Pierre Fabre Médicament
Represented by: Institut de Recherche Pierre Fabre
45, Place Abel Gance
F-92654 Boulogne Cedex

1. TITLE PAGE

CLINICAL STUDY REPORT

Treatment of iRon dEficiency Anaemia with Tardyferon®

Importance of prolonging iron supplementation in anaemic women of childbearing age.

Investigational product:	Tardyferon® tablet 80 mg
Study Design:	International, multicentre, placebo-controlled, randomised, on parallel group study
Protocol number:	L00008 CP 402
Phase of development:	IV
Date of first enrolment:	24/03/2007
Date of last completed:	8/07/2009
Co-ordinator:	Pr Pierre-Louis DRUAIS
Sponsor Representative(s) for study report:	Pascal Olier Pierre Fabre Médicament La Chartreuse, 81106 Castres Cedex, France Tel +33 5 63 71 39 10

Date of report: 7 December 2011 (Final)

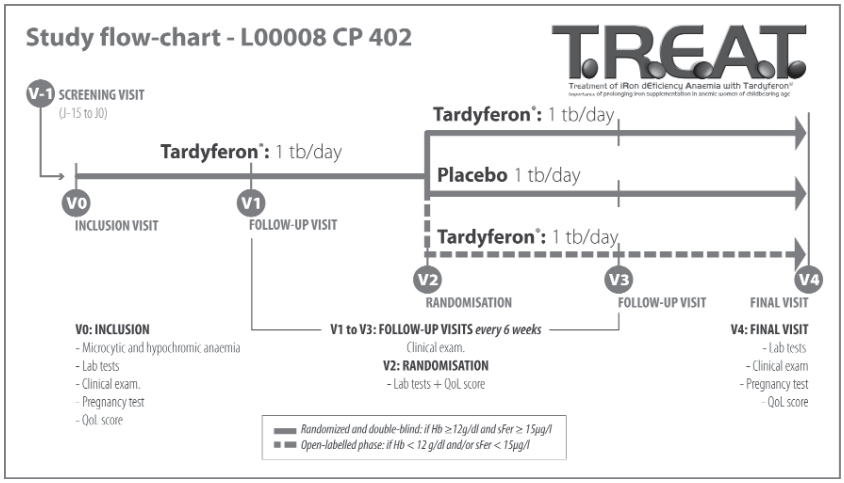
Study performed in compliance with Good Clinical Practice.

2. SYNOPSIS

Name of Company: Pierre Fabre Médicament	**Individual Study Table**	**(For National Authority Use Only)**
Name of finished product: Tardyferon®	**Referring to Module 5 of the Dossier**	
Name of active substance *(or ingredient)*: **Ferrous sulphate**	**Vol.: .....Page: .....**	

Title of study:	**T**reatment of i**R**on d**E**ficiency **A**naemia with **Tardyferon®** Importance of prolonging iron supplementation in anaemic women of childbearing age. **TREAT study (Protocol number L00008 CP 402)**	
Coordinating Investigators:	Pr Pierre-Louis DRUAIS (16 bis avenue Simon Vouet - 78560 Le Port-Marly – France)	
Study centre(s):	11 centres: 9 GPs centres in France and 2 Hospital centres in Argentina	
Publication (reference):	No publication at the time of the report.	
Studied period *(years, months ...)*: **(date of first enrolment)** **(date of last completed)**	17/04/2007 8/07/2009	**Phase of development:** IV
Rationale	Iron Deficiency Anaemia (IDA) is a common pathology occurring in less than 5% of women aged 15 to 49 years in western countries but can affect up to 30% of the world population, knowing that 9 of 10 anaemic patients live in developing countries. Treating at least 3 months allows to normalise haemoglobin but it is important to carry on the iron treatment in order to fully replenish the iron stores. This study investigated the potential positive effect of replenishing the iron stores to a ferritin level of above 15 µg/l.	
Objectives:	• To assess the kinetics of iron parameters under Tardyferon treatment. • To assess the necessity to treat at least 24 weeks, IDA patients, to help them recover their haemoglobin and restore their iron stores correctly (percentage of patients reaching serum ferritin > 40 µg/l and percentage of patients achieving haemoglobin > 12g/dl). If the patients are not correctly treated, their haemoglobin and/or serum ferritin are low, thus their quality of life can be affected (QoL scores).	
Methodology:	This was an international, multicentre, randomised, parallel group study including iron deficient anaemic female patients. This study had two phases: • one open-label phase for 12 weeks with Tardyferon® • one second phase of 12 weeks: - randomised, double-blind phase either on Tardyferon® or on Placebo if at V2 Hb $\geq$ 12g/dl and serum ferritin $\geq$15 µg/l - if at V2 Hb < 12g/dl and/or ferritin < 15µg/l, another open-labelled phase on Tardyferon® to maintain the treatment	

Name of Company: Pierre Fabre Médicament	Individual Study Table	(For National Authority Use Only)
Name of finished product: Tardyferon®	Referring to Module 5 of the Dossier	
Name of active substance *(or ingredient)*: Ferrous sulphate	Vol.: .....Page:	



Diagnosis and main criteria for inclusion:

Inclusion criteria:
Patients fulfilling all of the following criteria could be included in the study:

a) Demographic characteristics and other baseline characteristics:
- Female,
- Patient has attained her majority (between 18 years old and 21 years old according to country),
- Ambulatory,
- Menstruating,
- Anaemic (Hb level: 90-120 g/l) ,
- Hyposideremic: Serum Ferritin < 15ng/ml,
- MCV < 85µ3
- Patient under effective hormonal contraception (combination birth control pills, under-the-skin implants), intrauterine device, tubal sterilization or abstinence.

b) Diagnostic criteria:
- Mild to moderate iron deficiency anaemia.

c) Ethical considerations:
- Patients having signed their written informed consent.

Exclusion criteria:
Patients fulfilling at least one of the following criteria were not to be included in the study:

a) Disease characteristics:
- Anaemia with Hb level < 90 g/l
- Normocytic or macrocytic anaemia
- Normochromic or hyperchromic anaemia
- Normosideremic or hypersideremic anaemia

b) Differential diagnosis:
- Anaemia due to inflammatory disease,
- Hemochromatosis,
- Thalassemia,
- Anaemia due to medullar insufficiency,
- Refractory anaemia,
- Chronic renal insufficiency.

c) Other diseases:
- Any disease considered as life-threatening in a short or mid-term (e.g. cardiac insufficiency, cancer in evolution; severe renal insufficiency)
- Hepatic disease declared,
- Intestinal occlusion,

Name of Company: Pierre Fabre Médicament	Individual Study Table	(For National Authority Use Only)
Name of finished product: Tardyferon®	Referring to Module 5 of the Dossier	
Name of active substance *(or ingredient)*: Ferrous sulphate	Vol.: .....Page:	

- Glucose and /or galactose malabsorption syndrome,
- Sucrose isomaltase deficit syndrome,
- Fructose hypersensitivity,
- Chronic renal insufficiency.

d) Treatments:
- History of hypersensitivity to the test drug,
- Intake of other iron salts by oral or injectable route.

e) Habits:
- The following substances had to be used with caution:
  - Large quantities of tea, coffee, red wine inhibit iron absorption,
  - Dairy products and eggs can significantly reduce iron absorption when used simultaneously.

f) Other exclusion criteria:
- Was a family member or a work associate (secretary, nurse, technician) of the Investigator,
- Pregnancy,
- Extreme vegetarian diet,
- Regular blood donors or who gave blood within the 8 weeks before the study,
- Breastfeeding women,
- Having participated in any other clinical trial within the last month,
- Having received treatment with known remnant effects or having undergone investigation likely to interfere with the present clinical trial,
- Mentally unable to understand the nature, objectives and possible consequences of the trial on herself to its constraints,
- Having forfeited her freedom of an administrative or legal obligation or being under guardianship.

Test product, **Dose,** **Mode of administration,** **Batch number:**	Tardyferon® 80 mg 1 tablet/ day Oral route LOT G00469 / G00723
Reference therapy, **Dose,** **Mode of administration,** **Batch number:**	Placebo 1 tablet / day Oral route SB028
Duration of treatment	24 weeks
Criteria for evaluation:	
Efficacy:	<u>Primary efficacy endpoint:</u> Serum ferritin level at V4 (difference from baseline (V0), from V2 and absolute value) <u>The secondary efficacy endpoints were:</u> - Percentage of normalised (Hb ≥120 g/l and serum ferritin ≥ 40 ng/ml) patients in each randomisation group at 24 weeks - Haemoglobin level at V4 (difference from baseline (V0), from V2 and absolute value), - Serum iron at V4 (difference from baseline (V0), from V2 and absolute value), - Transferrin saturation at V4 (difference from baseline, from V2 and absolute value), - Serum Transferrin receptors V4 (difference from baseline, from V2 and absolute value), - QoL scores at 24 weeks, - Symptom development at each visit

Name of Company: Pierre Fabre Médicament	Individual Study Table	(For National Authority Use Only)
Name of finished product: Tardyferon®	Referring to Module 5 of the Dossier	
Name of active substance *(or ingredient)*: Ferrous sulphate	Vol.: .....Page:	

	- Percentage of patients with Hb ≥ 120 g/l and serum ferritin between 15 and 50 ng/ml at visit V4 - Treatment compliance
Safety:	Safety was assessed by the collection of local/regional and systemic adverse events at each visit.
Sample size calculation	The sample size calculation was performed according to the primary objective initially chosen: percentage of normalised (Hb ≥120 g/l and serum ferritin ≥ 40 ng/ml) patients in each randomisation group at 24 weeks. This objective was modified during the study (see admendments and changes in thr protocol) As this study initially used 2 primary variables, the sample size was calculated on the basis of 2 variables: the percentage of patients with serum ferritin ≥ 40 ng/ml (normalised serum ferritin) and the percentage of patients with Hb ≥ 120 g/l (normalised Hb). Among the randomised patients, the percentage of subjects with normalised serum ferritin at the end of the study was expected to be about 20% in the placebo group and 60% in the Tardyferon® group. With these assumptions, 23 patients per group among the randomised patients were required in the second period of the study to demonstrate a significant difference ($p<0.05$) with a power of 80%. Among the randomised patients, the percentage of subjects with normalised Hb at the end of the study was expected to be about 30% in the placebo group and 70% in the Tardyferon® group. With these assumptions, 24 patients per group among the randomised patients were required in the second period of the study to demonstrate a significant difference ($p<0.05$) with a power of 80%. Thus, 50 randomised patients on average (25 patients in each group) completing the study randomised phase with analysable data were required. Taking into account that in such a study the inclusion failure rate can exceed 20% and there may be up to 30% dropouts because of the study duration, further inclusions were planned. 80 patients needed to be included at V0 for a minimum of 50 patients to be analysed.
Statistical methods:	**DOUBLE-BLIND PHASE** Among the randomised patients, the different types of variables were compared between treatment groups as follows: - The ferritin rate (level at 24 weeks, absolute change from Baseline, absolute change from V2) was compared between treatment groups using Cochran-Mantel-Haenszel (CMH) with modified ridit scores, adjusting for country group. - The ferritin rate (adjusted according to treatment group and with the level at V2 as covariate) was analysed using a Non-parametric Analysis of Covariance. - The ferritin rate (adjusted according to treatment group and with the level at V2 and the HB level at V4 as covariates) was analysed using a Non-parametric Analysis of Covariance. - The percentage of normalised patients (Hb ≥ 120 g/l and serum ferritin ≥ 40 ng/ml) in both groups of randomised subjects (treatment group and placebo group), after 24 weeks of treatment was analysed, using a Cochran-Mantel-Haenszel test stratified by country group.

Name of Company: Pierre Fabre Médicament	Individual Study Table	(For National Authority Use Only)
Name of finished product: Tardyferon®	Referring to Module 5 of the Dossier	
Name of active substance *(or ingredient)*: Ferrous sulphate	Vol.: .....Page:	

- The percentage of patients with Hb ≥ 120 g/l and serum ferritin between 15 and 50 ng/ml at V4 was analysed, using a Cochran-Mantel-Haenszel test stratified by country group.
- Physical examination was analysed using Cochran-Mantel-Haenszel (CMH) with modified ridit scores, adjusting for country group. Only descriptive statistics were given for treatment compliance.
- Quantitative variables (haemoglobin, transferrin) was analysed using Cochran-Mantel-Haenszel (CMH) with modified ridit scores, adjusting for country group. Only descriptive statistics were given for changes levels of serum iron.
- Quantitative variables (quality of life scores) were compared between treatment groups using Cochran-Mantel-Haenszel (CMH) with modified ridit scores, adjusting for country group.
- All these parameters were also described by country.

For the physical examination which was a list of symptoms described by their presence or absence, a derived variable was calculated as follows between baseline and each visit, and between V2 and each visit, for randomised subjects:

- If the symptom was present at visit V0/V2 and still present at visit n+1, the state was stability.
- If the symptom was present at visit V0/V2 and absent at visit n+1, the state was improvement.
- If the symptom was absent at visit V0/V2 and present at visit n+1, the state was worsening.

Safety was assessed on the basis of adverse events.

- These were the subject of a descriptive analysis only, for each group.

OPEN-LABEL PHASE

Descriptive statistics for the patients continuing on Tardyferon open-label (Hb<12g/dl at visit 2)

- percentage of patients with Hb ≥12g/dl and serum ferritin between 15 and 50 µg/l at visit 3 and 4
- changes in haemoglobin and serum ferritin levels
- changes in SF-12 scores

Due to recruitment difficulties, the number of included subjects was not sufficient to test the original null hypothesis. Therefore the statistical analyses were changed (in Protocol, Amendment 3). The protocol amendments and changes in the statistical analysis plan are described below.

Amendments and changes in the protocol

Four protocol amendments were submitted. All these amendments were related to the difficulties of recruitment in the first part of the study.

Amendment N° 1, July 2007

The investigators from hospitals in Argentina had difficulties to recruit and as long as the French sites are concerned, the major reason for this situation was possibly linked to the pre-selection visit V-1: it was difficult to find patients with a biological analysis dating from less than one month that attested that they were anaemic, and untreated for this condition. Another possibility of eligibility at V-1 was added: a patient whose anaemia was clinically suspected by the investigator could also be eligible for the study.

Name of Company: Pierre Fabre Médicament	Individual Study Table	(For National Authority Use Only)
Name of finished product: Tardyferon®	Referring to Module 5 of the Dossier	
Name of active substance *(or ingredient)*: Ferrous sulphate	Vol.: .....Page:	

Amendment N° 2, October 2007

To facilitate recruitment, 2 inclusion criteria were modified:

- Upper limit of acceptable MCV value was increased from 80µ3 to 85µ3
- Because pregnancy was an exclusion criterion, patients had to use effective forms of contraception. The possibility to include patients abstinent during the study period was added.

Amendment N° 3, February 2008

- Considering the difficulties to find suitable patients, inclusion criteria were modified. Patients with mild anaemia (Hb level between 110 to 120 g/l) could be included in the study.
- Recruitment period was also prolonged for 15 months.
- The statistical analysis plan for the primary efficacy criterion was modified.

Amendment N° 4, September 2008

Considering the difficulties to recruit patients, an additional prolongation of recruitment period of 3 months was planned.

Final number of patients

Despite several protocol amendments performed to facilitate recruitment, a total number of 67 patients was included vs. 80 patients initially planned; 29 patients were randomised in the double-blind phase vs. 50 patients initially planned.

27 and 22 patients were analysed for efficacy in the ITT and PP populations respectively.

There was a doubt on the treatment intake of one patient. She was analysed in the safety population but she was not included in the ITT population. Finally 67 patients were analysed for efficacy and 68 for safety .

Consequences for the primary and secondary criteria

The primary efficacy endpoint originally planned was the percentage of normalised (Hb ≥120 g/l and serum ferritin ≥ 40 ng/ml) patients in each randomisation group at 24 weeks

Following recruitment difficulties, the number of patients was not adequate to test the null hypothesis. Therefore, the primary criterion was adapted and the ferritin level at the end of the study (V4) was used as new primary criterion. Ferritin level at V4 could be considered as the level that was globally reached by the patients from the two groups, reflecting the efficacy of treatment in correcting iron deficiency.

The percentage of patients reaching Hb ≥ 120 g/l and and serum ferritin ≥ 40 µg/l (initial primary criterion) was analysed as a secondary criterion.

Consequences on the statistical methods – changes in the Statistical Analysis Plan

Primary criteria (original)

The percentage of patients with normalised serum ferritin at 24 weeks to be analysed by the chi-square test or the Fisher exact test if required.

The percentage of patients with normalised haemoglobin at 24 weeks to be analysed by the chi-square test or the Fisher exact test if required.

Secondary criteria (original)

Binary criteria (percentage of patients with Hb ≥ 12g/dl and serum ferritin between 15 and 50 µg/l) to be compared between treatment groups by the chi-square test or the Fisher exact test if required.

Name of Company: Pierre Fabre Médicament	Individual Study Table	(For National Authority Use Only)
Name of finished product: Tardyferon®	Referring to Module 5 of the Dossier	
Name of active substance *(or ingredient)*: Ferrous sulphate	Vol.: .....Page:	

Quantitative criteria (haemoglobin, serum ferritin, transferrin saturation and quality of life scores) to be compared between treatment groups by Student's t-test.

Ordinal criteria (symptoms) to be compared between treatment groups by the Wilcoxon-Mann-Whitney test.

Descriptive statistics to be given for serum iron an serum transferrin receptors (changes levels of each parameter).

Missing data were not to be replaced.

Due to recruitment difficulties, statistical analysis methods had to be adapted :

Among the randomised patients, the different types of variables were compared between treatment groups as follows:

- The ferritin rate (level at 24 weeks, absolute change from Baseline, absolute change from V2) was compared between treatment groups using Cochran-Mantel-Haenszel (CMH) with modified ridit scores, adjusting for country group.
- The ferritin rate (adjusted according to treatment group and with the level at V2 as covariate) was analysed using a Non-parametric Analysis of Covariance.
- The ferritin rate (adjusted according to treatment group and with the level at V2 and the HB level at V4 as covariates) was analysed using a Non-parametric Analysis of Covariance.
- The percentage of normalised patients (Hb $\geq$ 120 g/l and serum ferritin $\geq$ 40 ng/ml) in both groups of randomised subjects (treatment group and placebo group), after 24 weeks of treatment was analysed, using a Cochran-Mantel-Haenszel test stratified by country group.
- The percentage of patients with Hb $\geq$ 120 g/l and serum ferritin between 15 and 50 ng/ml at V4 was analysed, using a Cochran-Mantel-Haenszel test stratified by country group.
- Physical examination was analysed using Cochran-Mantel-Haenszel (CMH) with modified ridit scores, adjusting for country group. Only descriptive statistics were given for treatment compliance.
- Quantitative variables (haemoglobin, transferrin) was analysed using Cochran-Mantel-Haenszel (CMH) with modified ridit scores, adjusting for country group. Only descriptive statistics were given for changes levels of serum iron.
- Quantitative variables (quality of life scores) were compared between treatment groups using Cochran-Mantel-Haenszel (CMH) with modified ridit scores, adjusting for country group.
- All these parameters were also described by country.

For the physical examination which was a list of symptoms described by their presence or absence, a derived variable was calculated as follows between baseline and each visit, and between V2 and each visit, for randomised subjects:

- If the symptom was present at visit V0/V2 and still present at visit n+1, the state was stability.
- If the symptom was present at visit V0/V2 and absent at visit n+1, the state was improvement.
- If the symptom was absent at visit V0/V2 and present at visit n+1, the state was worsening.

Safety was assessed on the basis of adverse events.

- These were the subject of a descriptive analysis only, for each group.

Results

Only 67 patients were included at Baseline (versus 80 patients initially planned) and 29 were randomised at V2 (versus 50 initially planned); 27 patients were analysed in the ITT population (11 in Tardyferon® group and 16 in placebo group) and 22 in the PP population.

Patients included in the study were aged between 19 and 51 years (mean (SD) = 36.8 (9.4) years).

The main causes of IDA in this population were menorrhagia/metrorrhagia (61.8% of the patients) and low consumption of meat (51.5% of the patients).

Name of Company: Pierre Fabre Médicament	Individual Study Table	(For National Authority Use Only)
Name of finished product: Tardyferon®	Referring to Module 5 of the Dossier	
Name of active substance *(or ingredient)*: Ferrous sulphate	Vol.: .....Page:	

Efficacy results (double blind phase)

After 24 weeks of treatment (V4), mean serum ferritin levels were 38.9 ng/ml in the Tardyferon® group and 13.6 ng/ml in the placebo goup.

Normalisation defined as Hb above 120 g/l and serum ferritin above 40 ng/ml was observed in 4/11 patients (36.4%) with Tardyferon®. No patient normalised with placebo after 24 weeks of treatment (0/16 patients).

In Tardyferon® group, serum ferritin levels continued to increase and Hb and transferrin saturation levels remained stable. Serum iron level decreased during this period.

All anaemia-related laboratory test results worsened in the placebo group.

The following laboratory tests results for efficacy were observed in Tardyferon® and placebo groups:

Visit	Baseline	Randomisation visit (V2)	Final visit (V4)
Serum ferritin (ng/mL)			
Tardyferon® N=11	6.4 (3.1)	29.2 (12.0)	38.9 (18.3)
Placebo N=16	9.6 (11.0)	25.2 (8.4)	13.6 (9.1)
Haemoglobin (g/l)			
Tardyferon® N=11	104.3 (5.9)	133.5 (9.2)	133.3 (13.1)
Placebo N=16	104.5 (9.4)	130.9 (4.9)	126.1 (11.6)
Transferrin saturation (%)			
Tardyferon® N=11	7.3 (2.8)	26.7 (16.2)	24.4 (12.3)
Placebo N=16	10.8 (10.4)	26.9 (16.2)	18.6 (8.6)
Serum iron (µmol/L)			
Tardyferon® N=11	5.5 (1.8)	17.2 (10.3)	13.7 (4.8)
Placebo N=16	8.9 (10.8)	17.9 (8.6)	13.3 (5.5)

Values are mean (SD)

A slight improvement of quality of life was observed in patients treated with Tardyferon® during at least 12 weeks. This effect mainly concerned the physical component of the QoL. This effect was maintained for patients with Tardyferon®. No relevant changes were observed with regard to the mental aspect of the QoL.

The frequency of symptoms of IDA decreased markedly in both groups of treatment between Baseline and V2, and stabilised between V2 and V4.

Safety results

No death was reported during the study. Overall, 32 AEs were reported by 20 patients (29.4%) including 4 patients with SAEs. No patient withdrew from the study because of AEs. Twenty-one AEs were of mild intensity, 8 of moderate intensity and 3 of severe intensity. Relationship with the study treatments was considered as not excluded for 5 AEs in Tardyferon® group, 6 AEs in placebo group and 5 AEs in the open-labelled phase (and unassessable for 1 AE (3.1%) in placebo group.

Most of the AEs (27/32) occurred during the first 12 weeks of treatment with Tardyferon®.

The AEs most frequently reported during the study were gastrointestinal disorders observed in 11 patients among the 20 patients with AEs. Gastrointestinal disorders are AEs known with Tardyferon®.

During the double-blind phase, 5 treatment emergent AEs (TEAE) were reported by 5 patients: 4 TEAEs with placebo (abdominal pain, gastroenteritis, arthralgia and headache) and 1 TEAE with Tardyferon® (gastroenteritis). Values of vital signs remained stable during the study.

Name of Company: Pierre Fabre Médicament	Individual Study Table	(For National Authority Use Only)
Name of finished product: Tardyferon®	Referring to Module 5 of the Dossier	
Name of active substance *(or ingredient)*: Ferrous sulphate	Vol.: .....Page:	

Overall, both treatments administered to the patients during the study were safe and well-tolerated.

Discussion

The aim of this study was to assess the necessity to treat IDA patients long enough and to assess the efficacy of Tardyferon® on serum ferritin and haemoglobin parameters in female patients suffering from iron deficiency anaemia.

Despite repeated efforts to increase the recruitment (4 amendments that modified study duration, inclusion criteria and the primary criterion), the planned number of patients was not reached. Only 67 patients were included at Baseline (vs. 80 patients initially planned) and 29 patients were randomised at V2 (vs. 50 initially planned).

Possible explanations for the difficulties encountered in the recruitment and the data analysis of the study are listed below :

- Too restrictive inclusion criteria that lead to 4 amendments
- Because of the prolonged recruitment period, the reduced number of patients could not be taken into account before randomisation
- The cause of anemia was probably insufficiently treated: while menorrhagia and metrorrhagia were reported as the main reasons of anaemia in this study (61.8% of the patients), only 1 patient received an appropriate teatment against such conditions. Then it can be assumed that Tardyferon® could not entirely compensate the iron depletion due to these concomitant disorders.
- Different population characteristics in France and Argentina :
    1. Menorrhagia and metrorrhagia were especially common in Argentina (85.7% of the patients) compared to France (50%).
    2. Past history of iron deficiency was reported by 30 patients in Argentina (85.7%) versus 4 patients in France (12.1%)
    3. Dietary habits such as the consumption of meat and of traditional drinks rich in polyphenol as Maté (very common in Argentina) are also expected to modify iron metabolism. However such data were not recorded during the study.
- One tablet of Tardyferon® was administered to the enrolled patients in accordance with the SmPC of this product in Argentina. As allowed in France, the increase of dosage of Tardyferon® from 1 to 2 tablets could have normalised more patients taking into account the fact that the Hb level at inclusion was low: 104.3 g/l in average.

Only 27 patients were analysed as ITT population (11 in the Tardyferon® group and 16 in the Placebo group). Consequently, the statistical tests were adapted to take into account the smaller sample size. At these small sample sizes, statistical tests lose some of their power.

Conclusion

In spite of recruitment difficulties and the resulting insufficient number of patients, the adapted statistical analyses show a positive effect of Tardyferon® on haemoglobin, serum ferritin, transferrin, iron levels, IDA symptoms, and the physical aspects of the quality of life.

The difficulties encountered in the recruitment and the data analysis of the study make the interpretation of results difficult.

Date of report Final, 7 December 2011