MEASUREMENT OF FACTOR VIII IN THE PRESENCE OF EMICIZUMAB IS DEPENDENT ON THE CHROMOGENIC ASSAY SELECTED FOR USE

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INTRODUCTION

Prophylaxis of persons with Haemophilia A (PwHA) includes the nonfactor therapy, emicizumab (Hemlibra®); a humanised bispecific antibody with two antigen binding domains (Factor IX/IXa and Factor X/Xa), resulting in FIXa mediated activation of FX, eliciting the haemostatic response[1].

Therapeutic Factor VIII (FVIII) concentrate in PwHA on emicizumab prophylaxis must be measured by chromogenic substrate assay (CSA) with bovine components, avoiding interference by emicizumab[2]. CSA may comprise human, partial bovine or fully bovine components.

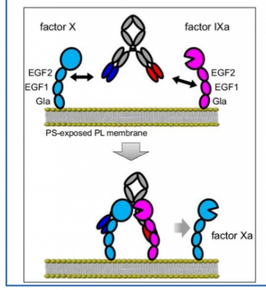

Fig 1. Mechanism of action of emicizumab[3]

AIM

To demonstrate that measurements of post-infusion FVIII concentrate in PwHA are influenced by the chromogenic assay used.

METHOD

- An in-vitro spiking study was conducted where congenital Factor VIII deficient plasma (Technoclone) and a sample pool from PwHA treated with emicizumab (PwHA/emi) were spiked with Elocta to give a range of FVIII concentrations from 0.0-2.0IU/ml.
- Factor VIII levels were measured by the one stage clotting assay (OSCA) and/or by two different CSA methods (Technoclone Technochrom® FVIII:C, partial bovine and HemosIL® Electrachrome™ FVIII, fully bovine) on the ACL Top 750.
- In addition, post-infusion FVIII levels were measured in samples from PwHA on emicizumab prophylaxis and treated with Elocta (PwHA/emi/Elocta) (n=38) using both CSA.

RESULTS

In the in-vitro spiking study, the FVIII levels in plasma spiked with a range of concentrations of Elocta were higher by both CSA compared with the OSCA; (median difference Technochrome® CSA / OSCA was 48%, Electrachrome™ CSA / OSCA was 17%) (Fig 2).

FVIII levels in plasma from PwHA on emicizumab prophylaxis (PwHA/emi) spiked with a range of Elocta concentrations were higher with Technochrome® CSA compared with Electrachrome™ CSA (median difference was 29%) (Fig 3).

Post infusion samples (n=38) from PwHA/emi/Elocta were assayed using both CSA. FVIII levels when measured by the Technochrome® assay were significantly higher than the ElectrachromeTM assay (median difference was 43%, p<0.05) (Fig 4).

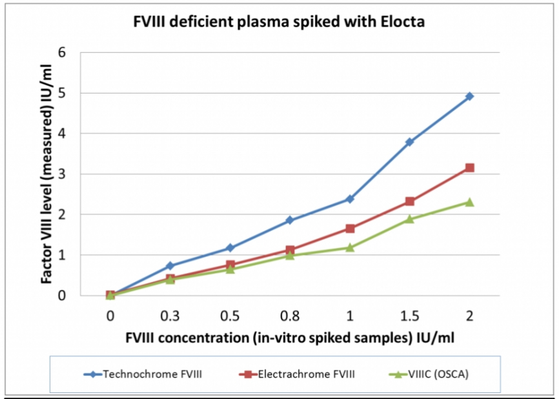

Fig 2. Comparison of FVIII levels in in-vitro spiking study in FVIII deficient plasma spiked with Elocta and measured with CSA and OSCA

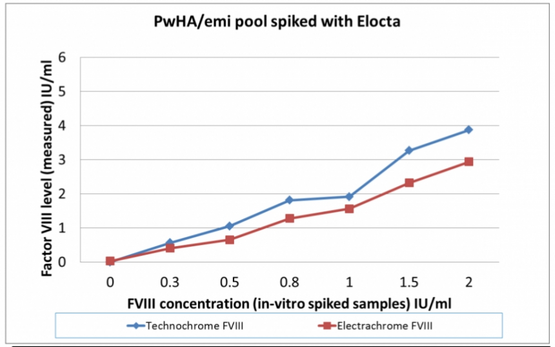

Fig 3. Comparison of FVIII levels in in-vitro spiking study in PwHA/emi pool spiked with Elocta and measured with CSA

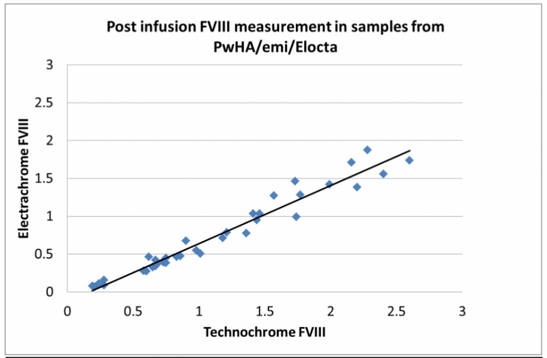

Fig 4. Comparison of FVIII levels from patient samples on emicizumab prophylaxis and treated with Elocta measured by Technochrome and Electrachrome FVIII CSA

CONCLUSIONS

- Emicizumab (Hemlibra®), the Factor VIII mimetic, will interfere with FVIII measurement when assayed using the one stage clotting assay
- Therapeutic FVIII in PwHA on emicizumab prophylaxis must therefore be measured using a chromogenic assay with bovine components to avoid interference by emicizumab in the assay.
- The selection of the appropriate FVIII CSA is vital to ensure appropriate measurement of therapeutic FVIII.
- In this study it was shown that measured FVIII levels will differ depending on whether a fully bovine FVIII CSA or partial bovine CSA is used for the assay.
- FVIII levels with the Technochrome® assay are higher when compared with the Electrachrome™ assay; this difference is magnified for PwHA/emi/Elocta patient samples.
- Measurement of FVIII in PwHA treated with Elocta in addition to emicizumab requires careful consideration when selecting the appropriate CSA to monitor this treatment.

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