iron overload have decreased markedly in urban black South African men over the past 25 years. This has been ascribed to a change in drinking habits, with Western liquors having replaced traditional beverages.[9] However, it has been shown recently that iron overload is still a major problem in men living in rural Zimbabwe.[10] The present study was undertaken to find out whether this is also the case in other rural areas in southern Africa.

Subjects and methods

A total of 370 subjects were studied. Over half (180 males and 34 females) were inpatients in Letaba, Elim and Tintswalo Hospitals in Gazankulu. A further 127 healthy male and 29 female Mozambican refugees living in the Gazankulu district were also evaluated. A brief medical history, including past and present alcohol consumption, was obtained using a simple questionnaire and the hospital-based patients were examined physically. A full blood count, erythrocyte sedimentation rate (ESR), serum iron (SI) value, total iron-binding capacity (TIBC), serum ferritin, C-reactive protein (CRP) and γ-glutamyltransferase (GGT) were measured in all subjects, and hepatitis B and human immunodeficiency virus (HIV) serology were also assessed. In an allied study 29 patients in the Raleigh Fitkin Memorial Hospital in Swaziland, who were being submitted to liver biopsy for medical diagnostic reasons, were similarly investigated. Half of each biopsy specimen was examined histologically, while the non-haem iron concentration of the other half was measured biochemically.

Biochemical measurements

Full blood counts were performed on either a Coulter M530 or a Coulter Model-S electronic counter with standard calibration. The ESR was determined by the Westergren method and the SI and TIBC by the methods recommended by the Iron Panel of the International Committee for Standardisation in Haematology.[11,12] Serum ferritin was determined by an enzyme-linked immunosorbent assay (ELISA),[13] the CRP by the method of Melamies[14] and the GGT by the method of Persijn and Van der Silk.[15] Hepatitis B serology was assessed using radio-immunoassay, while the serological test for HIV antibodies was done using an ELISA kit. Non-haem iron concentrations in liver biopsy specimens were measured by the method of Torrance and Bothwell.[16] White blood cell ascorbic acid concentration was determined according to the method of Denson and Bowers.[17]

The study was approved by the Committee for Research on Human Subjects of the University of the Witwatersrand. Subjects were admitted to the study only after informed written consent had been obtained.

Statistical considerations

Normality of distributions was assessed by the univariate procedure of the Statistical Analysis System (SAS) using an IBM 370 computer.[18] Data for ferritin, CRP and GGT concentrations were positively skewed, were normalised by logarithmic transformation and were expressed as geometric mean and SD ranges. Comparisons were by Student's *t*-test.[19] The level of significance was taken as 0,05. Correlation between the data were determined using Pearson correlation coefficients.

Results

Table I summarises the values obtained in each of the four groups studied. The mean ESR and CRP were significantly higher in the hospital-based group, reflecting a higher prevalence of infection and/or inflammation. The mean haemoglobin value was identical in hospital and community-based men (12,5 g/dl) and lower in both groups of women (10,7 g/dl and 11,1 g/dl, respectively). Geometric mean serum ferritin concentrations were significantly raised in men, both hospital based [1 581 μg/l (421 - 5 944 μg/l)] and community based [448 μg/l (103 - 1 945 μg/l)]. Concentrations were lower in women within each group. The lower TIBC in the hospital groups presumably reflects the fact that transferrin decreases in the acute phase response associated with inflammation.[20]

Alcohol had a profound effect on the serum ferritin values, with concentrations being significantly raised in both hospital- and community-based men who admitted to alcohol consumption (Table II). These comprised 72% and 80% of the study groups, respectively. The transferrin saturation level was also raised in the drinkers. The serum GGT concentration, which is a sensitive although relatively nonspecific marker of alcohol use,[21] suggested a similar association (Table III), since subjects

TABLE I. MEAN VALUES (± SD) FOR ESR, CRP, HAEMOGLOBIN, SERUM IRON, TOTAL IRON-BINDING CAPACITY, IRON SATURATION, SERUM FERRITIN AND GGT IN THE POPULATION SAMPLES STUDIED†

	Men		Women		Normal
	Hospital	Community	Hospital	Community	values
No. of subjects	180	127	34	29	—
Age (yrs)	56* ± 13	50 ± 14	54 ± 12	52 ± 17	—
ESR (mm/1st h)	51** ± 41	13 ± 20	58** ± 36	15 ± 15	< 15
CRP (mg/l)†	21** (8 - 53)	3 (1 - 5)	29** (13 - 69)	2 (1 - 3)	< 12
Hb (g/dl)	12,5 ± 3,2	12,5 ± 3,0	10,7 ± 2,3	11,1 ± 2,09	M > 13 F > 12
SI (μmol/l)	17,2 ± 9,7	23,3 ± 10,4	12,9** ± 8,4	23,2 ± 10,4	M 20,6 ± 5,6 F 20,6 ± 7,2
TIBC (μmol/l)	35,1** ± 10,0	51,4 ± 9,1	36,7** ± 10,8	50,4 ± 8,1	M 57,0 ± 6,5 F 62,1 ± 9,9
Saturation (%)	51 ± 26	46 ± 21	38 ± 25	48 ± 24	16 - 55
Ferritin (μg/l)†	1 581** (421 - 5 944)	448 (103 - 1 945)	486*** (105 - 2 252)	176 (23 - 1 340)	M < 300 F < 200
GGT (U/l)†	101 (16 - 164)	85 (18 - 136)	56*** (13 - 89)	35 (8 - 51)	< 60

*Hospital group significantly different from community group $P < 0,003$.
**Hospital group significantly different from community group $P < 0,0001$.
***Hospital group significantly different from community group $P < 0,03$.
†Serum CRP, ferritin and GGT values were positively skewed and results are therefore given as geometric mean values (SD ranges).

TABLE II. EFFECTS OF ALCOHOL CONSUMPTION ON MEAN (± SD) TRANSFERRIN SATURATION AND GEOMETRIC MEAN (SD RANGE) SERUM FERRITIN CONCENTRATION IN BLACK MEN

	Community-based		Hospital-based	
	Serum ferritin (μg/l)	Transferrin saturation (%)	Serum ferritin (μg/l)	Transferrin saturation (%)
No alcohol	145 (30 - 693)	36 ± 14	762 (205 - 2 829)	39 ± 25
Alcohol	584 (154 - 2 213)	48 ± 21	2 062 (600 - 7 088)	55 ± 26
***T* value**	4,2768	3,5087	4,6694	3,6505
***P* value**	0,0001	0,001	0,0001	0,0004

TABLE III. GEOMETRIC MEAN (SD RANGE) SERUM FERRITIN CONCENTRATIONS IN RELATION TO GGT LEVELS IN BLACK MEN

	GGT < U/l	GGT > 60 U/l	*P*	*F*
Hospital-based	1 204 (373 - 3 885)	2 360 (561 - 9 922)	0,0007	3,4509
Community-based	292 (66 - 1 292)	891 (281 - 2 828)	0,0001	4,3366

with GGT values below 60 U/l had significantly lower ferritin concentrations.

A feature of the study was the fact that serum ferritin concentrations were significantly higher in hospital-based men than in those subjects living in the community. This was true for both drinkers and non-drinkers. Differences in drinking habits did not seem to account for the findings, since GGT concentrations in black hospital-based and community-based men were not significantly different (Table I). A second possible reason for the observed differences was the presence of infection, inflammation or neoplasia. In this context it was noteworthy that hospital patients with raised CRP concentrations had geometric mean serum ferritin concentrations almost twice those found in patients with normal CRP levels [1 938 μg/l (553 - 6 794 μg/l) v. 1 206 μg/l (306 - 4 760 μg/l) ($t = 2{,}4091$; $P < 0{,}02$)] A similar analysis was not done on community residents, since all their serum CRP concentrations were within the normal range.

The evidence obtained in the community-based and hospital-based studies suggested that two factors were contributing to the high serum ferritin concentrations — excessive alcohol consumption and acute inflammatory diseases. However, it was still not clear how the alcohol was exerting its effects. In southern Africa a unique situation exists in which iron overload develops in subjects as a result of the ingestion over long periods of fermented beverages brewed in iron containers.[22,23] In such circumstances, the level of serum ferritin would be expected to reflect the degree of iron overload. However, alcohol can also raise the serum ferritin concentration by causing the release of ferritin from damaged liver cells.[24,25] In such circumstances serum ferritin concentrations may reach very high levels even in the absence of iron overload. The relative contributions of these two factors in the rural populations under study were addressed by carrying our measurements on a group of black patients who were undergoing percutaneous liver biopsies for diagnostic reasons. On questioning, 21 of the 29 patients admitted to the regular consumption of traditional fermented beverages. The geometric mean serum ferritin in the group was 2 650 μg/l (598 - 11 735 μg/l) and the mean transferrin saturation level was 62% ($\pm$ 30%). The high frequency of moderate-to-severe hepatic siderosis was confirmed by the findings on histological examination and chemical analysis (Table IV). Histological evidence of moderate-to-severe siderosis[2] was present in two-thirds of the patients and the geometric mean hepatic iron concentration ($\pm$ SD range) in this group was 128 μmol/g dry weight (32 - 519 μmol/g). In addition, 22 of the 29 patients showed evidence of significant portal fibrosis or cirrhosis. Of note was the fact that alcoholic hepatitis, which is associated with the excessive consumption of Western liquors and is rare in the siderosis associated with the drinking of iron-contaminated fermented beverages,[26] was only present in 2 patients. The degree of hepatic siderosis showed a positive correlation with the serum ferritin concentration ($r = 0{,}555$; $P = 0{,}0018$). The mean ($\pm$ SD) white cell ascorbic acid concentration was 15,8 $\pm$ 11,7 μg/10^8 leucocytes (normal range 20 - 40 μg/10^8 leucocytes) in the 9 subjects in whom it was measured.

As part of the overall survey, hepatitis and HIV status were also assessed. The percentage of subjects who were anti-HBc-, anti-HBS- and HBsAg-positive was 70,8%, 72,8% and 10,3%,

TABLE IV. BIOCHEMICAL AND HISTOLOGICAL FINDINGS IN 29 BLACK PATIENTS UNDERGOING DIAGNOSTIC LIVER BIOPSY

Age (yrs)	Sex	Serum ferritin (μg/l)	Transferrin saturation (%)	Serum GGT (U/l)	CRP (mg/l)	Hepatic non-haem iron (μmol/g dry weight)	Hepatic histology
40	M	9 424	87	34	47	818	Cirrhosis*
34	F	5 164	96	265	7	635	Cirrhosis*
55	F	1 417	—	52	12	612	Normal*
65	M	8 674	46	322	98	522	Fibrosis*
44	M	1 388	40	10	12	513	Fibrosis*
41	M	7 118	89	175	23	470	Fibrosis*
60	M	6 114	95	42	12	411	Fibrosis/cirrhosis*
72	M	6 509	36	174	40	407	Normal*
45	M	3 260	98	41	10	389	Cirrhosis*
70	F	2 960	56	36	95	346	Fibrosis*
50	M	5 670	97	63	8	316	Fibrosis*
45	M	4 081	89	58	5	305	Cirrhosis*
50	M	3 688	95	236	12	301	Cirrhosis*
30	M	4 164	93	6	12	215	Cirrhosis*
33	M	2 490	65	375	10	176	Cirrhosis*
49	M	3 272	44	174	63	156	Fibrosis*
50	F	37 566	61	217	5	146	Abscess/fibrosis*
61	M	5 820	63	148	43	99	Tuberculosis
59	M	13 862	33	117	76	72	Abscess*
40	M	5 870	73	496	12	65	Hepatitis/cirrhosis
38	M	1 121	45	221	12	60	Hepatitis
70	F	2 181	83	140	12	46	Hepatoma/cirrhosis*
60	M	3 624	92	229	18	36	Hepatoma
66	M	4 195	65	292	52	32	Cirrhosis*
34	F	48	7	80	11	28	Extramedullary erythropoiesis
68	M	249	20	472	85	16	Hepatoma
—	M	1 720	23	238	95	14	Hepatoma
17	M	35	10	631	5	14	Hepatoma/cirrhosis
70	M	743	21	190	62	5	Hepatoma

*Denotes grade III or IV iron overload on histological examination.
'Fibrosis' implies significant portal fibrosis.[37]

respectively. The corresponding figures in the community-based subjects were 70,5%, 65,5% and 11,5%. There was no correlation between positivity and the size of iron stores. All patients tested negative for HIV antibodies.

Discussion

Body iron homeostasis is maintained by the ability of the upper gastro-intestinal mucosa to regulate iron absorption according to the body's needs. Thus oral iron overload can occur when mucosal control is genetically altered (e.g. idiopathic haemochromatosis) or when there are large amounts of soluble iron in the diet.[1] It is this latter mechanism which accounts for the iron overload that occurs in southern African blacks.

The condition was first recognised pathologically in the 1920s and was subsequently investigated intensively over the next 40 years. The source of excess iron was shown to be the traditional beer, made from maize and sorghum, which was brewed in iron pots and drums.[22,27] During the fermentation process the iron was leached from the containers and entered the brew. This iron was highly bio-available because of its low pH, its ethanol and lactate contents, and its low content of solids.[28] As a result, iron overload of varying degrees of severity was present in the majority of middle-aged black men and was responsible for a variety of disease states.[29]

However, with amendments to the Liquor Act in South Africa in 1961 and 1963, there was a marked change in the drinking habits of the black urban population and a study done in 1976 indicated that there had been a marked reduction in the prevalence and severity of this condition in urban males, with virtually no evidence that there had been any further accumulation of iron between 1959 and 1976.[9] Because of these findings, interest in the condition waned, and iron overload in black men is now a rare necropsy finding in Johannesburg.

The present study was prompted by the recent finding by Gordeuk *et al.*[10] that iron overload remains prevalent in rural Zimbabwe, where traditional drinking habits persist. Our results, obtained in several rural black populations, support Gordeuk's assertion that iron overload remains a major problem in rural sub-Saharan Africa. The assertion is further supported by the findings in 29 consecutive liver biopsies carried out for medical diagnostic purposes in patients attending a rural hospital in Swaziland. A high frequency of severe iron overload was observed, with cirrhosis a common association. Direct questioning plus the results of serum GGT estimations indicated that the iron overload in rural communities continues to result from the consumption of iron-contaminated fermented alcoholic beverages.

Most of the previous work on iron overload in black subjects living in sub-Saharan Africa was done before the widespread application of serum ferritin level estimation as a means of assessing the size of body iron stores.[30] The present findings underscore both its strengths and weaknesses in this regard. While there is no doubt that it identified the high prevalence of iron overload in the groups under study, it was apparent that the actual concentrations in individual subjects were markedly influenced by other factors. These include liver damage due to alcohol abuse[3,25] and infection, inflammation and neoplasia[31,32] — all of which are associated with an inappropriately raised serum ferritin concentration, and ascorbic acid deficiency, which lowers serum ferritin levels in relation to body iron.[33] For example, although drinking habits, as judged by GGT concentrations, were similar in the community- and hospital-based men, serum ferritin concentrations in hospital patients were nearly 4 times higher. This difference can be explained, in part at least, by the hyperferritinaemic effect of inflammation, infection and neoplasia, since there was a significant association between a raised CRP, which is a sensitive marker of the non-immune inflammatory response,[31] and increased concentrations of serum ferritin in the hospital-based group. Those patients with a raised CRP had a mean serum ferritin concentration that was almost twice that found in patients without the biological marker of inflammation. At the same time, it should be noted that the geometric mean ferritin concentration of 608 $\mu g/l$ was also raised in those patients who had normal CRP concentrations. It was noteworthy that most of the subjects submitted to liver biopsy gave a history of a significant alcohol intake and had raised serum concentrations of both GGT and CRP.

Interpretation of the quantitative significance of the serum ferritin concentration in black subjects with iron overload is further bedevilled by the fact that many of these people are deficient in ascorbic acid. It has been shown that the iron deposits lead to increased catabolism of ascorbic acid[34] and the resultant ascorbic acid deficiency is associated with both a reduced release of iron into the circulation from reticulo-endothelial cells[35,36] and inappropriately low serum ferritin concentrations.[37]

The present findings, when taken in conjunction with those of Gordeuk *et al.*,[10] indicate that dietary iron overload remains a problem in sub-Saharan Africa, affecting a significant proportion of the rural population. As was mentioned earlier, this has considerable pathological implications, since the condition has previously been associated with diseases such as cirrhosis, diabetes, scurvy and osteoporosis. There may, however, be other equally important but less well-recognised effects. In this context, observations that other forms of iron overload may predispose to certain infections could have major implications in rural communities where the prevalence of infective disease is high. Finally, the results indicate the urgent necessity of preventing the further development of iron overload in rural black communities. This could be achieved by simple public health campaigns directed at the replacement of iron containers with earthenware pots in the production of traditional fermented beverages.

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