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Original Article

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Oral Iron Treatment Response and Predictors in Anaemic Adolescents and Adults with IBD: A Prospective Controlled Open-Label Trial

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Abstract

Background: Because of previous concerns about the efficacy and safety of oral iron for treating iron deficiency anaemia in inflammatory bowel disease [IBD], particularly in young people, we compared the effects of ferrous sulphate on haemoglobin response, disease activity and psychometric scores in adolescents and adults with IBD. We also assessed the relation of baseline serum hepcidin to haemoglobin response.

Methods: We undertook a prospective, open-label, 6-week non-inferiority trial of the effects of ferrous sulphate 200 mg twice daily on haemoglobin, iron status, hepcidin, disease activity (Harvey–Bradshaw Index, Simple Colitis Clinical Activity Index, C-reactive protein [CRP]), faecal calprotectin and psychometric scores in 45 adolescents [age 13–18 years] and 43 adults [>18 years].

Results: On intention-to-treat analysis, ferrous sulphate produced similar rises in haemoglobin in adolescents {before treatment 10.3 g/dl [0.18] (mean [SEM]), after 11.7 [0.23]: $p < 0.0001$} and adults (10.9 g/dl [0.14], 11.9 [0.19]: $p < 0.0001$); transferrin saturation, ferritin [in adolescents] and hepcidin [in adults] also increased significantly. On per-protocol univariate analysis, the haemoglobin response was inversely related to baseline haemoglobin, CRP and hepcidin. Oral iron did not alter disease activity; it improved Short IBDQ and Perceived Stress Questionnaire scores in adults.

Conclusion: Oral ferrous sulphate was no less effective or well-tolerated in adolescents than adults, and did not increase disease activity in this short-term study. The inverse relation between baseline CRP and hepcidin levels and the haemoglobin response suggests that CRP or hepcidin measurements could influence decisions on whether iron should be given orally or intravenously. *[ClinTrials.gov registration number NCT01991314]*

Key Words: inflammatory bowel disease; iron deficiency anaemia; hepcidin

1. Introduction

Iron deficiency anaemia [IDA] is a frequent complication of inflammatory bowel disease [IBD].[1,2] In children and adolescents IDA appears to be more common than in adults and is often undertreated,[1,3,4] perhaps reflecting paediatricians' concerns about possible side effects, including worsening of disease activity, and about young people's medication adherence.[1]

Quality of life [QOL] correlates negatively with the severity of anaemia in patients with IBD.[5] Prospective studies of oral and intravenous iron supplementation in adults with IBD have shown improvements in QOL when the haemoglobin [Hb] is corrected[6–10] but this effect has not been assessed in young people with IBD. Psychological distress and fatigue are common in people of all ages with IBD[11–13] but to our knowledge there are no prospective studies of the effects of oral iron supplementation on these factors in people with IBD.

It is widely stated that the Hb response to oral iron is reduced in patients with active IBD: this has been confirmed in one[14] but not all studies.[6,15] Such an effect could be explained by the involvement of hepcidin, which regulates iron homeostasis by inhibiting its uptake by enterocytes, macrophages and hepatocytes.[16] Serum hepcidin levels are increased by pro-inflammatory cytokines; conversely, in iron deficiency, in the absence of inflammation, hepcidin levels fall. Serum hepcidin concentrations at baseline are related inversely to the Hb response to oral iron in patients with rheumatoid arthritis and other diseases,[17] but whether this is true in IBD is unknown.

We therefore undertook a prospective phase IV, open-label, parallel group, 6-week non-inferiority clinical trial using oral ferrous sulphate to assess the hypotheses that: [1] there is no difference in the Hb response to oral iron treatment of IDA in adolescent compared to adult IBD patients; [2] oral iron does not worsen disease activity in IBD; [3] response to oral iron is inversely related to serum hepcidin concentrations at baseline; and [4] treatment of anaemia improves QOL, mood and fatigue in adolescent and adult patients with IBD.

2. Materials and Methods

2.1. Patients

Patients aged 13–18 years were defined as adolescents, and those aged ≥ 19 years as adults. Between January 2012 and April 2015, patients with IBD (ulcerative colitis [UC], Crohn's disease or IBD unclassified [IBDU]) diagnosed by standard clinical, radiological and pathological criteria and who within the next month were due to attend the adult, adolescent transition and paediatric IBD clinics at Barts and the Royal London Hospitals, or the paediatric IBD clinic at Chelsea and Westminster NHS Trust, London, UK, were screened for the result of their Hb concentration at their previous clinic attendance. Those found to be anaemic [see Figure 1] were sent a letter of explanation and invited to participate in the trial. They were telephoned 1–2 weeks after the letter and asked about current and previous iron therapy: those apparently eligible [see below] and verbally consenting to participate were then seen by a trial doctor or research nurse either at their next clinic appointment, or on another mutually convenient occasion. Informed written consent from the patients or their parents, as appropriate, was obtained at this meeting, and patients who remained eligible on the basis of their iron therapy history and clinical and haematological criteria were enrolled [Figure 1].

2.1.1. Definition of iron deficiency anaemia

Anaemia was defined by age and sex-adjusted World Health Organisation criteria [males <13.0 g/dl; females and adolescents aged <15 years <12.0 g/dl].[18] For inclusion in the trial, patients had to be both anaemic and have iron deficiency as defined by transferrin saturation <18%. They also had to report either tolerance of previous course[s] of oral iron, or to be naïve to this treatment.

2.1.2. Exclusion criteria

Patients were excluded if they did not meet the haematological inclusion criteria on the admission-to-study blood test ['screening failures'], if they had been given oral or intravenous iron within 3 months, or if they had previously been intolerant of oral iron [Figure 1]. Other exclusion criteria were age <13 years, vitamin B12 or folate deficiency, anaemia caused by drugs used to treat IBD, haemoglobinopathy, presence of stoma or ileoanal pouch, severely active IBD requiring hospital admission, severe cardiopulmonary, hepatic, renal or other disease, pregnancy, breast-feeding, use of cholestyramine, and inability to speak English well enough to complete the consent form or psychometric questionnaires [Figure 1].

2.1.3. Regulatory considerations

The trial was approved by the Southampton National Research Ethics Committee [number 10/H504/900] and was registered as a Clinical Trial of an Investigational Medical Product [CTIMP] with the Medicines and Healthcare Products Regulatory Agency [number 14620/0035/001-0001, EUDRACT number 2010-023797-39] and ClinTrials.Gov [NCT01991314]. The trial was sponsored by Barts Health NHS Trust.

2.2. Trial protocol and treatment

At enrolment, demographic data [Table 1] were recorded. Disease type, location, behaviour and extent, using Montreal classifications,[19] were noted from medical records. The Charlson index was used to score comorbidities.[20] Before and after treatment, patients completed four psychometric questionnaires to assess mood, fatigue and QOL: the Hospital Anxiety and Depression Scale [HADS-A and HADS-D],[21] the general Perceived Stress Questionnaires [PSQ-G],[22] the Multi-dimension Fatigue Inventory [MFI][23] and the Short Inflammatory Bowel Disease Questionnaire [SIBDQ].[24] Symptomatic disease activity was assessed using the Harvey–Bradshaw Index [HBI],[25] and Simple Clinical Colitis Activity Index [SCCAI].[26] Blood was collected for full blood count, including Hb, iron studies and hepcidin, and C-reactive protein [CRP]; and a stool sample was obtained for faecal calprotectin. Patients were given 6 weeks of treatment with 200 mg oral ferrous sulphate [Wockhardt Ltd, Ranbaxy Ireland Ltd] twice daily.

After 1 week, patients were telephoned to assess tolerance to treatment: those intolerant of or non-adherent to oral iron were withdrawn from the trial and asked to attend for repeat blood tests, questionnaires and faecal calprotectin as end of trial measures. After 6 weeks of iron treatment, the above measurements were repeated and adherence was assessed by counting of returned iron tablets.

2.2.1. Outcome measures

The primary outcome measure was mean increase in Hb concentration in the adolescent and adult groups after 6 weeks of treatment. Secondary outcome measures were tolerance of oral iron, changes in disease activity [HBI, SCCAI, CRP, faecal calprotectin], psychometric scores, and relation of serum hepcidin at baseline to Hb response to oral iron.

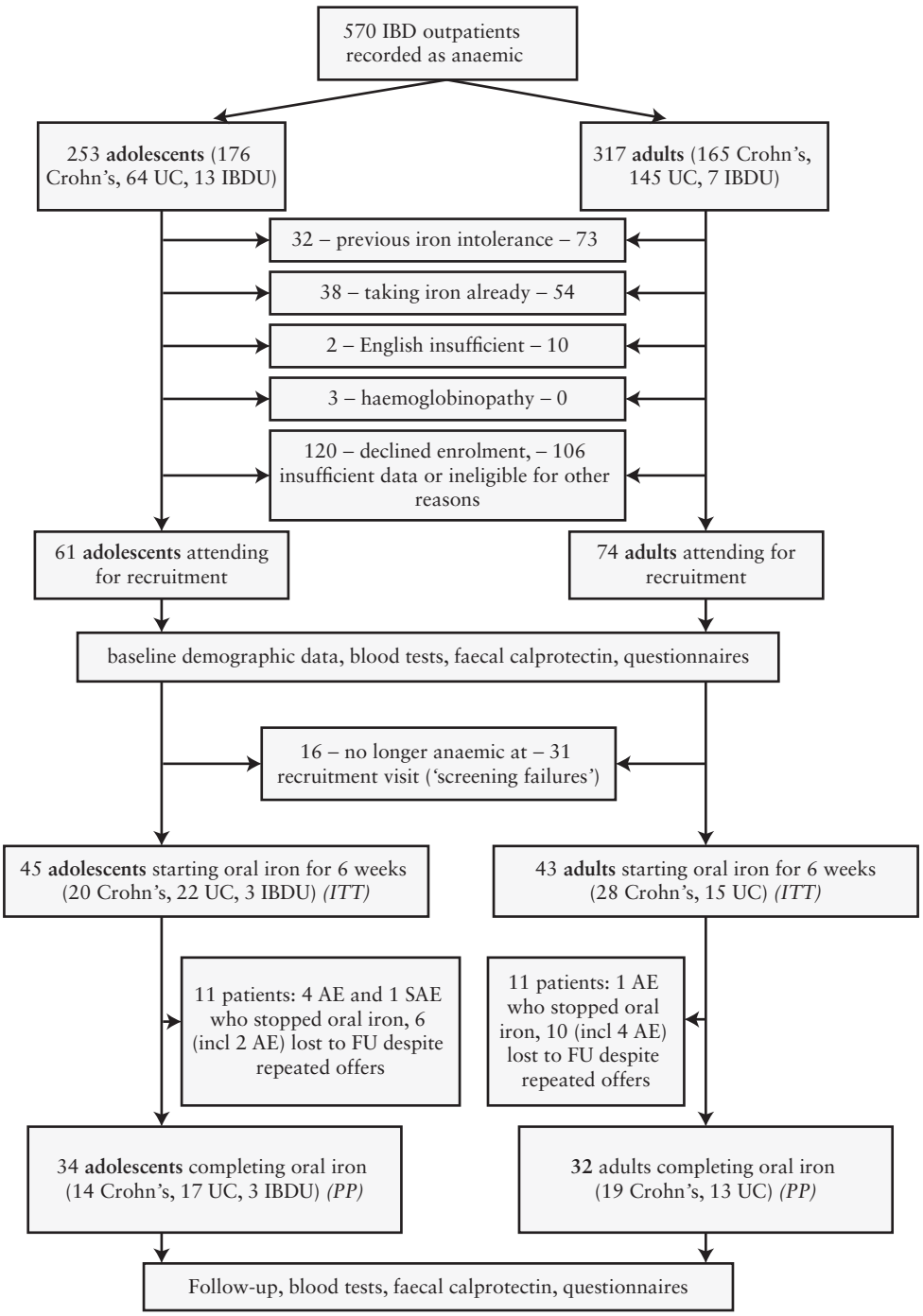

Figure 1. CONSORT-style diagram of trial recruitment. UC, ulcerative colitis; IBDU, IBD unclassified; AE, adverse event; FU, follow-up; ITT, intention to treat; PP, per protocol.

2.3. Assays

Hb, iron studies and biochemical tests were undertaken in the routine laboratories at the Royal London and Chelsea and Westminster Hospitals. Serum for hepcidin assays was stored at −80 °C until assay in duplicate in the University Birmingham by mass spectrometry.[27] Stable isotope-labelled synthetic hepcidin was used as an internal standard for each assay but the mass spectrometer, being a research instrument, was not serviced annually and, as such, did not meet strict Good Clinical Practice [GCP] compliance regulations. Stool samples were stored at −80 °C until the end of recruitment: they were then quantified for calprotectin in duplicate by enzyme-linked immunosorbent assay [ACCUSAY Calprotectin, Launch Diagnostics] in the Clinical Immunology laboratory at Barts Health NHS Trust.

2.4. Statistics

2.4.1. Sample size calculation

We calculated, on the premise that there is non-inferiority between adolescent and adult groups in the difference in increase of mean Hb levels of 0.35 g/dl, with estimated standard deviation 0.7 g/dl[6] following treatment, that 45 patients in each group would be required, derived using

Table 1. Demographic, clinical and laboratory characteristics and psychometric scores of the recruited adolescent and adult patient groups at baseline [ITT groups]. Means ± SEM are shown unless otherwise stated.

Characteristic		Adolescents [*n* = 45]	Adults [*n* = 43]	*p*-value
Sex	Male	23 [51%]	20 [47%]	0.68
Age	Age [years]	14.9 [0.25]	32.5 [1.74]	**<0.0001**
Ethnicity	White Caucasian	24 [53%]	22 [51%]	0.59
	Asian	17 [38%]	13 [30%]	
	Afro-Caribbean	3 [7%]	6 [14%]	
	Other	1 [2%]	2 [5%]	
Smoking	Current	1 [2%]	4 [9%]	0.054
	Ex	1 [2%]	5 [12%]	
	Never	43 [96%]	34 [79%]	
Education	Degree	0 [0%]	22 [51%]	**<0.0001**
	A-level and equivalent	8 [18%]	8 [19%]	
	GCSE and equivalent	30 [67%]	8 [19%]	
	None	7 [16%]	5 [12%]	
Marital status	Married or partner	0 [0%]	18 [42%]	**<0.0001**
Employment	Unemployed	0 [0%]	7 [16%]	**<0.0001**
	Retired	0 [0%]	3 [7%]	
	Working	1 [2%]	23 [54%]	
	Full-time education	44 [98%]	10 [23%]	
Comorbidity	Charlson score (median [range])	0 [0–1]	0 [0–1]	0.50
Nutritional status	Weight [kg]	54.6 [1.76]	67.4 [2.30]	**<0.0001**
	BMI	21.0 [0.55]	23.7 [0.83]	**0.008**
Disease type	Crohn's disease	20 [44%]	28 [65%]	**0.03**
	Ulcerative colitis	22 [49%]	15 [35%]	
	IBDU	3 [7%]	0 [0%]	
Duration	Age at diagnosis [years]	12.2 [0.4]	22.4 [1.4]	**<0.0001**
	Disease duration [years]	2.7 [0.38]	9.5 [1.3]	**<0.0001**
Crohn's	A1: Age <17 years	20 [100%]	9 [32%]	**<0.0001**
disease	A2: 17–40 years	–	17 [61%	
[Montreal	A3: >40 years	–	2 [7%]	
Classification]	L1: Ileal	5 [25%]	5 [18%]	0.38
	L2: Colonic	2 [10%]	7 [25%]	
	L3: Ileocolonic	13 [65%]	16 [57%]	
	+L4: Upper GI	7 [35%]	2 [7%]	**0.024**
	B1: Inflammatory	16 [80%]	7 [25%]	**<0.0001**
	B2: Stricturing	3 [15%]	8 [29%]	
	B3: Penetrating	1 [5%]	13 [46%]	
	+ p: Perianal	0 [0%]	11 [39%]	**0.001**
Disease activity	Harvey Bradshaw Index	2 [0.4]	3 [0.5]	**0.04**
UC	E1: Proctitis	3 [14%]	2 [13%]	0.64
[Montreal	E2: Left	7 [32%]	7 [47%]	
Classification]	E3: Total	12 [55%]	6 [40%]	
Disease activity	Simple Clinical Colitis Activity Index	2 [0.4]	3 [0.5]	0.11
Medications	5 ASA	32 [71%]	17 [40%]	**0.005**
	Prednisolone/budesonide	7 [16%]	7 [16%]	0.93
	Enteral nutrition	3 [7%]	0 [0%]	024
	Thiopurine	23 [51%]	20 [47%]	0.67
	Methotrexate, ciclosporine	2 [4%]	0 [0%]	0.50
	Anti-TNF	2 [4%]	3 [7%]	0.67
	Antidepressants	0 [0%]	3 [7%]	0.11
Oral iron treatment	Tolerant of previous course[s]	18 [40%]	31 [72%]	**0.005**
	Naïve	25 [56%]	12 [28%]	
	Uncertain of history	2 [4%]	0 [0%]	
Inflammatory markers	Serum CRP [mg/l]	9.9 [3.4]	11.4 [2.9]	0.75
	Faecal calprotectin [µg/g]	295 [48]	299 [73]	0.96
Psychometric scores	SIBDQ	51 [1.9]	44 [2.3]	**0.02**
	HADS-A	7.3 [0.8]	8.1 [0.7]	0.45
	HADS-D	4.4 [0.6]	6.0 [0.6]	0.06
	PSQ-G	57 [2.3]	71 [2.4]	**<0.0001**
	MFI	58 [1.1]	60 [1.1]	0.28

Bold denotes significant *p*-value. Abbreviations: A-level, Advanced-level; GCSE, General Certificate of Secondary Education; BMI, basal metabolic index; IBDU, inflammatory bowel disease of uncertain type; 5ASA, 5-aminosalicylic acid; CRP, C-reactive protein; SIBDQ, Simple IBD Questionnaire; HADS-A and -D, Hospital Anxiety and Depression Scale for Anxiety (-A) and Depression (-D); PSQ-G, Perceived Stress Questionnaire-General; MFI, Multidimension Fatigue Inventory.

80% power, one-sided significance level of 0.05 and *R* 0.5 for the covariate; this calculation took into account the planned analysis of covariance [ANCOVA] methodology [see below][28] and the 20% patients who might be lost to follow up or withdraw from the trial.

2.4.2. Baseline demographic data

Differences between categorical variables were sought using chi-squared analysis or Fisher's exact test, and between continuous variables using Student's *t*-test. Regression analyses assessed associations between clinical, laboratory and psychometric data at baseline.

2.4.3. Primary endpoint efficacy analysis

To test the hypothesis of non-inferiority with maximal statistical power, ANCOVA was used to compare the change in mean haemoglobin levels between adults and adolescents after accounting for necessary covariates.[28] 95% confidence intervals were established for the treatment effects to determine the status of the primary hypothesis. Using the chi-squared test, we also compared the proportions of patients in each group in whom ferrous sulphate produced a normalisation of haemoglobin concentration by WHO criteria.[18]

The primary outcome measure [Hb response in each patient group] and tolerance to oral iron were assessed on both intention-to-treat [ITT] and per-protocol [PP] bases, while the effects of oral iron on the other variables measured were assessed only in those patients completing treatment [PP].

2.4.4. Secondary endpoint analysis

Iron-induced changes in symptom scores, laboratory measures and psychometric scores were compared between and within groups with the Mann–Whitney U test and Wilcoxon signed rank test or unpaired and paired Student's *t*-tests, as appropriate. The psychometric outcomes in responders and non-responders were compared by Student's *t*-test. Relations between baseline Hb and psychometric scores were assessed by linear regression.

2.4.5. Identification of factors related to the Hb response to iron therapy

Univariate linear regression analysis was conducted to identify the factors, including hepcidin, that might predict response to oral iron. Multivariate forwards regression analysis was used to confirm predictive factors.

2.4.6. Baseline demographics and safety analysis

Demographic factors and adverse effects of iron therapy in each group were compared by chi-squared test or Fisher's exact test, as appropriate.

Two tailed p values <0.05 [apart from the ANCOVA analysis, where one-tailed values were used] were considered significant. Results are expressed as mean [SEM] unless otherwise stated. The last observation carried forward [LOCF] and Last observation carried backward [LOCB] methods were used for data points missing at the follow-up or baseline assessments, respectively. Statistical Package for the Social Sciences [SPSS] [version 16] was used for the statistical analysis.

3. Results

3.1. Baseline data

Forty-five adolescents and 43 adults were recruited to the trial and included for the ITT analysis [Figure 1]. Their baseline demographics are shown in Table 1. The adolescent patients had a slightly lower Hb [p = 0.01] and HBI [p = 0.04] than the adults [Tables 1 and 2]. Adults had poorer QOL as assessed by the short IBDQ [SIBDQ] and perceived stress scores [PSQ-G] at baseline than adolescents.

When both groups were analysed together, baseline Hb was directly related to baseline transferrin saturation [R = +0.53, p < 0.0001], serum ferritin [R = +0.22, p = 0.044] and age [R = +0.30, p = 0.005], but showed no significant correlations with gender, HBI, SCCAI, CRP, faecal calprotectin [FCP], disease type or any of the psychometric scores. Otherwise, transferrin saturation at baseline correlated only with age [R = +0.21, p = 0.046]. Serum hepcidin at baseline was directly related to CRP [R = +0.79, p < 0.0001], FCP [R = +0.45, p < 0.0001] and SCCAI [R = +0.41, p = 0.01], but not to Hb, transferrin saturation, ferritin or HBI. Serum CRP was related to FCP [R = +0.43, p = <0.0001], SIBDQ [R = -0.25, p = 0.02] and the perceived stress score [PSQ-G] [R = +0.26, p = 0.02].

QOL at baseline as assessed by the SIBDQ also showed significant correlations with disease symptoms (HBI [R = –0.38, p = 0.01], SCCAI [R = –0.62, p < 0.0001]) and mood scores (HADS-A [anxiety] [R = –0.38, p < 0.0001], HADS-D [depression] [R = –0.41, p < 0.0001], PSQ-G [R = –0.55, p < 0.0001]). HADS-A correlated with HADS-D [R = +0.62, p < 0.0001] and PSQ-G [R = +0.46, p < 0.0001], while the latter correlated with HADS-D [R = +0.44, p < 0.0001] and SCCAI [R = +0.38, p = 0.02].

3.2. Response to oral iron therapy

3.2.1. Haematological response to oral iron therapy [ITT analysis]

Using ANCOVA, there were no differences in the Hb responses between the two patient groups: adolescents mean change +1.22 g/dl [0.21], 95% confidence interval [CI] 0.79–1.64; adults +1.30 [0.27], 95% CI 0.84–1.75, p = 0.80, with the covariates appearing in the model being evaluated at the following baseline values: Hb 10.55 g/dl, CRP 10.9 mg/l, transferrin saturation 7.3% and hepcidin 30.1 ng/ml.

On ITT analysis of the raw data, ferrous sulphate produced similar small but statistically significant rises in serum Hb in both patient groups [Table 2]. The proportion of patients whose Hb concentrations were normalised by WHO criteria[18] were 13/45 [29%] in the adolescent group, and 16/43 [37%] in the adult group. Percentage transferrin saturation also rose in both groups after oral iron [Table 2].

3.2.2. Haematological response to oral iron therapy [PP analysis]

Demographic data of the 66 patients completing the trial on a PP basis were similar to those of the ITT population [results not shown]. In these patients, oral iron for 6 weeks produced small but statistically significant rises in Hb, percentage transferrin saturation, ferritin [adolescents only] and serum hepcidin [adults only] [Table 3]. Again, there were no statistically significant differences in Hb, transferrin saturation, ferritin or hepcidin responses between the two age groups. The proportions of patients whose Hb concentrations were normalised by oral iron were 13/34 [38%] in the adolescent group and 16/32 [50%] in the adult group.

In a post-hoc analysis, we compared the Hb responses to iron in patients classified as having either 'pure' IDA [arbitrarily defined by CRP <5 mg/l and/or FCP < 100 µg/g] or combined IDA/anaemia of chronic disease [ACD] [CRP ≥5 mg/l and/or FCP ≥100 µg/g] [see Supplementary Material Tables A and B]. While there was a trend for the Hb responses to be smaller in the combined IDA/ACD than

Table 2. Haematological results of the 88 recruited adolescent and adult patient groups before and after 6 weeks of oral iron [ITT analysis]. Results are shown as mean [SEM].

Variable	Adolescents [n = 45] Before after change	p for difference within adolescent group	Adults [n = 43] Before after change	p value for difference within adult group	p value between groups at baseline	p between groups for change in variable
Hb [g/dl]	10.3 [0.18]		10.9 [0.14]			
	11.7 [0.23]		11.9 [0.19]			
	+1.4 [0.24]	**<0.0001**	+1.0 [0.23]	**<0.0001**	**0.01**	0.23
Transferrin saturation [%]	7.0 [0.5]		7.8 [0.6]			
	17.6 [2.2]		18.6 [2.3]			
	+10.5 [2.3]	**<0.0001**	+10.7 [2.2]	**<0.0001**	0.32	0.98
Ferritin [µg/l]	14.2 [2.9]		22.9 [8.9]			
	24.5 [3.3]		99.5 [59]			
	+10.3 [2.0]	**<0.0001**	+76.6 [60.0]	0.21	0.36	0.27
Serum hepcidin [ng/ml]	31.3 [3.9]		28.7 [2.9]			
	36.7 [4.3]		35.3 [3.8]			
	+4.8 [2.4]	0.056	+6.6 [2.6]	**0.013**	0.61	0.60

Bold denotes significant p-value.

in the pure IDA groups, these differences did not reach statistical significance.

3.2.3. Relation between Hb response to oral iron and baseline variables [PP analysis]

Univariate analysis of both groups showed that the rise in Hb in response to oral iron was significantly negatively correlated with baseline Hb itself, transferrin saturation, hepcidin concentration and CRP [Table 4]; it was unrelated to age, gender, ethnicity, BMI, Charlson index, disease type or location, HBI, SCCAI, FCP or ferritin [data not shown]. However, multivariate regression analysis showed that only baseline Hb and CRP were significant predictors of the Hb response [Table 5]. Reworking the multivariate analysis to include age group [adolescent/adult] as a cofactor made no overall difference to the model [see Supplementary Material Table C].

3.2.4. Effect of oral iron on disease activity and psychometric scores [PP analysis]

Oral iron had no significant effect on disease activity in either patient group [Table 3]. In the adult group, but not the adolescents, oral iron produced significant improvements of both the SIBDQ and the PSQ-G scores. When all patients were analysed together oral iron was associated with small but statistically significant improvements in SIBDQ [+4.4 units, p = 0.009] and HADS-A [–0.9 units, p = 0.042]. There were no differences in psychometric responses between patients whose Hb normalised after oral iron [n = 29] and the remainder [n = 37] [data not shown].

3.2.5. Adherence to treatment in PP study

Twenty-five of 34 adolescents and 19/32 adults made tablet returns. In the adolescent group returning their packets, the median number of tablets returned was 0 [range 0–28]; in the adults, these figures were 9 [0–57]. In the three patients [all adults] returning more than half the tablets they were issued with, all showed rises in their serum Hb levels [+0.5 to +1.3 g/dl] at the end of the study.

3.2.6. Tolerance of oral iron therapy: adverse and serious adverse events

In the 88 patients entering the study, there were 15 adverse events [AEs] and three serious adverse events [SAEs] [Table 6]. Overall, there was no difference in tolerance of ferrous sulphate during the trial between the two groups, with 13% [6/45] of adolescents and 12% [5/43] of adults stopping treatment because of AEs. AEs occurred in 16% [6/37] patients naïve to oral iron. Eight AEs and all three SAEs [Table 6] occurred in the 49 patients reporting that they had tolerated previous courses of oral iron.

Six of the 15 patients with AEs [Table 6] failed to return for follow-up and provide blood or stool for repeat FCP. In the patients with AEs who provided follow-up samples, there were no significant changes in inflammatory markers [results not shown].

Of the three protocol-adherent patients with SAEs, CRP was 60 ± 41 mg/l before, and 111 ± 43 after the SAE [n = 3]; FCP, available in only two patients, was 552 and <12.5 µg/g before, and 403 and 1341 µg/g afterwards, respectively.

4. Discussion

Most of the baseline demographic and phenotypic differences between the adolescents and adults shown in Table 1 are a consequence of their age difference. The adolescents were slightly more anaemic than the adults at baseline, perhaps because of a reluctance of paediatricians to use oral iron.[1] Our patients' serum hepcidin concentrations at baseline were directly related to IBD activity: the variability of results previously reported[29–33] is probably due to different methods of assaying hepcidin and IBD activity, differences between the IBD populations studied, and the contrasting effects on hepcidin levels of active inflammation and iron deficiency.

The prevalence of anxiety, depression and fatigue in IBD, particularly when active, is substantially increased in patients with IBD.[11–13] As before,[34] we found a tendency for mean scores for anxiety to exceed those for depression, and strong associations between SIBDQ, HADS and perceived stress scores. The lack of significant associations between the psychometric scores and baseline Hb could result from our recruitment only of patients with IDA and consequently a narrow spread of Hb. Like others, we found high mean fatigue scores in both groups of patients.[12,35,36] The absence of any association between MFI and IBD activity contrasts with previous reports,[35,37] but could result from the fact that all our patients were anaemic.

The increases in Hb concentration and transferrin saturation after 6 weeks of oral iron in the adolescents were not inferior to those of the adults. The inverse relation found between the Hb response and the severity of the anaemia does not support the recent recommendation, in the ECCO Guidelines on management of IDA in

Table 3. Laboratory results and psychometric scores before and after ferrous sulphate 200 mg twice daily for 6 weeks in the 34 adolescent and 32 adult patients completing the trial [PP analysis].

Variable	Adolescents [n = 34] Before after change	*p* for difference within adolescent group	Adults [n = 32] Before after change	*p* value for difference within adult group	*p* value between groups at baseline	*p* between groups for change in variable
Hb [g/dl]	10.2 [0.2] 12.0 [0.3] 1.8 [0.3]	**<0.0001**	10.9 [0.2] 12.2 [0.2] +1.3 [0.3]	**<0.0001**	**0.02**	0.21
Transferrin saturation [%]	6.7 [0.6] 20.7 [2.7] +13.9 [2.8]	**<0.0001**	8.1 [0.6] 22.6 [2.7] +13.8 [2.5]	**<0.0001**	0.10	0.98
Ferritin [µg/l]	16.2 [3.7] 29.9[3.9] + 13.7 [2.4]	**<0.0001**	25.6 [11.7] 127 [78.2] +101.4 [79.4]	0.21	0.43	0.26
Serum hepcidin [ng/ml]	29.2 [2.8] 35.4 [3.7] +6.2 [3.1]	0.06	27.8 [2.8] 36.2[4.2] +8.4 [3.2]	**0.01**	0.72	0.45
C-reactive protein [mg/l]	7.7 [2.1] 8.2 [2.0] +0.5 [1.5]	0.75	8.6 [2.2] 10.1 [3.0] +1.5 [3.4]	0.66	0.76	0.78
FCP [µg/g]	293 [38] 257 [35] −35 [38]	0.37	301 [80] 260 [61] −40 [81]	0.62	0.93	0.96
HBI	2 [0.5] 2 [0.6] +0.3 [0.7]	0.69	3 [0.7] 3 [0.8] +0.1 [0.7]	0.88	0.11	0.39
SCCAI	3 [0.5] 3 [0.6] +0.6 [0.4]	0.12	3 [0.6] 4 [0.8] +0.8 [1.0]	0.43	0.39	0.85
SIBDQ	51 [2] 54 [2] +3.2 [2.6]	0.22	42 [3] 47 [2.3] +5.7 [2.0]	**0.009**	**0.007**	0.49
HADS-A	6.7 [0.8] 6.0 [0.8] −0.7 [0.5]	0.21	8.2 [0.8] 7.1 [0.8] −1.1 [0.7]	0.11	0.87	0.85
HADS-D	4.2 [0.7] 3.8 [0.7] −0.5 [0.5]	0.38	5.8 [0.7] 4.9 [0.6] −0.9 [0.7]	0.18	0.46	0.62
PSQ-G	55 [2.5] 59 [3.2] +4.1 [2.5]	0.12	70 [3] 58 [3] −11.8 [2.5]	**< 0.0001**	**< 0.0001**	**< 0.0001**
MFI	58 [1.2] 59 [1.1] +0.9 [1.4]	0.20	59 [1.3] 61 [1.1] +1.9 [0.7]	0.13	0.79	0.69

Bold denotes significant *p*-value. Abbreviations: FCP, faecal calprotectin; HBI, Harvey-Bradshaw Index; SIBDQ, Simple IBD Questionnaire; HADS-A and -D, Hospital Anxiety and Depression Scale for Anxiety (-A) and Depression (-D); PSQ-G, Perceived Stress Questionnaire-General; MFI, Multidimension Fatigue Inventory.

Table 4. Univariate analysis of baseline factors significantly related to Hb response in 66 patients completing the protocol [PP analysis].

	R [beta]	Beta SE	*t*	*p*
CRP	−0.42	0.02	−3.7	**<0.0001**
Hb	−0.57	0.15	−5.55	**<0.0001**
Transferrin saturation	−0.42	0.05	−3.71	**<0.0001**
Hepcidin	−0.28	0.01	−2.3	**0.025**

Bold denotes significant *p*-value. Abbreviation: CRP, C-reactive protein.

IBD, that use of oral iron should be restricted to patients with mild anaemia [Hb >11 g/dl].[2]

Because of the possibility that iron absorption may be reduced in patients with active disease, the ECCO guidelines also suggest that such patients should be given intravenous rather than oral iron.[2] In support of this recommendation, we found negative associations between the Hb response and baseline CRP [as previously reported[14]], and, on univariate analysis, baseline hepcidin concentrations. There was also a trend to a smaller Hb response in patients with mixed IDA/anaemia of chronic disease than with 'pure' IDA [Supplementary Material Tables A and B]. Therapeutic decisions in anaemic IBD patients might therefore be guided by measurement of their CRP, and, if available, serum hepcidin concentrations:[14,33] those with raised levels might be offered intravenous rather than oral iron so as to bypass the inhibitory effect of hepcidin on intestinal absorption of iron.

Oral iron in anaemic adults with IBD has previously been shown to improve QOL.[6–10] We found that the Hb response to oral iron was associated with small but significant improvements in SIBDQ and PSQ-G in adults, and in SIBD-Q and HADS-A when both groups were analysed together. Reasons for our otherwise largely negative

Table 5. Multivariate regression analysis of baseline factors found on univariate analysis to be related to Hb response to oral iron.

	Beta [*R*]	*B*	SEM	*t*	95% CI [B]	*p*
Hb	−0.40	−0.57	0.18	−3.2	−0.93 to −0.21	**0.003**
CRP	−0.30	−0.04	0.02	−2.73	−0.07 to −0.001	**0.008**
Hepcidin	−0.06	−0.007	0.012	−0.58	−0.03 to +0.02	0.56
Transferrin saturation	−0.15	−0.08	0.06	−1.24	−0.2 to +0.05	0.22

Table 6. Adverse events and serious adverse events in the adolescent and adult patients during treatment with oral iron. UC denotes ulcerative colitis.

	Adolescents [*n* = 45]	Adults [*n* = 43]
Adverse events [AEs] [*n* = 15] [8 previously oral iron tolerant, 6 iron-naïve, 1 uncertain of iron use history]	2 Crohn's patients, abdominal pain 1 Crohn's, abdominal pain, constipation 1 Crohn's, abdominal pain, diarrhoea 1 UC, abdominal pain, diarrhoea 1 UC, nausea, constipation 1 UC, nausea, vomiting 1 UC, headache	2 Crohn's, abdominal pain 1 Crohn's, abdominal pain, diarrhoea 1 Crohn's, abdominal pain, constipation 3 UC, diarrhoea
Serious adverse events [SAEs] [*n* = 3] [all previously oral iron tolerant]	1 Crohn's, admission with abdominal pain, vomiting 1 Crohn's, admission with abdominal pain, vomiting, constipation	1 UC patient, admission with relapse of colitis

findings include the possibility that the Hb increases induced by oral iron given for only 6 weeks were too small to influence these measures, inaccurate completion of the questionnaires [see below] and under-powering of the study for these outcome measures.

It has been suggested that oral iron can exacerbate mucosal inflammation in patients with IBD. Although some studies of oral iron supplementation in adults have reported worsening disease activity symptom scores,[6,38–40] others have not.[7,8,41] Furthermore, there have been no studies reporting a consistent increase in inflammatory markers in patients with IBD given oral iron. In our 6-week study there was no suggestion of an oral iron-induced increase in disease activity in either patient group overall on the basis of symptom scores, CRP or faecal calprotectin. However, two recent studies in African children without IBD[42,43] have shown that supplementation of oral iron intake for 4–6 months is associated with a rise in faecal calprotectin. These results indicate that any pro-inflammatory effects of oral iron, whether acting as an oxidant and/or through changing the gut microbiome, take more than 6 weeks to become apparent,[10,42–44] a conclusion which may be specially relevant for IBD patients needing prolonged or repeated courses of oral iron to maintain their serum Hb.

Oral iron reportedly causes side effects leading to its discontinuation in up to 50% of patients,[45] but only about 17% of our iron-naïve patients withdrew. We found no evidence to support anecdotal claims that patients with Crohn's disease tolerate oral iron less well than those with UC [Table 6].

Unsurprisingly for an outpatient trial involving a standard oral drug which does not clearly improve disease symptoms, adherence to treatment was less than perfect. In this study, adherence to oral iron therapy, as assessed by returned tablets, was no worse in adolescents than adults, perhaps reflecting parental supervision in the younger patients. The fact that both groups' Hb levels rose despite their imperfect adherence provides support for evidence that only low doses of oral iron [about 60 mg elemental Fe daily] may be needed to improve iron stores and Hb levels.[46]

The study has several limitations. First, the findings may not be generalisable to other IBD populations. Our study was performed in an inner city tertiary referral centre where the case-mix is different from that in district hospitals or in the community. Second, the trial was not blinded. Blinding in oral iron trials is not feasible, as the stool often goes black. However, we used an objective primary outcome measure not influenced by the open label design. Third, we studied patients who had tolerated oral iron previously or were naïve to it. It would have been impracticable and unethical to attempt to recruit patients known to be intolerant of oral iron. Fourth, only about half of the recruited patients returned their medication containers for tablet counting. Despite the limitations of pill counting as a way of confirming medication adherence, this undermines the strength of the conclusions that can be drawn from the PP analysis. Finally, although we selected psychometric scales with simple wording which we believed would be readily intelligible by adolescent patients [E. Szigethy, personal communication], the questionnaires we used have not been validated for use in young people. While this potentially compromises comparisons between the adolescent and adult groups, it should not impair those made between individual patients' pre- and post-treatment psychometric scores.

In conclusion, contrary to previous suggestions, oral ferrous sulphate was as effective and well-tolerated in adolescents as in adults with IBD, and did not appear to increase disease activity during short-term use. Oral iron slightly improved QOL as measured by SIBDQ and perceived stress levels in adults, and by SIBDQ and HADS-A when both groups were analysed together. Adherence to, and adverse events resulting from, oral iron therapy were similar in adolescent and adult patients. These findings make it illogical not to treat IDA in adolescents with IBD in the same way as in adults. The findings that baseline serum CRP and, on univariate analysis, hepcidin levels were inversely related to the increase in Hb concentration produced by oral iron suggests that in patients with IBD these measures might help clinicians to decide by which route to offer iron replacement.

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Conflict of Interest

DSR has received speakers' fees and travel expenses from Vifor and Pharmacosmos, and payments for advisory board work from Pharmacosmos.

JRG, NMJ, A-BK, YK, FMB, LM, DGW, JE and IRS have no conflicts to declare. THI has received speaker fees from Vifor, Pharmacosmos and Shield Therapeutics. JMF has received fees for advisory board work for Abbvie and Janssen Pharmaceuticals.

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Author Contributions

JRG, DSR and IRS conceived the study, collected and analysed the data, and prepared the early drafts of the article. A-BK, FB, NMJ and LM recruited patients and/or collected samples, TI and DW analysed serum hepcidin concentrations, and YK, JE and JF coordinated the study and/or contributed to interpretation of the results. All authors approved the final text.

Supplementary Material

Supplementary data to this article can be found at *ECCO-JCC online*.

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