2 Synopsis

Trial Registration ID-number: NCT00914589	**IND Number:** 11188 **EudraCT number:** 2008-006324-62 **Japanese Registration number:** 101078
Title of Trial A Multi-Centre, Randomised, Double-Blind, Placebo Controlled Trial on Efficacy and Safety of FXIII Replenishment with Two Different Doses of Recombinant Factor XIII following Cardiopulmonary Bypass Surgery	
Investigators There was one principal investigator for each trial site. Signatory investigator: Dr. [redacted].	
Trial Sites Of a total of 32 initiated trial sites, 30 sites randomised and dosed at least one patient. The country distribution for these 30 sites was as follows (number of sites per country in parenthesis): Canada (5), Denmark (1), Germany (4), Great Britain (3), Israel (2), Italy (2), Japan (4), Spain (3) and the United States (6).	
Publications None	
Trial Period 27 July 2009 - 23 February 2011	**Development Phase** 2
Objectives **Primary Objective**: To investigate whether postoperative treatment with either of two doses (17.5 or 35 IU/kg lean body mass (LBM)) of recombinant factor XIII (rFXIII) is effective in avoiding any allogeneic transfusions in cardiopulmonary bypass (CPB) surgery subjects with a pre-operative transfusion risk score of 2 or 3. **Secondary Objective:** To document the safety profile of rFXIII used as replenishment therapy in cardiopulmonary bypass (CPB) surgery.	
Methodology This was a randomised, double-blind, placebo controlled, multi-centre, multi-national, dose finding, efficacy and safety trial. A total of 409 eligible subjects were randomised equally (1:1:1) to a single dose of either 17.5 or 35 IU/kg rFXIII or placebo, with dosing according to lean body mass. Trial drug was administered intravenously following termination of CPB and dosing with protamine. Administration of trial product was to occur 15-30 minutes after initiation of protamine administration. Trial drug was given in addition to standard antifibrinolytic treatment. A transfusion protocol was developed to reduce inter-centre variation in treatment practice and to standardize management of administration of RBC and blood products, and an external adjudication committee was established to ensure compliance with the transfusion protocol. Patients were monitored closely for 7 days after rFXIII dosing or until discharge (whichever came first), with a follow-up visit 5-7 weeks after trial drug administration. Adverse events were reported from the time of rFXIII dosing until day 7 or discharge (whichever came first), while serious adverse events were reported from the time of rFXIII dosing until the follow-up visit at 5-7 weeks after trial drug administration. An external, independent data monitoring committee reviewed accumulating safety data to ensure the safety of trial subjects.	
Number of Subjects Planned and Analysed A total of 408 dosed patients (136 in each of the three treatment arms) were planned. Of 479 randomised patients, the full analysis set consisted of the 409 patients who received trial drug (128 patients received placebo, and 143 and 138 patients received 17.5 and 35 IU/kg rFXIII, respectively).	
Diagnosis and Main Criteria for Inclusion Patients aged 18–80 years (both inclusive) who were scheduled to undergo cardiac surgery requiring cardiopulmonary bypass were included. Additional inclusion criteria were intraoperative use of an antifibrinolytic agent and that the patient was planned for one of the following surgical interventions: 1) coronary artery bypass grafting (CABG), 2) CABG plus single heart valve replacement/repair, or 3) replacement/repair of a single heart	

valve. Patients were furthermore required to have a pre-operative transfusion risk score of 2 or 3 according to predefined criteria. A number of pre operative exclusion criteria were specified to select a population of patients in whom transfusion avoidance would be a realistic target. Subjects with a history of thromboembolic events apart from coronary artery events or with current thromboembolic disease other than myocardial infarction were excluded. Post-randomisation exclusion criteria (but before trial drug administration) comprised any allogeneic transfusion since hospitalisation; haematocrit < 22% prior to cross-clamp removal; pump run > 3 hours; surgery having included the aortic arch and/or descending aorta or any implantable ventricular assist device; having been subjected to hypothermic (< 28°C) circulatory arrest; perioperative use of fibrin sealants or any investigational agent; severe haemodynamic instability following CPB; and more than 30 minutes elapsing from protamine administration to trial drug administration. Eligibility for trial drug administration according to these criteria was to be evaluated 15 minutes after initiation of protamine dosing.

Test Product, Dose and Mode of Administration, Batch Number

Recombinant FXIII was supplied as a sterile lyophilized powder for injection in single use vials of 15 mg (2505 IU) per vial. Each vial was to be reconstituted in 3.2 mL sterile water for injection, resulting in a rFXIII concentration of 835 IU/mL when reconstituted. Trial product was to be administered via a slow intravenous push at a rate not exceeding 2 mL per minute. rFXIII batch numbers: TR40400 and VR40104.

Duration of Treatment

A single-dose of trial product was administered following termination of cardiopulmonary bypass and dosing with protamine.

Reference Therapy, Dose and Mode of Administration, Batch Number

Each placebo vial was to be reconstituted in 3.2 mL sterile water for injection. Trial product was to be administered via a slow intravenous push at a rate not exceeding 2 mL per minute. Placebo batch number: VR40103.

Criteria for Evaluation – Efficacy

Avoidance of allogeneic transfusion, allogeneic transfusion requirements, need for re-operation, health economic parameters and patient-reported outcomes (health surveys).

Criteria for Evaluation – Safety

Adverse events, vital signs and clinical laboratory tests.

Statistical Methods

All analyses were pre-specified in the statistical analysis plan. For all efficacy endpoints, statistical tests were two-sided and the significance level was set to 5%.

Efficacy: The **primary endpoint** was the percentage of subjects avoiding any allogeneic transfusions (RBC, FFP, platelets, cryoprecipitate, fibrinogen concentrate, clotting factor concentrate) for 7 days post-operative or until discharge, whichever came first. The primary endpoint was analysed by logistic regression adjusting for trial site; transfusion risk score (2 or 3); age group (< or ≥ 65 years); redo, i.e., whether or not the surgical intervention was the patient's second (or more) independent cardiac surgery intervention (yes/no); pre-TDA fibrinogen level; pre-TDA thrombocyte level; type of surgery (elective vs. non-elective); and treatment. A variety of sub-group analyses were prespecified for the primary endpoint.

Based on blinded data review it was found that the distribution of number of units of transfused blood product was clearly not consistent with a Poisson distribution. Most patients received a few units, a few received a lot, and in particular one patient received more than 80 units. For that reason the amount (units) of blood product transfused was analysed using a cumulative logit model on 0, 1, 2, 3, 4 and > 4 units. The analysis was adjusted for covariates as for the primary endpoint. The number of transfused RBC units was analysed similarly. Incidence of re-operation, percentage of subjects avoiding any RBC transfusion and percentage of subjects avoiding massive RBC transfusion (≥ 5 units RBC) were compared between treatment groups using logistic regression analysis adjusting for covariates as for the primary endpoint.

Since it is expected that a certain level of fibrinogen is needed for FXIII to have full effect, it was considered of interest to investigate whether the effect of rFXIII increases with increasing fibrinogen level. This was specifically tested in a separate model for interaction between treatment and fibrinogen with respect to transfusion avoidance. The statistical model was otherwise as specified for the primary endpoint.

Restoration of FXIII plasma activity to > 95% lower normal range as defined from screening data was compared

between treatment groups using logistic regression analysis adjusting for transfusion risk score (2 or 3), age, pre-TDA FXIII plasma activity, and treatment. The plasma concentration of FXIII at each time point was compared among treatment groups using ANOVA adjusting for the same covariates as above.

The impact of the achieved FXIII activity level on avoidance of any transfusions was analysed in a manner analogous to the primary analysis. The only difference was that treatment was replaced by FXIII plasma activity at 30 minutes, 24 hours and 48 hours in the respective analyses.

The interaction effect between pre-dose factor FXIII activity and treatment on avoidance of allogeneic transfusions was analysed in a model similar to the primary analysis but with pre-dose FXIII activity as a covariate both on its own and in interaction with treatment.

The analysis of patient-reported outcomes and health economic parameters are reported separately.

Safety: Thromboembolic events (AMI, cerebrovascular thromboembolic event, peripheral artery occlusion, DVT, pulmonary embolism) and critical events (thromboembolic events, renal dysfunction, re-operation and death) were compared between treatments using logistic regression analysis adjusting for transfusion risk score (2 or 3), age, type of surgery (elective vs. non-elective) and treatment. Treatment emergent adverse events were defined as all adverse events with onset after trial drug administration and until the end of trial from was completed.

Demography of Trial Population

Demographic characteristics were similar across treatment groups. The mean age of the trial population was 68.8 years, and the majority of patients were males, as expected. The proportion of females was slightly higher in the placebo group (22%) than in the active treatment groups (15-17%). The vast majority (91%) of patients were white, reflecting the demography of the participating countries.

Efficacy Results

A total of 409 patients received trial drug and were included in the full analysis set.

- No effect of rFXIII treatment was apparent for the primary endpoint of avoidance of allogeneic transfusion for 7 days post-operative or until discharge, whichever came first. The proportion of patients who avoided transfusion was 64.8% with placebo versus 64.3% and 65.9% with 17.5 and 35 IU/kg rFXIII, which translated to odds ratios close to unity for the comparisons of active treatments against placebo in the applied logistic regression analysis. Trial site (p=0.0001) and type of surgery (p=0.0372) were identified as statistically significant covariates. The following conclusions could be drawn from sub-group and interaction analyses of the primary endpoint of transfusion avoidance: 1) The lack of effect of rFXIII on transfusion avoidance was apparent in all analysed sub-groups defined by pre dose FXIII activity, pre-dose fibrinogen concentration, transfusion risk marker and type of surgery. 2) No interaction between treatment and pre-dose FXIII activity level was identified from logistic regression analysis of transfusion avoidance (p=0.60). 3) There was no impact of FXIII activity level at 30 minutes post-dose on avoidance of allogeneic transfusion (p=0.64). A correlation between FXIII activity level at 24 and 48 hours and avoidance of allogeneic transfusion was identified, but a direct causal relationship is considered unlikely when considering the overall lack of efficacy of rFXIII in transfusion avoidance and the fact that there was no corresponding impact of FXIII activity level in the early post-dose phase.
- Administration of rFXIII quickly and dose-dependently restored FXIII activity to pre-operative levels, and the FXIII activity level was statistically significantly increased in both active treatment groups relative to placebo across all 7-day post-operative time points. The proportion of patients who at 30 minutes post-dose had achieved restoration of FXIII activity to above the lower 2.5% percentile for the distribution of pre-operative (screening) FXIII activity values (0.845 IU/mL) increased from 49% of patients in the placebo group to 85% and 95% of patients in the 17.5 and 35 IU/kg rFXIII groups, respectively, which was highly statistically significant. As expected, the mean concentration of free B-subunit decreased with increasing dose following rFXIII administration, returning to baseline levels within 7 days. This observation supports the assumption that rFXIII in the form of the A_2-dimer combines with available B-subunit to form the A_2B_2 heterotetramer.
- No effects of rFXIII treatment were apparent for the remaining efficacy endpoints (transfusion requirements; avoidance of red blood cell (RBC) transfusion or massive (≥ 5 units) RBC transfusion; percentage of patients undergoing re-operation; patient-reported outcomes; and health economic parameters).

Safety Results

- In total, 2309 treatment-emergent adverse events were reported for 400 patients. The overall incidence of adverse events was similar between dose groups. For all adverse events as well as for serious adverse events, individual

types of events did not occur with noticeably higher frequency in the active dose groups relative to placebo, neither overall nor when considering the subset of events that investigators considered probably or possibly related to trial product. The most commonly reported events (proportion of patients with event given in parenthesis) were pleural effusion (37%), atrial fibrillation (35%), procedural pain (30%), nausea (29%), anaemia (17%), peripheral oedema (16%) and hypotension (16%).

- No overall increase in the incidence of critical events (see table below) or thromboembolic (TE) events as defined for the trial was seen with rFXIII administration, and no consistent trends across treatment groups were noted for the individual types of these events. Thromboembolic events were recorded for 8% and 7% of patients receiving 17.5 and 35 IU/kg rFXIII, respectively, versus 9% for placebo, which includes events of acute myocardial infarction identified by the independent, central ECG laboratory.

Critical Events - Safety Analysis Set

	Placebo		rFXIII 17.5 IU/Kg		rFXIII 35 IU/Kg		Total rFXIII	
	N	(%) E	N	(%) E	N	(%) E	N	(%) E
Number of Subjects	128		143		138		281	
All Events	19	(14.84) 24	24	(16.78) 31	16	(11.59) 20	40	(14.23) 51
Peri-operative Acute Myocardial Infarction*	8	(6.25) 8	10	(6.99) 10	7	(5.07) 8	17	(6.05) 18
Renal dysfunction	9	(7.03) 9	10	(6.99) 10	5	(3.62) 5	15	(5.34) 15
Re-operation	2	(1.56) 3	8	(5.59) 8	4	(2.90) 4	12	(4.27) 12
Cerebrovascular TE Event	3	(2.34) 3	2	(1.40) 2	0	(0.00) 0	2	(0.71) 2
Death	0	(0.00) 0	1	(0.70) 1	1	(0.72) 1	2	(0.71) 2
Deep vein thrombosis	1	(0.78) 1	0	(0.00) 0	1	(0.72) 1	1	(0.36) 1
Peripheral Artery Occlusion	0	(0.00) 0	0	(0.00) 0	1	(0.72) 1	1	(0.36) 1

* Includes events identified from a central evaluation of ECGs and troponin T results by an independent, central ECG laboratory.

- Two adverse events with a fatal outcome were recorded. Both events (myocardial infarction and sepsis) occurred on active treatment but were considered unlikely related to trial drug by investigator and sponsor. The observed postoperative mortality of 0.7% in the active treatment groups is lower than the expected mortality of 4% in the studied population.
- The remaining results on safety laboratory parameters and safety-related examinations did not indicate clinically significant changes as a result of rFXIII administration.

Conclusions

Although administration of 17.5 or 35 IU/kg rFXIII following cardiac surgery and cardiopulmonary bypass dose-dependently restored FXIII activity to pre-operative levels, no effect of rFXIII dosing was observed for the investigated efficacy endpoints of transfusion avoidance, transfusion requirements, need for re-operation, health economic parameters and patient-reported outcomes. No safety issues were identified.

The trial was conducted in accordance with the Declaration of Helsinki and ICH Good Clinical Practice (refer to applicable edition).

The results presented reflect data available in the clinical database as of 11 March 2011.