2. SYNOPSIS

Name of Sponsor/Company:

Defiante Farmaceutica LDA

Product:

SIMVAPUFA

Pharmaceutical Form:

Simvapufa soft-gel capsules (simvastatin 20 mg/1 g omega-3-PUFA)

Dosage:

simvastatin 20 mg/1 g omega-3-fatty acids/day

Compared Drugs:

Sivastin® (Simvastatin 20 mg coated tablets), Sigma-Tau IFR S.p.A., Italy co-administered with Eskim® (omega-3-PUFA1g softgel capsules), manufactured by Cardinal Health, Italy and distributed by Sigma-Tau IFR S.p.A., Italy.

Pharmaceutical Forms:

Coated Tablets (Simvastatin) and softgel capsules (omega-3- PUFA)

Dosage:

Simvastatin 20 mg/day co-administered with omega-3-PUFA 1g/day

Title of the Study:

A randomized multicentre phase II study testing tolerability and efficacy on surrogate endpoints (lipid profile, vascular inflammatory markers and thrombogenesis) of SIMVAPUFA formulation (association of simvastatin and omega-3 fatty acids) in comparison with the administration of the two separate compounds, in patients with acute coronary syndrome and positive biochemical markers of myocardial necrosis.

Study Centres:

The study was conducted in 8 centers in Italy. The centers were: AORN "Antonio Cardarelli" – Napoli, Azienda Ospedaliera "S.Maria di Terni" – Terni, Azienda Ospedaliera Ospedale "S. Anna e S. Sebastiano" - Caserta, Ospedale "S. Giuseppe e Melorio" - Santa Maria Capua Vetere, Ospedale Provinciale "C.G. Mazzoni" – Ascoli Piceno, Ospedale "San Pio da Pietralcina" – Vasto, Azienda Ospedaliera "Pugliese-Ciaccio" – Catanzaro, Ospedale "F. Renzetti" – Lanciano. The latter one never recruited patients.

Publication (reference):

No publication till Dec 30th, 2013

Study Period:

The first dose of study medication was administered on 12 June 2008 and the last dose of study medication was administered on 2 December 2009.

Phase of Development:

Early phase II

Objectives:

The main objective of this study was to show the safety and tolerability of Simvapufa, a new fixed dose combination of Simvastatin and omega-3-PUFA, administered to patients with acute coronary syndrome.

The secondary objective was to compare the effect of Simvapufa and extemporaneous combination of Simvastatin and omega-3-PUFA on lipid metabolism, incorporation of omega-3-PUFA in red blood cell membranes, systemic inflammatory markers, vascular inflammation markers and thrombogenesis in patients with acute coronary syndrome.

Methodology:

This study was an early phase II, multicentre, randomized, parallel group, open, controlled study.

In this trial two treatment groups, SIMVAPUFA (experimental treatment) and SIMVASTATIN+PUFA (active control treatment) were compared.

The study protocol provides a screening visit within the 72 hours from the hospitalization due to an acute coronary syndrome event. During the **Screening Visit** patients were evaluated for inclusion/exclusion criteria, demographic data, patient's history, and a complete physical examination was collected. Screening visit was followed by a **run-in period** of treatment with simvastatin, 20 or 40 mg o.d., lasting for minimum 15 and maximum 30 days from the event. During this period the baseline evaluations were performed. After this period, if eligible, the patients were randomised to enter a treatment period consisting of 16 weeks with an intermediate visit after 4 weeks and a **final visit** at the end of the 16 weeks.

Number of Subjects:

A total of 90 patients (45 patients per group of treatment) were included in the study. Patients with acute coronary syndrome STEMI (ST-Elevation Myocardial Infarction) and NSTEMI (Non-ST-Elevation Myocardial Infarction), occurred within the previous 72 hours with positive biochemical marker of myocardial necrosis (troponin or CK-MB), were enrolled and those who satisfied all other entrance criteria were eligible for the study. Moreover, patients had not to be already under treatment with omega-3-PUFA.

Diagnosis and Main Criteria for Inclusion:

Patients with acute coronary syndrome STEMI (ST-Elevation Myocardial Infarction and NSTEMI (Non-ST-Elevation Myocardial Infarction), occurred within the previous 72 hours with positive biochemical marker of myocardial necrosis (troponin or CK-MB), were enrolled. Moreover, patients were not already under treatment with omega-3-PUFA.

Enrolment Inclusion Criteria:

- Capability of understanding the nature of the trial and willingness to participate as documented by signing the written informed consent form.
- Male or female patients aged between 18 and 75 years.
- Acute coronary syndrome STEMI and NSTEMI, with hospitalization occurred in the previous 72 hours and with positive biochemical marker of myocardial necrosis (troponin or CK-MB).
- No indication for aggressive therapy with statins (simvastatin >40 mg/day).

Randomization Inclusion Criteria:

- Negative pregnancy test for women of childbearing potential only.
- Acute coronary syndrome (STEMI and NSTEMI) with positive biochemical marker of myocardial necrosis (troponin or CK-MB) occurred in the previous 30 day.
- Previous treatment with simvastatin (20 or 40 mg) for at least 15 days during the run-in period.

Enrolment exclusion criteria:

- Congestive Heart Failure (NYHA Class $\geq$ 2) and/or depressed left ventricular function (LVEF < 35%) documented by an echocardiogram performed during the hospitalisation.
- Coronary Artery By-pass Grafting in the previous 6 months.
- Percutaneous Trans luminal Coronary Angioplasty during the hospitalization performed without stent insertion
- Surgery or CABG (Coronary Artery Bypass Graft) or PTCA (PercutaneousTrans luminal Coronary Angioplasty) planned during the study period.
- Patient under treatment with omega-3-PUFA
- Presence of atrial fibrillation
- On-going infectious or inflammatory diseases.
- Known allergy or intolerance to one of the study drugs.

- Chronic concomitant treatment with corticosteroids or NSAID (only low dose (maximum 100 mg) acetyl salicylic acid will be allowed).
- Pregnant or nursing women or women of childbearing potential not taking contraceptive medication (from the previous 3 months and agreeing to continue during all the study period) or not utilising intra-uterine devices.
- Patient having a diagnosis or under investigation for a cancer or a chronic inflammatory disease.
- Patient not agreeing to avoid grapefruit consumption during the all study period.
- Any other clinically significant laboratory or medical condition, which in the opinion of the Investigator makes the patient unsuitable for evaluation in the study.

Randomization exclusion criteria:

- Patient under treatment with anticoagulant therapies.
- Liver disease (SGOT or SGPT levels > 1.5 the upper normal limit.
- CPK level above the upper limit of normality.
- Renal failure (Serum creatinine> 2.5 mg/dl).

Test Product, Dose and Mode of Administration:

Patients assigned to the *Simvapufa* group were treated with 1 soft gel capsule a day, for 16 weeks, taken immediately after dinner. Patients assigned to the control treatment group, were treated with 1 coated tablet of Sivastin® and 1 soft gel capsule of Eskim®, once a day for 16 weeks, taken immediately after dinner.

The investigational treatment was SIMVAPUFA (supplied as soft gel capsules containing 20 mg of SIMVASTATIN plus 1 g of OMEGA-3-PUFA). The control treatment was SIMVASTATIN and OMEGA-3-PUFA (supplied as Sivastin® coated tablets containing 20 mg of simvastatin and Eskim® soft gel capsules containing 1 g of OMEGA-3-PUFA). The study protocol included a screening visit within the 72 hours from the hospitalization due to an acute coronary syndrome event, followed by a Run-In period of treatment with simvastatin, 20 or 40 mg (oral dose) lasting for minimum 15 and maximum 30 days from the event. During the Run-In period, the Baseline evaluation was performed.

Afterwards, the eligible patients were randomized to enter a treatment period lasting for 16 weeks with an intermediate visit after 4 weeks and a final visit at the end of the 16 weeks. Patients were followed-up for 18-20 weeks from enrolment.

Assessment of Efficacy:

The efficacy endpoints were the changes from Randomization to Week 16 evaluated on the following surrogate endpoints:

- Traditional markers of lipid metabolism:
  Total cholesterol, HDL-Cholesterol, LDL-cholesterol, VLDL-cholesterol , APO B and APO A1, Triglycerides
- EPA and DHA levels in plasma and red blood cell membranes
- Systemic inflammatory markers:
  White cell count, C-reactive protein (high sensitivity method)
- Serum glucose and serum insulin
- Vascular inflammation markers (expression of adhesion molecule):
  P-selectin, E-selectin, ICAM-1, VCAM-1
- Immune system activation: Circulating "Tissue Factor"
- Platelet Activation: Urinary 11-dehydro-TxB2, serum TxB2, CD40L
- Oxidative stress: Urinary 8-iso-prostaglandin-$F_{2\text{-alpha}}$ (8-iso-PGF2α)

Assessment of Safety and Tolerability:

Patients were screened during the 72 hours after the acute syndrome event and following hospitalization, in which the informed consent was signed, and vital signs and medical history were collected. Following the screening visit, the patient entered in a run-in period lasting minimum 15 and maximum 30 days from the event, in which vital signs collection, medical history, physical examination, previous/concomitant treatments recording, ECG parameters, standard laboratory testing (for renal and liver functionality), haematology and pregnancy test for childbearing potential women, were performed. The patients had also to undergo a 24 hour dynamic Holter monitoring. During this period, they were treated with simvastatin 20 or 40 mg o.d, according to the investigator opinion. At the end of the run-in period, patients were randomised. Physical examination, vital signs, standard ECG, haematology, renal and liver functionality, pregnancy test and concomitant treatments, were evaluated at each of the visits foresee by the study protocol., ie 4 and 16 weeks. 24 hour dynamic Holter monitoring was performed again in occasion of the week 16 visit. Safety was assessed by evaluating the incidence and severity of adverse events.

Statistical Methods:

All variables were descriptively analyzed per treatment and visit (mean, standard deviation, median, minimum and maximum for continuous variables; frequency distribution for categorical variables) and comparisons between the two treatments were performed by means of the 95% confidence interval for the mean difference. Data changes from Baseline to Week 16 were considered relevant.

The inferential analyses were applied both to the Completer and the Per-Protocol population. The assumption of normality was verified by means of the Shapiro-Wilk test at the 0.1 significance level and constructing a vertical bar chart.

The assumption of homoschedasticity was verified comparing the variances in the two treatment groups by means of the F test at the 0.1 significance level.

If the assumption of normality was verified, but not the homoschedasticity, Satterthwaite test included in the SAS procedure procttest was used.

In case of serious deviation from normality, the two-tailed 95% non-parametric confidence interval for the comparison of medians was used.

The following populations were considered for analysis:

Safety Population, defined as all randomized patients who took at least one study treatment dose. This population were considered for the safety analysis only.

Completer Population (all analyzable patients), defined as all randomized patients who had the baseline and the week16 evaluation. Patients prematurely withdrawn by the study due to an AE but having the week 16 evaluation performed, were included in this population.

Per-Protocol Population (PP), defined as the patients who fulfilled all of the study procedures and all of the inclusion and exclusion criteria of the Study Protocol.

Statistical Analysis:

Demographic and Baseline Characteristics

In order to account for adequate interpretation of the results, treatment balance for demographic and anamnestic variables and for all the other patient characteristics at study entry were verified by means of descriptive statistics (mean, standard deviation, minimum and maximum for continuous variables; frequency distribution for categorical data).

Efficacy Analysis

Evaluation criteria considered for the pharmacological efficacy on relevant biomarkers were finalized to assess the essential equivalence between the experimental and the control treatments; so, for all the surrogate efficacy endpoints the comparisons between the two treatments was performed by means of the 95% confidence interval for the mean difference.

Data changes from baseline to Week 16 were considered relevant.

The inferential analyses were applied both to the Completer and the Per Protocol population.

Results from the Completer Population were considered the primary ones.

Due to the explorative nature of this study, all the outcomes coming from the statistical analyses performed will not be considered inferential; they will have descriptive meaning only.

Safety and Tolerability Analysis

The Safety population was used to evaluate safety and tolerability data.

Physical examinations, ECGs, 24-hour dynamic Holter, vital signs, laboratory tests, adverse events and concomitant medications were considered for the safety and tolerability evaluation.

Categorical variables (ECG abnormalities, physical examination abnormalities, etc.) were analysed with shift tables (baseline vs. final visit), while continuous variables (laboratory parameters, vital signs, etc.) by descriptive summaries.

Laboratory data were analysed with shift tables (baseline vs. final visit), considering each value as being normal/abnormal with respect to the appropriate normal range.

Adverse events were coded using the MedDRA dictionary. Adverse events and all related information were listed by patient. Descriptive statistics were performed stratifying the events by system organ class and preferred term; they were stratified by seriousness and relationship with the study treatment.

Number of Subjects (total and in each arm)

	Screened	Randomized	Completer Population	Per-Protocol (PP)	Safety Population
Total	99	90	89	56	90
Simvapufa		45	44	32	45
Simvastatin+Pufa		45	45	24	45

A total of 99 patients were screened and 90 of them were randomized (and included in the Safety population), 45 (50.0%) to SIMVAPUFA and 45 (50.0%) to SIMVASTATIN+OMEGA-3-PUFA. Of these, 89 patients were included in the Completer population, defined as all randomized patients who had the Baseline and the Week 16 evaluation. Patients prematurely withdrawn by the study due to an AE but having the Week 16 evaluation performed, were included in this population. The all-randomized population comprised 82.2% males and 17.8% females.

Extent of Exposure and Compliance:

The number of days of treatment interruption was computed using the date when the patient temporarily stopped study medication and date when the patient re-introduced the experimental medication.

The exposure to study medication was calculated subtracting the days of treatment interruption to the extent of exposure.

The percentage of time of observed treatment compliance in respect to expected time of treatment was assessed at the end of the study as follows:

$$\text{Compliance} = \frac{100 \cdot [\#\text{days of exposure to study medication}]}{\#days \quad of \quad treatment}$$

Efficacy Results:

At the end of the study, the patients treated with SIMVAPUFA and those treated with SIMVASTATIN+PUFA showed an increasing trend for Total cholesterol, LDL-cholesterol, HDL-cholesterol, APO B, APO A1, and a decreasing trend for Triglycerides. VLDL-cholesterol was nearly constant in both the treatment groups.

EPA and DHA levels in plasma and red blood cell membranes increased in both study groups considering the difference between Week 16 visit and Randomization visit.

Systemic inflammatory markers decreased in both study groups.

Glucose showed an increasing trend in SIMVAPUFA and a constant trend in SIMVASTATIN+PUFA.

With reference to Insulin, patients reported a nearly constant trend.

Vascular inflammation markers showed different trends.

Immune system activation marker (Tissue Factor) showed an increasing trend in patients treated with SIMVAPUFA and a decrease in those treated with SIMVASTATIN+PUFA.

The effect of treatments on all platelet activation markers was similar in the two groups: Urinary 11-dehydro-TxB2 showed a decreasing trend, while serum-TxB2 and CD40L showed a constant trend.

Urinary 8-iso-PGF2α, a marker of oxidative stress, showed a small increase in the SIMVAPUFA group and a strong decrease in the SIMVASTATIN+PUFA group.

The Per-Protocol population results were similar to those shown in the Completer population.

In conclusion, the results obtained with reference to the set of surrogate endpoints seem to confirm the approximate equivalence of the two treatment groups.

Safety results

Adverse Events: In the SIMVAPUFA group 19 patients (42.2%) reported 35 AEs and 15 patients (33.3%) of SIMVASTATIN+PUFA group reported 27 AEs.

The most frequent AEs were: "Gastrointestinal disorders", "General Disorders and administration site conditions", "Nervous system disorders" and "Vascular disorders".

For the SIMVAPUFA group, AEs occurred with an incidence of 10% or greater in 2 organ classes ("Gastrointestinal disorders" and "Vascular disorders"), while for SIMVASTATIN+PUFA group the number of organ classes in which AEs occurred with an incidence of 10% or greater was one ("General Disorders and administration site conditions").

Just 1 patient reported only 1 related AE ("Hepatic enzyme increased" in the "Investigations" class) in the SIMVAPUFA group, but none in the second group, during the study period.
The unique reported related AE was classified as "Non Serious". During the overall period, for the "Mild" category 15 AEs were reported by 10 patients (22.2%) in SIMVAPUFA group and 8 AEs by 7 patients (15.6%) in SIMVASTATIN+PUFA group; for the "Moderate" category 19 AEs were reported by 13 patients (28.9%) in SIMVAPUFA and 18 AEs by 10 patients (22.2%) in SIMVASTATIN+PUFA; for the "Severe" category 1 AE by 1 patient (2.2%) was reported in each group.
Serious Adverse Events: No deaths were reported to the sponsor during the course of the study or at any time since the last dose of study medication.

In the SIMVAPUFA's group 2 SAEs were reported by 2 patients. The System Organ Classes interested were "Gastrointestinal disorders" and "Infections and infestations" with, respectively, "Proctitis" (2.2%) and "Staphylococcal sepsis" (2.2%) as PPTs.

Also in the SIMVASTATIN+PUFA's group 2 SAEs were reported by 2 patients. The System Organ Class interested was "Cardiac disorders" with "Acute myocardial infarction" (2.2%) and "Angina unstable" (2.2%) as PPTs.

Laboratory Data: Laboratory data report little differences between the two treatment groups: regarding to lipidic profile (Table L.14), both groups have shown the same trends and very similar median values both at the screening visit and at Week 4 visit.

Vital Signs and Physical evaluation: The number of patients with non-missing observations was 45 in both groups at Randomization, Week 4 and Week 16. All the physical abnormalities appear in SIMVAPUFA group. At Randomization 1 abnormality was observed (Heart and Vessel), while 4 abnormalities were observed at Week 4 (Eyes, Lungs, Abdomen and Other). No abnormality was found at Week 16.

Conclusions:

The study showed a slight not clinically significant lower tolerability of SIMVAPUFA formulation in patients with an acute myocardial infarction, compared with the two separate compounds. As to efficacy on surrogate endpoints, the two treatments group were approximately equivalent.