FACTOR VIIA-CTP (MOD-5014), A NOVEL LONG-ACTING COAGULATION FACTOR PROPOSING AN IMPROVED PROPHYLACTIC AND ON DEMAND TREATMENT OF HEMOPHILIC PATIENTS, EXTENSIVE COMPARATIVE IN–VITRO CHARACTERIZATION IN PREPARATION FOR FIRST IN HUMAN STUDY

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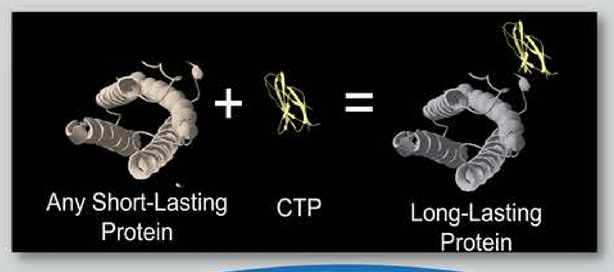
Introduction

PROLOR Biotech is a clinical stage public company developing biobetter long acting versions of existing therapeutic proteins utilizing a technology termed CTP. The technology involves fusion of the C terminus peptide of hCG to one or both ends of the target protein.

The aim of this work was to asses the safety, PK and PD of FVIIa-CTP following administration to rats and monkeys as part of toxicological studies and in preparation to the upcoming FIH clinical study in haemophilc patients .



Methods

FVII-CTP was expressed in CHO cells, purified and activated utilizing a CTP specific purification process. FVIIa-CTP activity was characterized in-vitro and compared to commercial hFVIIa using different in vitro or plasma based assays.

MOD-5014 (FVIIa-CTP) activity was compared to commercial FVIIa in a dose dependent manner by STAClot assay (Stago), FVII chromogenic assay and by PT and aPTT on mass basis . In addition, the physiological interaction and affinity of FVIIa-CTP to co-factors like tissue factor (TF), Factor X and inhibitory proteins like TFPI and ATIII was also characterized.

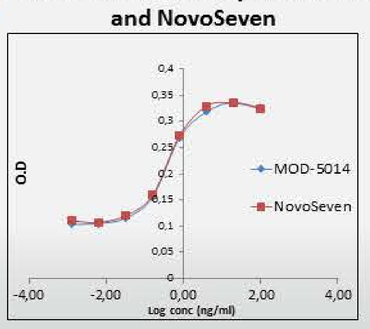
Conclusions

- MOD-5014 activity is comparable to NovoSeven as measured in a qualified STAClot FVIIa assay
- factor X activation by chromogenic assay in the presence of tissue factor (TF), phospholipids and calcium also similir activity of MOD-5014 as reflected by the EC50 value
- Incubating MOD-5014 or NovoSeven at two fixed concentrations with increasing concentrations of TFPI resulted in a dose-dependent reduction in clotting or FXa enzymatic activity. Both compounds demonstrated a very similar de-activation pattern, reflected by clotting % inhibition
- AT III demonstrated a similar inhibition pattern of both MOD-5014 and NovoSeven® when both compounds were spiked with increasing concentrations of AT III.

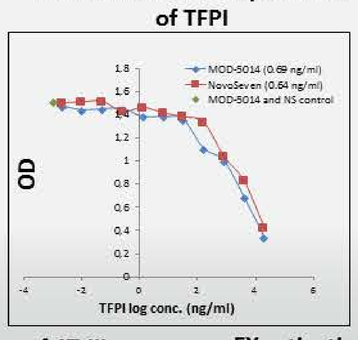
MOD-5014 , following the attachment of CTP, demonstrates similar activity and interaction with co-factors and inhibitors as NovoSeven®. These findings propose that MOD-5014 has a similar mechanism of action as NovoSeven® with the main advantage of extend activity as reflected by its increased half –life, reduced clearance and long term hemostatic effect.

MOD-5014 Characterization by FX activation

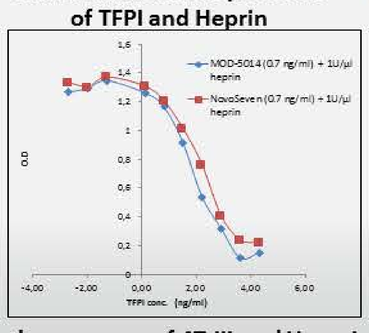
Factor X activation by MOD-5014 and NovoSeven



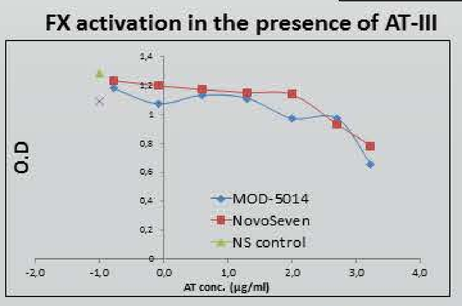
FX activation in the presence of TFPI



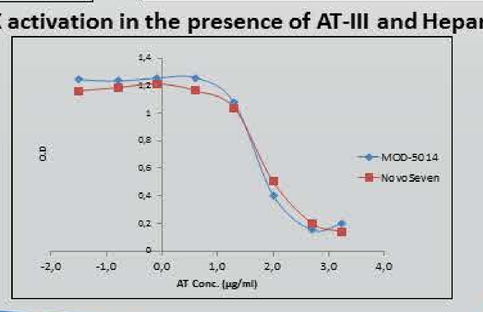
FX activation in the presence of TFPI and Heprin



FX activation in the presence of AT-III

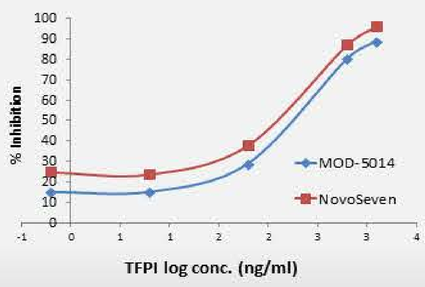


FX activation in the presence of AT-III and Heparin

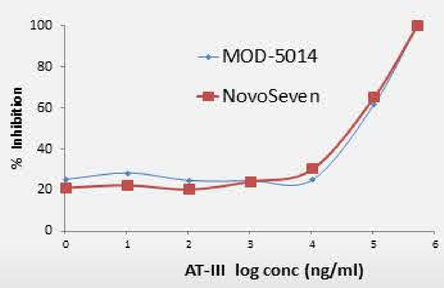


MOD-5014 Characterization by Clotting activity (STAClot)

Clotting Activity in the presence of TFPI



Clotting Activity in the presence of AT-III



Clotting Activity in the presence of Heparin

	NovoSeven		MOD-5014	
Heparin Conc. (U/µl)	Activity (mU/ml)	% Inhibition	Activity (mU/ml)	% Inhibition
0	68.8	NA	56.9	NA
1	0	100	0	0
0.5	0	100	0	0
0.25	0	100	0	0
0.1	2.1	97	2.1	96

PT & aPTT in Deficient Plasma

		PT (sec)		aPTT (sec)	
Test article	**Tested concentration (mg/ml)**	**FVIII Def. Hemophilic Plasma**	**In FVII Def. Plasma**	**FVIII Def. Hemophilic Plasma**	**In FVII Def. Plasma**
MOD-5014	0.5	10.0	10.2	21.0	21.0
	0.1	8.7	8.9	22.8	21.0
	0.02	8.6	8.8	30.9	<21.0
	0.004	8.5	8.8	45.0	24.0
hFVIIa	0.5	No coagulation		No coagulation	
	0.1	8.5	8.7	21.0	21.0
	0.02	8.3	8.6	26.6	21.0
	0.004	8.3	8.6	38.9	22.7
Control (Only Plasma)	0	11.9	No coagulation	87.1	27.5
Normal values		11-13.5		25-35	
		Extrinsic pathway		**Intrinsic pathway**	

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Clotting Factor Concentrates
Hart Gili
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