SYNOPSIS

Name of Company:	Vifor (International) Inc.
Name of Finished Product:	FERINJECT®
Name of Active Ingredient(s):	Ferric carboxymaltose

Title of Study:	A Multi-Center, Randomized, Controlled, Single-Blinded, Phase II Study to Investigate the Safety and Efficacy of Intravenous Infusions of FERINJECT® Versus Placebo in Patients with Thrombocytosis Secondary to Iron Deficiency and Chronic Inflammatory Bowel Disease (ThromboVIT)		
Investigators:	001 - Christoph Gasche 002 - Juergen Stein 003 - Stefanie Howaldt 004 - Oliver Mickisch		
Study Centre(s):	The study was conducted at four centers; three in Germany and one in Austria		
Publication (Reference):	None to date		
Studied Periods: **(Date of First Enrolment)** **(Date of Last Completed)**	22 December 2006 – 28 January 2010	**Phase of Development:**	2
Objectives:	The primary objective of the study was to evaluate the efficacy of FERINJECT in reducing elevated platelet counts. The secondary objectives were: • To evaluate the effect of FERINJECT on coagulation and platelet activation parameters. • To evaluate the efficacy of FERINJECT in normalizing iron deficiency. • To evaluate the change in quality of life and disease activity. • To evaluate the safety of FERINJECT.		
Methodology:	This was a phase II, multi-center, randomized, placebo-controlled, single-blind, parallel group study in irritable bowel disease (IBD) subjects with thrombocytosis and iron deficiency. Subjects attended a screening visit (Visit 1) up to two weeks before the start of		

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Name of Finished Product: FERINJECT®
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	treatment. Subjects were randomized to one of four treatment groups (500 mg FERINJECT, 1000 mg FERINJECT, 1500 mg FERINJECT and placebo). Protocol amendment 2 reduced the number of treatment groups to two (1500 mg FERINJECT and placebo) to adjust for recruitment that was more difficult than expected. At Visit 2, subjects underwent baseline assessments and received their first intravenous (IV) infusion of study treatment. Subjects attended the study center seven days later (Visit 3) for their second IV infusion and a further seven days later (Visit 4) for their third/final IV infusion. Subjects attended for a final study visit (Visit 5) six weeks after their first IV infusion. The duration of the study was six weeks, including three weeks of treatment and a three-week observation period.
Number of Patients:	It was planned to enroll 80 subjects (20 subjects in each of four treatment groups). However, the sample size was reduced to 50 subjects (25 subjects in each of two treatment groups) in protocol amendment 2 (dated 20 February 2009). A total of 19 subjects had been enrolled at the time of this amendment. A summary of subject disposition is provided in Table S1. Of 26 subjects assigned to study treatment, 25 subjects were treated (four, two and 10 subjects in the 500 mg, 1000 mg and 1500 mg FERINJECT treatment groups, respectively and nine subjects in the placebo treatment group). One subject was randomized (placebo) but did not receive study drug. Twenty three (23) subjects completed the study. Two subjects discontinued the study: one subject (1500 mg FERINJECT) due to other reason (received treatment with erythropoietin) and one subject (placebo) due to an AE (unexpected worsening of underlying disease/severe deterioration of ulcerative colitis). The majority of subjects in the study were female (20 of the 25 [80.0%] subjects treated). Mean age (standard deviation [SD]) for all subjects was 34.1 (9.97) years. The majority of subjects were Caucasian (22 of 25 [88.0%] subjects).

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Table S1 Disposition of subjects

	FERINJECT			Placebo
Number of subjects	500 mg	1000 mg	1500 mg	
Assigned to study treatment	4	2	10	10
Treated	4	2	10	9
Completed	4	2	9	8
Discontinued	0	0	1	1
Analyzed for efficacy[a]	0	0	10	9
Analyzed for safety	4	2	10	9

[a] The efficacy analysis was performed on the 1500 mg FERINJECT and placebo groups only. Week 6 data for Subject 3007 (placebo) was excluded from the analyses as data was not collected until Week 19.

Diagnosis and Main Criteria for Inclusion:

Male or female subjects (at least 18 and less than 60 years of age) with a platelet count >450G/L, transferrin saturation (TfS) <20% or ferritin <100 µg/L who had been previously diagnosed with IBD (Crohn's disease or ulcerative colitis) were eligible for this study. Subjects with a Crohn's disease activity index (CDAI) >220 and colitis activity index (CAI; Rachmilewitz) >6, or significant anemia (Hb [hemoglobin] <10.5 g/dL) or anemia not caused by iron deficiency were excluded from the study.

Test Product, Dose and Mode of Administration, Batch Number(s):

Subjects received a single IV infusion of FERINJECT or placebo at Visits 2, 3 and 4 each separated by seven days. FERINJECT or placebo was administered IV into a peripheral vein in the arm. 500 mg FERINJECT was diluted to a total volume of 100 mL in 0.9% saline for infusion and administered over 15 minutes duration.

The original protocol included four treatment groups. The number of treatment groups was reduced to two (1500 mg FERINJECT and placebo) in protocol amendment 2. The original four treatment groups were:

1500 mg FERINJECT: Subjects received one infusion of 500 mg FERINJECT diluted in 100 mL 0.9% sodium chloride every seven days for a total of three infusions (1500 mg).

1000 mg FERINJECT: Subjects received one infusion of 500 mg FERINJECT diluted in 100 mL 0.9% sodium chloride every seven days for a total of two infusions (1000 mg) followed by one placebo infusion of 100 mL 0.9% sodium chloride.

500 mg FERINJECT: Subjects received one infusion of 500 mg FERINJECT diluted in 100 mL 0.9% sodium chloride (500 mg)

Name of Company: Vifor (International) Inc.	
Name of Finished Product: FERINJECT®	
Name of Active Ingredient(s): Ferric carboxymaltose	

followed by two placebo infusions of 100 mL 0.9% sodium chloride every seven days.

Placebo: Subjects received one placebo infusion of 100 mL 0.9% sodium chloride every seven days for a total of three infusions.

The sponsor supplied each site with sufficient FERINJECT solution for the study. Study treatments were to be stored in a locked area at room temperature between 4 and 25°C. Lot numbers are presented in Table S2.

Table S2 FERINJECT Lot Numbers

Study Drug	Dosage Form	Lot Numbers
FERINJECT	Small volume parenteral, terminally sterilized	597200 (expiry 09/2007), 507210 (expiry 10/2008), 518010 (expiry 11/2008), 882200 (expiry 08/2011)

Duration of Treatment:	Subjects received one infusion per week for three weeks.
Reference Therapy, Dose and Mode of Administration, Batch Number(s):	Placebo treatment consisted of intravenous infusions of 0.9% sodium chloride solution. Batch numbers were not recorded.
Criteria for Evaluation:	
Efficacy:	Efficacy evaluations included: • Platelet count: assessed at all visits. • Platelet activation markers (p-selectin, sCD40L), thrombopoietin and reticulated thrombocytes: assessed at Visits 2, 4 and 5. • Coagulation parameters (partial thromboplastin time [PTT], prothrombin time [PT], factors of the intrinsic coagulation pathway): assessed at Visits 2, 4 and 5. • Iron parameters (ferritin, Hb, transferrin, TfS, soluble transferrin receptor, hepcidin): assessed at all visits except hepcidin assessed at Visits 2, 4 and 5 only. • Quality of life (inflammatory bowel disease questionnaire [IBDQ] and functional assessment of cancer therapy-anemia scale [FACT-An]) and disease activity (CDAI/CAI) questionnaires. Quality of life

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Name of Finished Product: FERINJECT®
Name of Active Ingredient(s): Ferric carboxymaltose

questionnaires were completed at Visits 2, 4 and 5 and disease activity questionnaires at all visits.

- C-reactive protein, erythrocyte sedimentation rate (ESR): assessed at all visits
- IL-3, IL-6, IL-11 and calprotectin levels: assessed at Visits 2, 4 and 5.

Safety: Safety evaluation included adverse events (AEs), serious adverse events (SAEs), safety laboratory tests, and vital signs (blood pressure, heart rate and body temperature).

Statistical Methods: The primary endpoint was a binary response variable, where a positive response was defined as a decrease in platelet count of >25% at Week 6 as compared to baseline. The Week 6 percentage change in platelet count for each subject was derived as follows:

$$\frac{[\text{Week 6 platelet count} - \text{Baseline platelet count}]}{\text{Baseline platelet count}} \times 100$$

Subjects with a Week 6 percentage decrease in platelet count >25% were classified as responders. Subjects not achieving this level of response were classified as non-responders.

The baseline value was defined as the Visit 2 measurement collected prior to the first infusion of study treatment (or the Visit 1 measurement if the Visit 2 measurement was missing). Week 6 platelet count was the Visit 5 measurement.

Platelet counts for each visit, change from baseline at each visit, Week 6 percentage change from baseline, and the response rate were listed and summarized by treatment group.

The difference between the response rates in the 1500 mg FERINJECT and placebo treatment groups were analyzed using Fisher's exact test. The exact one-sided p-value from this test was presented. No statistical analyses were performed on data from the 500 mg and 1000 mg FERINJECT treatment groups.

Clinical disease activity scores (CDAI and CAI) were collected at all visits. Quality of Life questionnaires (IBDQ and FACT-An) were collected at Visits 2, 4 and 5. Baseline values for each of these endpoints were defined as the Visit 2 measurements collected prior to the first infusion of study treatment.

Clinical disease activity scores (CDAI and CAI) and quality of life questionnaires (IBDQ and FACT-An) were listed. CDAI, CAI, IBDQ and

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Name of Finished Product:
FERINJECT®
Name of Active Ingredient(s):
Ferric carboxymaltose

FACT-An scores at each visit and the change from baseline to each visit were summarized by treatment group and visit.

AEs were summarized using MedDRA (Version 13.1).

AEs were listed and summarized by treatment group.

Clinical laboratory parameters were listed and summarized (where appropriate) by treatment group and visit.

The following vital signs variables were collected: systolic blood pressure, diastolic blood pressure, heart rate and temperature. Baseline values for each of these variables were defined as the Visit 2 measurements collected prior to the first infusion of study treatment. For each variable, the change from baseline value at each post-baseline visit was calculated as the difference between the measurements obtained at the specific post-baseline visit and the baseline value. Vital signs data were listed and summarized by treatment group and visit.

Physical examination and medical history data were listed.

Summary of Results:

Efficacy Results:

The efficacy analysis was performed on the 1500 mg FERINJECT and placebo groups only; all 19 subjects treated in these groups were included in the full analysis set and analyzed for efficacy. Week 6 data for Subject 3007 (placebo) was excluded from the analyses as data was not collected until Week 19.

The proportion of females:males and the mean age of subjects in the 1500 mg FERINJECT and placebo treatment groups (i.e., treatment groups analyzed for efficacy) were similar.

The primary endpoint was a binary response variable, where a positive response was defined as a decrease in platelet count of >25% at Week 6. Out of 10 subjects randomized to each of the 1500 mg FERINJECT and placebo treatment groups, only eight subjects in the 1500 mg FERINJECT treatment group and seven subjects in the placebo treatment group were evaluable for the primary efficacy assessment. Four of eight (50.0%; Subjects 02001, 03003, 03004 and 03009) subjects in the 1500 mg FERINJECT treatment group and one of seven (14.3%; Subject 04003) subjects in the placebo treatment group were classified as responders. The difference between these two treatment groups was not statistically significantly different (p=0.182).

Having started at similar baseline platelet counts, the mean percentage change in platelet count from baseline to Week 6 was greater in the 1500 mg FERINJECT treatment group compared to the placebo treatment group (-26.8% and -2.4%, respectively). The mean percentage change in platelet count from

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Name of Finished Product: FERINJECT®
Name of Active Ingredient(s): Ferric carboxymaltose

baseline to Week 6 was also greater in the 500 mg and 1000 mg FERINJECT treatment groups (-13.4% and -15.9%, respectively) compared to the placebo treatment group (-2.4%); however, the numbers of subjects in the 500 mg and 1000 mg treatment groups were small (four and two, respectively).

Secondary endpoints included clinical disease activity (CDAI and CAI) and quality of life questionnaire (IBDQ and FACT-An) scores and changes from baseline. The number of subjects with data available for CAI and FACT-An was low (≤50% of subjects in each treatment group at Week 6); no interpretation of the data is possible due to the number of subjects with data available.

Safety Results:

All 25 treated subjects were analyzed for safety. Table S3 summarizes treatment-emergent AEs (TEAEs) (all causality and treatment related). Five (50.0%) subjects experienced at least one TEAE in the 1500 mg FERINJECT treatment group and three (33.3%) subjects experienced at least one TEAE in the placebo group. One (25.0%) subject in the 500 mg FERINJECT treatment group experienced a TEAE. No TEAEs were reported in the 1000 mg FERINJECT treatment group.

Three (30.0%) subjects in the 1500 mg FERINJECT treatment group and one subject (25.0%) in the 500 mg FERINJECT treatment group experienced at least one TEAE considered related to study treatment by the investigator.

The most frequently reported TEAEs (classified according to the MedDRA System Organ Class) were gastrointestinal disorders, infections and infestations, and skin and subcutaneous tissues disorders

One subject (placebo) discontinued from the study due to a TEAE of severe deterioration of ulcerative colitis. This was not considered related to study treatment by the investigator. The event was considered an SAE due to hospitalization.

Three subjects experienced a total of four SAEs (two subjects in the 1500 mg FERINJECT treatment group and one subject in the placebo treatment group). One SAE (severe systemic inflammatory response syndrome; 1500 mg FERINJECT) was considered related to study treatment by the investigator. No deaths were reported during the study.

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Ferric carboxymaltose

Table S3 Summary of Treatment-Emergent Adverse Events – All Causality (Treatment-Related)

	FERINJECT			
Number of subjects	**500 mg (N=4)**	**1000 mg (N=2)**	**1500 mg (N=10)**	**Placebo (N=9)**
Subjects evaluable for adverse events	4	2	10	9
Number of TEAEs	1	0	14	3
Subjects with TEAEs	1 (1)	0 (0)	5 (3)	3 (0)
Subjects with severe TEAEs	0	0	1 (1)	1 (0)
Subjects with SAEs	0 (0)	0 (0)	2 (1)	1 (0)
Subjects discontinued due to TEAEs	0 (0)	0 (0)	0 (0)	1 (0)
Deaths	0	0	0	0
N = number of subjects treated Related TEAEs are defined as TEAEs where the relationship to study medication was recorded as 'unlikely', 'possibly'. 'probably', 'certainly' or missing.				

Greater changes from baseline in iron parameters (ferritin, serum iron, soluble transferrin receptor, transferrin, and TfS) were observed at each visit following treatment with 1500 mg FERINJECT compared to placebo indicating that FERINJECT may normalize iron deficiency in IBD subjects with thrombocytosis and iron deficiency. Additionally, a mean increase in Hb of 1.40 g/dL was observed at Week 6 in subjects treated with 1500 mg FERINJECT compared with a mean decrease of 0.13 g/dL in the placebo treatment group.

There were no clinically significant changes in vital signs.

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Conclusion:

- More subjects treated with 1500 mg FERINJECT than placebo were classified as responders (defined as a decrease in platelet count of >25% at Week 6). The difference between these groups was not statistically significant. The mean percentage change in platelet count from baseline to Week 6 was also greater in the 1500 mg FERINJECT treatment group compared to the placebo treatment group.
- Due to the limited amount of data available, the effects of FERINJECT on coagulation and

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Name of Finished Product: FERINJECT®
Name of Active Ingredient(s): Ferric carboxymaltose

platelet activation parameters were not analyzed.

- There was some evidence that FERINJECT may normalize iron deficiency in IBD subjects with thrombocytosis and iron deficiency.
- Limited data are available for clinical disease activity and quality of life assessments so no meaningful conclusions can be drawn.
- FERINJECT, at doses of 500 mg, 1000 mg and 1500 mg, was generally well tolerated.
