REVERSAL OF HEPARIN BY NOVEL SYNTHETIC ANTAGONIST PMX-60056 EXHIBITS A LINEAR DOSE-RESPONSE RELATIONSHIP

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Abstract

PMX-60056 is a small-molecule first-in-class new chemical entity designed to bind to the pentasaccharide group in unfractionated heparin (UFH) and low-molecular-weight heparins (LMWH), and reverse their anticoagulation effects. It has been shown to reverse UFH and the LMWH tinzaparin to date, and without rebound anticoagulation.

When no UFH or LMWH is present, PMX-60056 dosing has been limited by transient hypotension at doses over 0.4 mg/kg in volunteers, but its use for heparin reversal should eliminate this effect as PMX-60056 preferentially binds to heparin. A study with UFH at 70 U/kg and reversal with 0.3 mg/kg PMX-60056 showed full efficacy, and mild blood-pressure reductions in half the subjects that suggested excess PMX-60056.

Therefore, we examined the dose-response, efficacy, and safety at higher doses of UFH as used in cardiac surgery. Hemodynamic effects were recorded with lithium-dilution cardiac output, and blood pressures were continuous intra-arterial measurements. PMX-60056 dose was titrated using activated clotting times (ACTs), and protamine requirements were also estimated by heparin-protamine titrations to determine heparin levels.

350 U/kg of heparin was reversed in 6 normal volunteers, after an initial 10-minute infusion of 0.7 mg/kg PMX-60056 and subsequent smaller doses as needed to normalize ACT. Hemodynamics were unaffected until total PMX-60056 dose exceeded 1 mg/kg – the initial dose of 0.7 mg/kg never produced a change. When a hemodynamic effect did occur (in 3 of the 6), it was initiated by a fall in systemic vascular resistance. Only 1 of these 6 subjects had a clinically significant hypotension, which lasted 15 minutes with pressor agents and limb elevation; this subject was subsequently found to have a past history suggestive of vaso-vagal instability. The other 5 subjects had no appreciable change in blood pressure.

Per protocol the antagonist doses were in discrete amounts, so some overshoot was inevitable. Therefore, the total dose of PMX-60056 to the point immediately preceding full reversal of ACT was correlated with ACT, with initial protamine requirement according to heparin levels, and with estimated residual heparin (calculated from dose given and time elapsed). These data were highly correlated with a straight line from zero: R-squared was 0.90 with initial ACT, 0.96 with initial protamine requirement according to heparin levels, and 0.97 with estimated residual heparin (calculated).

These data suggest that measurements routinely available during cardiac surgery are sufficient for predicting a single reversing dose of PMX-60056 that will safely and effectively neutralize UFH-induced anticoagulation.

After reversal, the ability to re-anticoagulate is sometimes important. The same study also established that reversal of anticoagulation with PMX-60056 did not inhibit subsequent repeat anticoagulation with heparin a few minutes later, which was then also reversed with PMX-60056.

This study was conducted according to cGCP at Duke University's Anesthesiology Research Unit. More results will be forthcoming as the analysis proceeds.

Background

PMX-60056 is a synthetic small molecule (MW=1126) designed to bind to the pentasaccharide group in heparin and all LMWH. This prevents binding to antithrombin, and therefore blocks the anticoagulant effect. The real-time Accelerated Cotting Tme (ACT) indicates the degree of anticoagulation by timing clot formation after addition of a thrombogenic substance; and alternatively, the amount of protamine required to neutralize the heparin in the plasma can be determined by titration. We utilized both these methods.

The dose-limiting effect of PMX-60056 when no heparin product is present can be hypotension.

Objectives

Cardiac surgery typically involves heparin doses of 350 U/kg, resulting in ACT times of 450 seconds or more and a protamine requirement of over 3 mg/kg. The present study was planned to evaluate the safety and efficacy of PMX-60056 with such high levels of anticoagulation, and to provide a dose-response relationship for use in subsequent studies.

Since actual cardiac surgery may sometimes require that anticoagulation be immediately re-established, the ability to do so, and to reverse it soon after, was also evaluated in this study.

Methods

Normal male volunteers were given intravenous unfractionated heparin (UFH) as a bolus. 20 minutes later, PMX-60056 was given as a series of intravenous infusions at a constant rate of 4.2 mg/kg/hr. The first dose was always 0.7 mg/kg (a 10-minute infusion). If the ACT was not within 10 seconds of the pre-heparin baseline, subsequent smaller infusions were given (0.2 mg/kg or less) until that goal was reached. Blood pressures and heart rate were recorded by intra-arterial sensors, and lithium-dilution cardiac output (LiDCO) recording also provided CO and systemic vascular resistance (SVR) on a continuous basis. Anti-Xa effect was measured as well (although not available in real time).

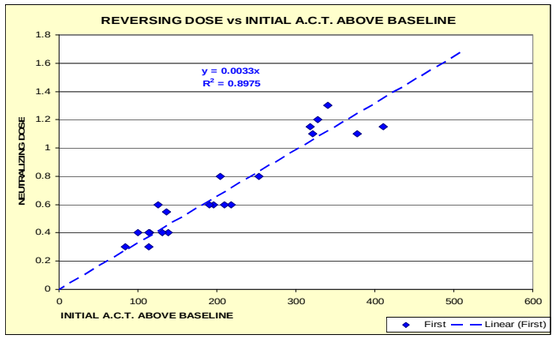
Following this neutralization, anticoagulation was re-established with a second bolus of 100 U/kg UFH, and a second neutralization was begun with PMX-60056 infusions of 0.2 mg/kg or less.

Results

No subject had a blood pressure response to the initial 0.7 mg/kg of PMX-60056. Since it was a titration study, dosing was continued until full neutralization, which usually meant some excess drug was given.

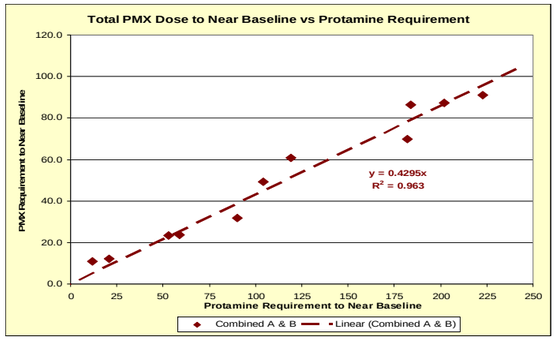
Only one of the six exhibited clinical hypotension, and that was seen only after the 4th infusion.

The total neutralizing dose of PMX-60056 was taken to be the dose exclusive of any excess, with excess defined as doses associated with hypotensive responses (even if not symptomatic). Plotting this against the number of seconds reduction in ACT resulted in the following figure:



The resulting distribution was fit very well by a straight line through the origin, whereby the neutralizing dose (in mg/kg) was predicted by 0.0033 times the number of seconds reduction in ACT desired, with a correlation coefficient of 90%.

Although ACT is used by a majority of clinics in the U.S. to monitor anticoagulation, protamine titration by automated instruments is used in some. We also examined the relationship between the total protamine requirement predicted to reverse the heparin, and the neutralizing dose of PMX-60056. The results are shown here:



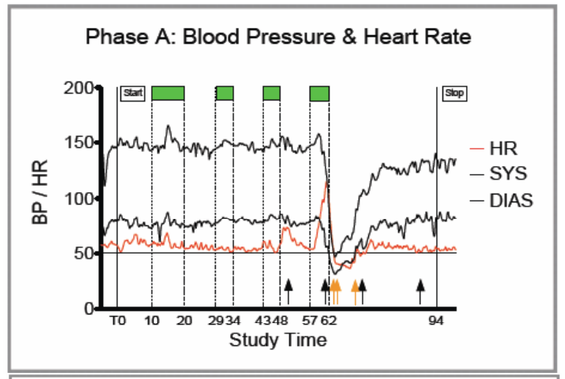
(Excludes Overshoot Past Baseline)

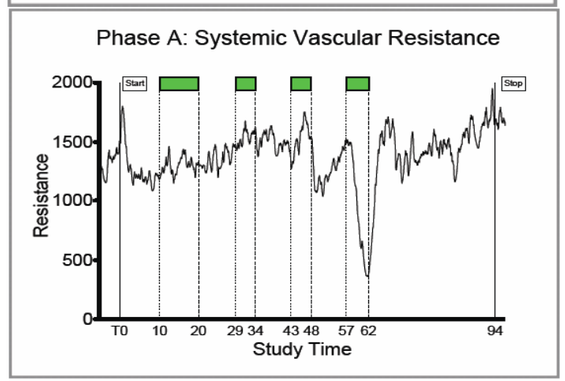
Again the distribution is fit by a straight-line relationship, with a correlation coefficient of 96%: the neutralizing dose of PMX-60056 is given in mg/kg by 0.4295 times the protamine requirement.

The dose to reverse the anti-Xa effect of the heparin was similarly linear.

The LiDCO tracings showed that once all the heparin had been neutralized, the remaining PMX-60056 caused a reduction in SVR. This appeared to be the initial event, which if sufficiently large, would then result in a drop in blood pressure once the heart could not elevate CO sufficiently to offset it.

The following tracings show the one symptomatic hypotensive event. No BP changes occurred until the 4th infusion. A small decrease in PVR after the 3rd infusion was easily compensated, but the next infusion (apparently all excess PMX-60056) produced a large change not compensated, and blood pressure fell:





Summary

PMX-60056 predictably, safely, and completely reverses the anticoagulation effects of high doses of UFH in man. Complete reversal can be obtained without adverse effect, by computing the appropriate dose using a simple linear relationship.

Discussion

A linear dose-response relationship has been demonstrated in these normal subjects, enabling calculation of the appropriate dose of PMX-60056 for completely (or if desired, partially) reversing the anticoagulation produced by unfractionated heparin at high doses. This dose can be calculated based on either ACT or protamine requirement.

PMX-60056 binds to heparin very rapidly, before any peripheral vascular relaxation can occur; but when heparin is exhausted, any excess PMX-60056 results in peripheral vasodilation and possible hypotension. With the dose-response relationship as given here, excess (and therefore hypotension) can be completely avoided.

PMX-60056 is owned and under development by PolyMedix, Inc., Radnor, PA 19087, U.S.A.

(Dr. McAllister is a full-time employee of PolyMedix, Inc. and holds stock options in the corporation)