Prevention of Postpartum Hemorrhage With Tranexamic Acid
10/27/2023

NCT#03287336

Statistical Analysis Plan

Study Population

This was a prospective, single-center, open-label, dose-ranging study that recruited thirty pregnant women at The George Washington University from February 2018 to May 2019. Inclusion criteria were normal renal function (serum creatinine < 0.9 mg/dL), and greater than 34 +0 weeks gestation age with planned cesarean delivery. Patients were excluded if they had a thromboembolic history or hypersensitivity to TXA (or any antifibrinolytic therapies). The study protocol was approved by the institutional review board (IRB# 041737), and written consent was obtained from each patient before entering the study.

Dosing and Blood Sampling

Thirty patients were treated sequentially: the first ten patients received 5 mg/kg of TXA, the second ten received 10 mg/kg of TXA, and the last ten received 15 mg/kg of TXA. Total body weight was used to calculate the actual doses, which were capped at 1000 mg. TXA (Cyklokapron®) was given intravenously at the time of umbilical cord clamping and was infused via an infusion pump for approximately 15 minutes (500 mL/hour of 110 mL volume).

Maternal blood samples were collected from the opposite arm of drug administration through peripheral venous catheters or separate venous blood draws. Three samples per patient were taken at each sample time, including pre-drug, 10 minutes, 30-60 minutes, 1.5-3 hours, 4-6 hours, 7-8 hours, and 24 hours post end-of-infusion. One of the three samples for PK analysis was drawn into a tube containing potassium ethylenediaminetetraacetic acid (Becton Dickinson Vacutainer tubes, K2 EDTA 7·2 mg). This sample was further centrifuged at 3000 xg for 15 minutes to obtain platelet-poor plasma. The other two samples for PD analysis were drawn into tubes coated with sodium citrate (Becton Dickinson Vacutainer tubes, Na Citrate 0·109 M, 3·2%). Samples were stored on dry ice until further analysis.

Biomarkers and covariates

TXA concentration determined by ultra-high-performance liquid chromatography-tandem mass spectrometry was used as the PK metric. A modified rotational thromboelastometry (ROTEM) analysis[9] was used to quantify the anti-fibrinolysis effect of TXA. Maximum lysis (ML), the proportion of clot lysed from the maximum clot (range 0-100%), was chosen as the PD marker to

reflect the lysis potential of a formed clot.[10] Briefly, clotting was initiated with tissue factor and recalcification in the presence of exogenous recombinant tissue plasminogen activator, as described.[11] The bioanalysis details regarding TXA concentration and ML measurements are published elsewhere.[11]

Patient-specific routine clinical data were obtained from the electronic medical record. Demographic data included age, height, weight, and BMI. Pre-delivery laboratory data included serum creatinine level (Scr), platelet count, and hematocrit level. Cockcroft-Gault[12] equation was used to estimate creatinine clearance (CrCl). Patient comorbidities (diabetes and hypertension) were also recorded.

Data Analysis

The population approach was adopted using Pumas v1.0.5 (www.pumas.ai).[13] Non-linear mixed effects modeling with first-order conditional estimation with interaction approach was applied to characterize TXA concentration and its effect on ML. Models were built hierarchically: a base model containing a structural component and a variability component was determined first, followed by the exploration of covariate models to explain the variability in the parameters. The graphic presentation of the data and output was performed using R (version 4.0.2)[14] and ggplot2 (version 3.0.0).[15]

Pharmacokinetic modeling

Base model development: One-compartment and two-compartment models with first-order elimination from the central compartment were explored and compared. One-compartment models were parameterized by clearance (CL) and central volume of distribution (Vc). Additional parameters including inter-compartmental clearance (Q) and peripheral volume of distribution (Vt) were used to parametrize the two-compartment models. Between subject variability (BSV) in all the PK parameters was assumed to be log-normally distributed as shown in Equation 1, where P_i denotes the individual parameter, tvP denotes the population typical parameter and η_i denotes the patient-specific deviation from the population parameters that follows a normal distribution with 0 as mean, ω^2 as variance.

$$P_i = tvP \cdot e^{\eta_i} \qquad \textit{Equation 1}$$

$$\eta_i = (0, \omega^2)$$

Different residual error models were also tested to account for the unexplained residual variability between observations and model predictions, including additive, proportional and combined (both additive and proportional) models. Equation 2 shows the formula of the combined error model. $OBS_{i,j}$ and $IPRED_{i,j}$ are the observation and prediction for the i[th] patient at time j. The difference between these two variables was characterized by a proportional error term $\varepsilon_{1_{i,j}}$ and an additive error term $\varepsilon_{2_{i,j}}$ that follow normal distributions with mean at 0 and variance at ${\sigma_{1or2}}^2$.

$$OBS_{i,j} = IPRED_{i,j} \cdot \left(1 + \varepsilon_{1_{i,j}}\right) + \varepsilon_{2_{i,j}} \qquad \textit{Equation 2}$$

$$\varepsilon_{1or\,2_{i,j}} \sim (0,\ {\sigma_{1or2}}^2)$$

Covariate model development: Covariate models were tested to leverage patient-specific covariates to explain the variability in parameters. Potential relationships were assessed from the graphical diagnosis first. Body weight was included in the model using allometric scaling to reflect the impact of organ size on PK. Given the hydrophilic properties of TXA, lean body mass (LBM) calculated by Boer equation[16] was used in the allometry function to compare with total body weight. To explain clearance variability resulting from different levels of kidney functions, kidney function related covariates (Scr, CrCl) were tested upon clearance using a power function.

The final model was chosen based on statistical diagnosis and graphical diagnosis. A model was determined to be significantly better if it reduced the objective function value (OFV) by more than 3.84 units ($P < 0.05$, for 1 degree of freedom). Covariates included in the final model are physiological or mechanistically relevant, and can explain the parameter variability significantly compared to the base model.

Pharmacodynamic modeling

A sequential population approach was used to perform the TXA concentration-ML analysis. This standard approach has been used previously.[17] Briefly, first the PK was developed, and the individual predicted concentrations from the final PK model were used as an independent variable for the PD modeling. Due to the ex vivo characteristics of the ML bioassay, only direct effect models are reasonable. As shown in Equation 3, the PD model was parameterized in terms

of baseline ML (ML_0), maximal fractional inhibition (Imax), concentration of TXA causing 50% of maximal fractional inhibition (IC_{50}), and hill factor (γ). Variabilities of all the parameters were assumed to be log-normally distributed. Different error models were evaluated and compared to account for the random residual variability. The effect of covariates on PD parameters was evaluated in the same manner as for PK parameters.

Proportional PD effect model: $E = ML_0 \cdot (1 - \frac{Imax \cdot (conc\_pred)^{\gamma}}{IC_{50}{}^{\gamma} + conc\_pred^{\gamma}})$ *Equation 3*

Model Evaluation

Five-hundred bootstrap samples, each containing 30 patients, were generated by randomly sampling with replacement from the original dataset. Final PK and PD models from previous steps were used to re-estimate the parameters for the 500 bootstrap samples. Subsequently, the 95% confidence interval of the bootstrap results was calculated.

Simulations to derive optimal dosing

In vitro studies have found that 10 mg/L of TXA is required to obtain 80% inhibition of fibrinolysis; thus, 10 mg/L was chosen as the PK therapeutic target.[18,19] Based on data obtained from women after non-hemorrhagic deliveries, the normal reference of maximum lysis 17% was chosen as the PD therapeutic target.[20] Individual TXA concentration and ML were simulated using a dose range of 100-1000 mg. The post-hoc individual parameters from the final PK and PD models were employed for the simulations. The proportion of patients who met the PK or the PD targets were determined for each dosing regimen.