



**MEMORANDUM**

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**To** STN 125506/0

**Through** William M. McCormick, Director OCBQ/DBSQC, HFM-680

**Company** Bio Products Laboratory Limited (BPL), Inc.

**Product** Coagulation Factor X (Human), High Purity Concentrate

**Subject** Review Memo for the Release Tests for the Drug Product and the Factor X Potency Assay for the (b) (4)

**Recommendation:** CR Letter—Incomplete Submission

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**Summary**

A new BLA was submitted by Bio Products Laboratory Limited (BPL), Inc. for Coagulation Factor X (Human), STN: 125506, High Purity Concentrate for the treatment of hereditary factor X deficiency. This document constitutes the Review Memo from DBSQC for the following quality control analytical methods and their validations. These methods are used for lot release of the (b) (4) and the drug product, as described below.

(b) (4)

1. Determination of Factor X (Chromogenic Assay)

## Drug Product

1. Identity (by factor X assay) and Determination of Factor X (Chromogenic Assay)
2. (b) (4) Moisture Determination
3. Determination of Total Protein by (b) (4)
4. Determination of Non-Activated Partial Thromboplastin Time (NaPTT) (Clotting Assay)
5. Determination of Fibrinogen Clotting Time (FCT) at (b) (4)
6. Determination of Factor II ((b) (4) Assay)
7. Determination of Factor IX ((b) (4) Assay)
8. Determination of (b) (4)
9. Determination of Citrate
10. Determination of (b) (4)
11. Determination of (b) (4)
12. Sucrose Determination by (b) (4)
13. Determination of Sodium Content by (b) (4)
14. Determination of Chloride
15. Determination of Phosphate
16. Determination of (b) (4)
17. Determination of (b) (4)
18. Characteristics, solubility, appearance of reconstituted solution

The analytical methods provided by the sponsor with the initial submission were not in sufficient details to permit review. This is particularly important for critical assays, including assays for potency and critical impurities. In addition, the sponsor provided very limited information on method validation in the initial submission. They did not evaluate critical validation characteristics for many of the assays, including those for critical assays related to the potency and critical impurities of the product. Furthermore, they did not validate two of the critical (NaPTT and FCT) methods citing that they were compendial methods and did not require method validations.

The review of the submission led to several information requests (IR), asking for large amount of additional information, including SOPs for the critical assays and necessary method validation data. The sponsor submitted requested information in part on 23 October 2013, 17 December 2013, 13 January 2014, and 31 January 2014, and indicated that they would provide the additional data in three additional installments by 28 February, 14 March, and 11 April 2014.

## Conclusion

Having reviewed all information submitted by the sponsor before 31 January 2014, we conclude that the following methods have been described and validated adequately and can be approved for the quality control lot-release tests.

- Determination of Chloride
- Determination of Phosphate
- Determination of (b) (4)
- Determination of (b) (4)
- Characteristics, solubility, appearance of reconstituted solution

However, due to the large amount of incomplete validation results and pending responses to our IRs, we do not have sufficient information to complete our review and could not conclude that other methods can be approved as lot release tests (numbered 1-13 listed in the Summary section of this memo) for (b) (4) the drug product. Thus, it is recommended that the application is not approved at this time but a Complete Response (CR) letter is sent to the sponsor asking them to address the outstanding issues and comments, which are summarized in “CR Letter” section at the end of this memo.

### **Background of Submission**

On July 10, 2013, Bio Products Laboratory Limited (BPL), Inc. submitted an original BLA for Coagulation Factor X (Human) product. The product was granted orphan drug status by the U.S. FDA (No. 07-2469, 08 November 08, 2007) and Fast Track Designation on April 12, 2012 meeting the criteria for Priority Review. The product was developed as a replacement therapy to treat hereditary factor X deficiency, a rare bleeding disorder, for which no specific coagulation factor replacement therapy is currently available. The product contains a human coagulation factor X concentrate indicated for control and prevention of bleeding episodes as well as for peri-operative management in adults and children (aged 12 years and over) with a hereditary factor X deficiency. The product is formulated as a sterile, (b) (4), freeze-dried concentrate of coagulation factor X and is presented as two doses, 250 and 500 International Units (IU). Following reconstitution with sterile water for injection, the two dose sizes contain the same concentration of active ingredient and differ only in the corresponding volumes at the point of use.

### **Submitted Information and Documents**

This is an electronic submission. Information submitted and reviewed includes:

- 125506/0 – Cover letter, dated 10 July 2013
- 125506/0 – 3.2.P.5.1 Specifications (Drug Product)
- 125506/0 – 3.2.P.5.4 Batch Analyses
- 125506/0 – 3.2.P.5.2 Analytical Procedures
- 125506/0 – 3.2.P.5.3 Validation of Analytical Procedures
- 125506/0/14 – Quality Information Amendment; Response to IR dated 21 October, 2013.
- 125506/0/25 – Quality Information Amendment; Response to IR dated 9 December 2013
- 125506/0 – 3.2.P.5.2.3.1 Analytical Procedures: Determination of Factor X
- 125506/0/14 – Quality Information Amendment; Response to IR dated 9 October 2013 – Appendix 1, QCA/00179/14/SOP: (b) (4) Method for the Determination of Factor X
- 125506/0 – 3.2.P.5.3.3.3 Validation of the procedure for determination of Factor X
- 125506/0 – 3.2.P.5.2.5.1 Analytical Procedures: Determination of Factor II

- 125506/0/14 – Quality Information Amendment; Response to IR dated 9 October 2013 – Appendix 1, QCA/00178/12/SOP: (b) (4) Method for the Determination of Factor II
- 125506/0 – 3.2.P.5.3.5.1 Validation of the procedure for determination of Factor II
- 125506/0 – 3.2.P.5.2.5.2 Analytical Procedures: Determination of Factor IX
- 125506/0/14 – Quality Information Amendment; Response to IR dated 9 October 2013 – Appendix 1, QCA/00149/14/SOP: The Operation of the (b) (4) for Single Factor Assays of Factor IX.
- 125506/0 – 3.2.P.5.3.5.2 Validation of the procedure for determination of Factor IX
- 125506/0 – 3.2.P.5.2.3.5 Analytical Procedures: Determination of Fibrinogen Clotting Time
- 125506/0/14 – Quality Information Amendment; Response to IR dated 9 October 2013 – Appendix 1, QCA/00011/14/SOP: The Fibrinogen Clotting Time Test
- 125506/0 – 3.2.P.5.3.3.5 Validation of Method for Determination of Fibrinogen Clotting Time
- 125506/0 – 3.2.P.5.2.3.4 Analytical Procedures: Determination of Non-activated Thromboplastin Time
- 125506/0/14 – Quality Information Amendment; Response to IR dated 9 October 2013 – Appendix 1, QCA/00008/16/SOP: Non-activated Thromboplastin Time
- 125506/0 – 3.2.P.5.3.3.4 Validation of Method for Determination of Non-activated Thromboplastin Time
- 125506/0 – 3.2.P.5.2.1.4 Analytical Procedures: Determination of Moisture by the (b) (4) Method
- 125506/0/14 – Quality Information Amendment; Response to IR dated 9 October 2013 – Appendix 2, QCA/00178/12/SOP: Moisture Determination in Freeze-Dried Products by (b) (4) Method
- 125506/0 – 3.2.P.5.3.1.4 Validation of (b) (4) Moisture Determination
- 125506/0 – 3.2.P.5.3.4.4 Validation of Procedure for Determination of Sucrose
- 125506/0 – 3.2.P.5.2: Analytical Procedure: Determination of (b) (4)
- 125506/0/14 – Quality Information Amendment; Response to IR dated 9 October 2013 – Appendix 2, QAC/00391/13/SOP: Determination of (b) (4)
- 125506/0 – 3.2.P.5.3: Validation of Procedure for Determination of (b) (4)
- 125506/0 – 3.2.P.5.2: Analytical Procedure: Citrate by (b) (4)
- 125506/0 – 3.2.P.5.3.4.3 Validation of Procedure for Determination of Citrate
- 125506/0/14 – Quality Information Amendment; Response to IR dated 9 October 2013 – Appendix 2, QAC/00402/10/SOP: Determination of Citrate and (b) (4) by (b) (4)
- 125506/0 – 3.2.P.5.2.4.1 Analytical Procedures: Determination of chloride
- 125506/0 – 3.2.P.5.3.4.1 Validation of Procedure for Determination of Chloride
- 125506/0 – 3.2.P.5.2.4.2 Analytical Procedures: Determination of Phosphate
- 125506/0 – 3.2.P.5.3.4.2 Validation of Procedure for Determination of Phosphate
- 125506/0 – 3.2.P.5.2 Analytical Procedures: Determination of Total Protein

- 125506/0/14 – Quality Information Amendment; Response to IR dated 9 October 2013 – Appendix 1, QAC/00419/10/SOP: Protein Determination using (b) (4) Assay
- 125506/0 – 3.2.P.5.3.3.2 Validation of Methods for Determination of Total Protein
- 125506/0 – 3.2.P.5.2 Analytical Procedures: Determination of (b) (4)
- 125506/0/14 – Quality Information Amendment; Response to IR dated 9 October 2013 – Appendix 2, QAC/00336/09/SOP: Determination of (b) (4)
- 125506/0 – 3.2.P.5.3.53 Validation of Procedure for Determination of (b) (4)
- 125506/0 – 3.2.P.5.2.4.5 Analytical Procedures: Determination of Sodium
- 125506/0/14 – Quality Information Amendment; Response to IR dated 9 October 2013 – Appendix 2, QAC/00319/12/SOP: Use of (b) (4) to Determine Sodium and (b) (4)
- 125506/0 – 3.2.P.5.3.4.5 Validation of Procedure for Determination of Sodium by (b) (4)
- 125506/0 – 3.2.P.5.2.5.5 Analytical Procedures: Determination of (b) (4)
- 125506/0/14 – Quality Information Amendment; Response to IR dated 9 October 2013 – Appendix 2, QAC/00452/02/SOP: (b) (4) Determination by (b) (4)
- 125506/0 – 3.2.P.5.3.5.5 Validation of Procedure for Determination of (b) (4) by (b) (4)
- 125506/0 – 3.2.P.5.2.5.1.1 Analytical Procedures: Determination of Characteristics, solubility, appearance of reconstituted solution and stability at (b) (4)
- 125506/0 – 3.2.P.5.3.1.1 Validation of methods to determine characteristics, solubility, appearance of reconstituted solution and stability at (b) (4)
- 125506/0 – 3.2.P.5.2.5.1.2 Analytical Procedures: Determination of (b) (4)
- 125506/0 – 3.2.P.5.3.1.2 Validation of procedure to determine (b) (4)
- 125506/0 – 3.2.P.5.2.5.1.3 Analytical Procedures: Determination of (b) (4)
- 125506/0 – 3.2.P.5.3.1.2 Validation of (b) (4) Determination

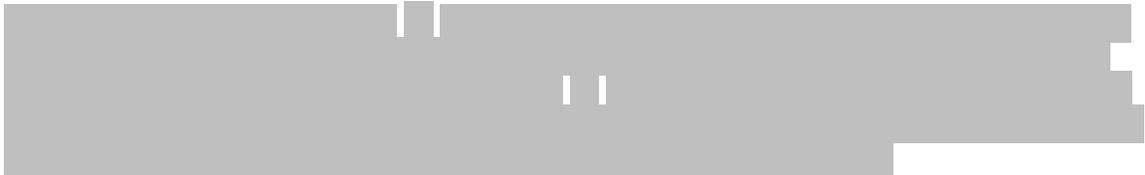
## Review Narrative

### 1. Determination of Factor X

The factor X assay is used for both the identity and potency of (b) (4) the final container product and is based on (b) (4) principle. The specifications for the factor X activity in the final container product are proposed to be 80 – (b) (4) IU/mL and 200 – (b) (4) IU/vial for a 250 IU dose and 400 – (b) (4) IU/vial for 500 IU dose, after reconstitution. The information provided with the original submission on the assay procedure in module 3.2.P.5.2.3.1 of “Analytical Procedure” was insufficient to perform adequate review of the assay method. The SOP (QCA/00179/14/SOP) was provided subsequently in response to our IR.



(b) (4)



Information Requests and Review

The following IR was sent to the sponsor on October 9, 2013. The sponsor responses were received as amendment 125506/14.

- You have not studied specificity of this assay citing that it is a (b) (4). procedure. You need to perform specificity study to demonstrate that the method works for your product without interference from the product matrix. Please provide data to demonstrate specificity of this assay based on analysis of representative product samples and matrices.

Response: Specificity (b) (4)



- You have demonstrated accuracy of the method by testing one standard (3rd International Standard) against another standard ((b) (4))  
Please provide results of accuracy of your method using your product for which this assay is intended. We suggest you evaluate accuracy using spike-recovery method in which unspiked samples at different concentrations and the same samples after spiking with known concentrations (IU/mL) of the standard are analyzed.

Response: The accuracy of the method was determined from (b) (4)

[Redacted]

- Please provide data obtained using representative product samples by multiple analysts to demonstrate intermediate precision of this assay.

Response: The sponsor provided data obtained by (b) (4) analysts over a period of (b) (4) weeks.

Review of Response: The results met acceptance criteria. This is acceptable.

- We could not understand what material you used to evaluate linearity of this assay? Please provide data to show linearity of factor X response in the representative product matrix and parallelism between the standard and sample regression lines.

Response: Linearity was evaluated using the (b) (4)

[Redacted]

(b) (4)

(b) (4)

[Redacted]

Review of Response: The linearity of the standard was adequately demonstrated in the response.

(b) (4)

- Please provide data on the range of the assay based on your results of repeatability, accuracy and linearity studies obtained using representative product samples.

Response: As the method is the (b) (4)

Review of Response: The range should be demonstrated for the product for which the assay is intended to be used. A review of the range study will be completed when received.

- Please provide data to demonstrate appropriate robustness of the assay method using representative product samples for which this assay is intended. The data should demonstrate effect of small deliberate changes of critical method parameters, such as reagent concentration, incubation time, etc.

Response: The robustness of the method was not determined by making small deliberate changes to critical method parameters as the method is from the (b) (4). Additional validation will be carried out using representative product samples as suggested. This validation work will be completed by the 31st January 2014.

The following IRs were submitted on 17 December 2013. The sponsor's responses were received on 13 January 2014.

- Please submit SOP QCA/00037 Reconstitution of Freeze Dried Finished Product for Estimation of Potency.

Response: SOP QCA/00037 version 15, Reconstitution of Freeze Dried Finished Product for Estimation of Potency is presented in Appendix I.

Review of Response: The information submitted provided sufficient details to permit review. The SOP describes the procedure completely and is adequate for its intended purpose.

- Please provide a detailed description of the control, (b) (4), used in the Factor X assay (SOP QCA/00179/15). Please provide data to show how this material is qualified as the control for this assay.

Response: The control was made following SOP QCA/00089 Preparation of in-house Standards and Controls in Biochemistry. A copy of this SOP is presented in Appendix II.

(b) (4)

(b) (4) [Redacted]

Review of Response: SOP QCA/00089/07 Preparation of in-house Standards and Controls in Biochemistry details the procedure used to prepare In-house control

(b) (4) [Redacted].

(b) (4) [Redacted]

The information provided adequately demonstrated the elements required (detailed in 10.1.1 to 10.1.19) to freeze dry the control material. However, details of the testing performed and calculation of potency were not clearly described.

- Please revise SOP QCA/00179/15 to include assay validity criteria and submit for review.

Response: SOP QCA/00179 version 16 (Date effective 18 Nov 13) is provided and states the assay validity criteria in the calculation section 11. Refer to Appendix III for copy of the revised SOP.

Review of Response: SOP QCA/00179 version 16 (Date effective 18 Nov 13) was revised to update sample dilution and remove reference to (b) (4) [Redacted]. The assay specification remains as outlined in SOP QCA/00179/14.

(b) (4) [Redacted]

Based on the information provided, this reviewer concludes that the validity criteria for the sample and control are acceptable. However, this recommends that the validity/acceptance criteria for the standard should be expressly stated.

Conclusion

We are unable to conclude that the method is suitable for lot release of (b) (4) [Redacted] the final container drug product due to outstanding IRs.

**2. Determination of Total Protein by (b) (4)**

Total protein was determined as a release test of the drug product. The proposed specification is (b) (4). The sponsor provided the SOP (SOP QAC/00419/10) for the assay procedure and the validation report (3.2.P.5.3).

Method

(b) (4)

Method Validation

(b) (4)

(b) (4)

[Redacted]

Information Request

The following IR was sent to the sponsor on October 9, 2013. The sponsor responses were received as amendment 125506/14.

- What protein was added to the Factor X product for the accuracy study?

Response: Protein (b) (4) [Redacted]  
[Redacted] before being added to the final product.

Review of Response: The response is adequate.

- Please provide results obtained using representative product samples by multiple analysts on multiple days to demonstrate intermediate precision of this method.

Response: The data provided in the BLA (reference 3.2.P.5.3.3.2.3) to demonstrate intermediate precision was obtained using a number of analysts. Table of Intermediate Precision results for Final Product carried out by (b) (4) Analysts over (b) (4) period. BLA reference 3.2.P.5.3.3.2.3 (Table 3.2.P.5.3-T37)

(b) (4)

Review of Response: The response is adequate.

- Please provide data of the linearity study using representative final container product samples and to demonstrate parallelism between the linear regression fits for the final container product samples and the standard protein used in the linearity study.

Response: Validation was previously carried out in accordance with ICH guidelines using standard protein. Additional validation as requested using Factor X final product will be carried out. This validation work will be completed by the 31st January 2014.

- Please recalculate range of this assay based on your evaluation of precision, accuracy and linearity studies and submit your results.

Response: As there is insufficient data to recalculate range, additional validation as requested using Factor X final product will be carried out. This validation work will be completed by the 31st January 2014.

- Please provide detail of composition of the Internal Quality Control sample used in robustness study? How is this sample related to the final container product? If this product is not representative of the final container product, please provide data for robustness studies performed with representative final container product samples.

Response: The composition of IQC sample: (b) (4)

[Redacted]

(b) (4)

(b) (4)

**ACCEPTANCE CRITERIA:**

(b) (4)

Review of Response: The specification for this assay is (b) (4)

- You have performed robustness study using an Internal Quality Control sample that contains about (b) (4) of protein, which is much higher than your proposed specification limit (b) (4). Robustness should be evaluated at a concentration of protein at or below the specification limit. Please submit robustness data by analyzing representative product samples that contain appropriate concentrations of protein.

Review of Response: We have not received response to this IR yet.

**Conclusion**

This method is validated by the sponsor as a quantitative assay. The method is described in sufficient details. However, the validation report has significant deficiencies, which were brought to the attention of the sponsor through IRs. The review could not be completed due to the pending IRs.

**3. Moisture Determination in Freeze-Dried Products by (b) (4)  
Method**

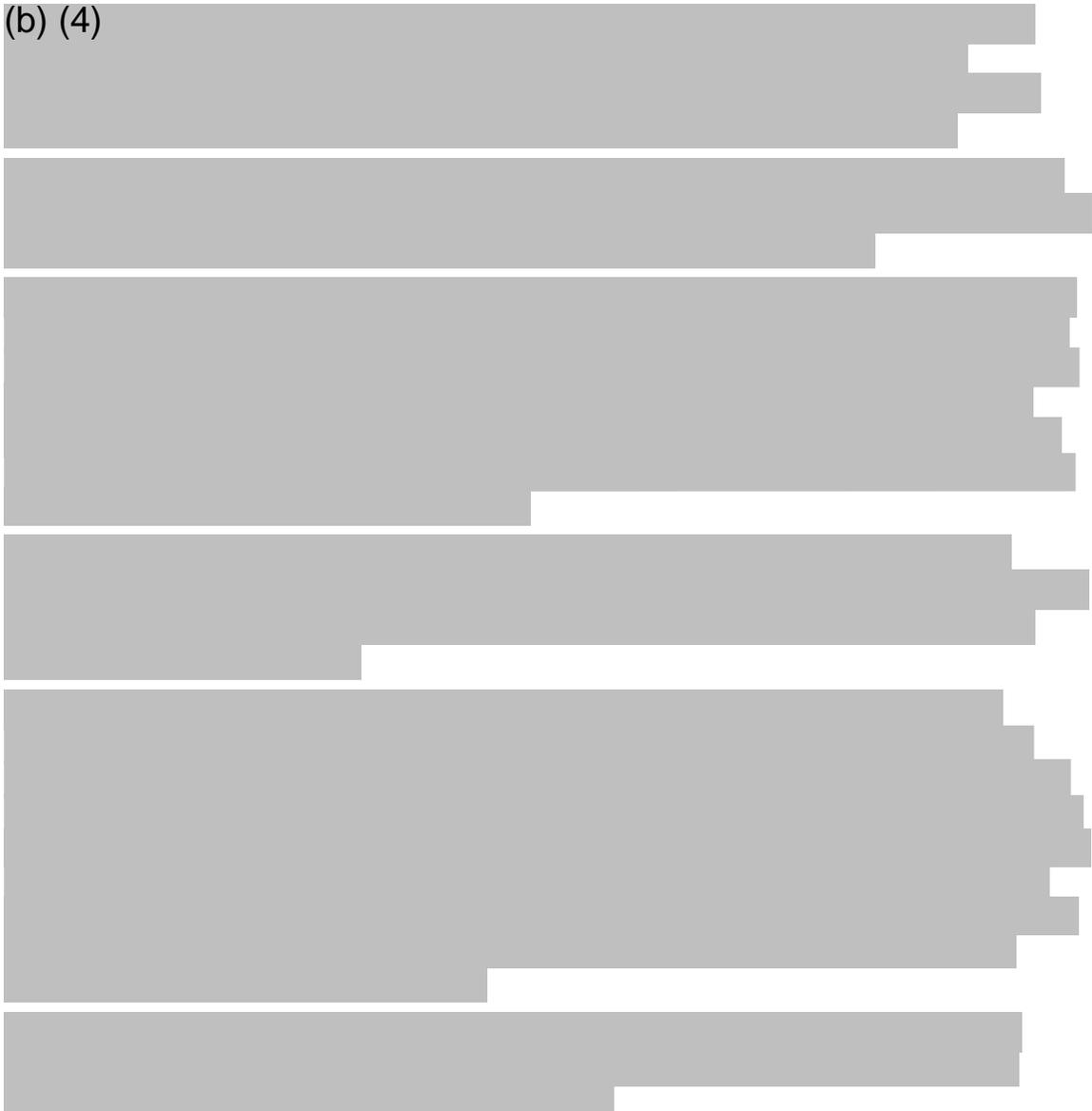
The residual moisture of the final drug product, human coagulation factor X, was measured by (b) (4) method. The proposed specification for the residual moisture in the drug product is (b) (4). No detailed information was provided on this method in the original submission, except the statement that “The method is as described in the current edition of the (b) (4)”. The SOP (QAC/00331/18/SOP) was provided in response to our IR. The sponsor also provided validation data in the original submission.

**Method**

(b) (4)

**Method Validation**

(b) (4)



Information Requests

- For specificity study, you mentioned, “Specificity was not examined as substances known to interfere in the (b) (4) assay are not present in FACTOR X.” Please provide data using representative product samples to substantiate your statement.

Response: The sponsor indicated that the result to be completed by Jan 31, 2014.

- Please provide data for the recovery using representative product samples to demonstrate accuracy of the assay. We suggest that you spike your sample with different known amount of water and then assay both unspiked and spiked samples to calculate recovery.
- Please provide data using representative product samples to demonstrate repeatability, intermediate precision (multiple analysts, multiple days), linearity, and limit of quantitation of the assay.

- Please provide robustness study data using representative product samples, which address the effect of small variations of critical method parameters.

Review of the responses to IR# b, c, and d:

As for Information requests b, c, and d, the sponsor agreed to reassess the assay's Accuracy, Precision, Linearity, LOQ, and Robustness as suggested, and the data is to be completed by Jan 31, 2013, and the decision is pending on the expected data.

Conclusion

Based on the limited information provided by the sponsor, the reviewer could not come to the conclusion that the assay has been validated for its intended use.

**4. (b) (4) Method for the Determination of Factor II ((b) (4) Assay)**

The assay measures indirectly the Factor II (FII) activity present in the final container product as a product-related impurity. The method adapted from (b) (4) is described in detail in the SOP entitled (b) (4) Method for the Determination of Factor II, document number QCA/00178/12/SOP. The proposed specification for the residual moisture in the drug product release is  $\leq 1$  IU/mL.

Method

(b) (4)



### Method Validation

A formal validation according to a protocol with pre-determined acceptance criteria was not submitted due to a claim by the sponsor that the method is compendial as described in (b) (4), and does not need validation. As a result, the following IR was sent to the sponsor. The points of contention and the response to the IR were received on October 21, 2013 and are outlined below.

### Information Request and Review

The following IR was sent to the sponsor on October 9, 2013. The sponsor responses were received as amendment 125506/14.

- You have not studied specificity of this assay citing that the assay is performed as described in (b) (4). You need to perform specificity study to demonstrate that the method works for your product without interference from the product matrix. Please provide data to demonstrate specificity of this assay based on analysis of representative product samples.

Response: Work will be completed by 31 January 2014.

- You have demonstrated accuracy of the method by testing one standard (3rd International Standard) against another standard ((b) (4)). Please provide results of accuracy of your method using your product for which this assay is intended. We suggest you evaluate accuracy using spike-recovery method by analyzing unspiked samples at different concentrations and the same samples after spiking with known concentrations (IU/mL) of the standard in such a way that the total concentrations of factor II in the samples are between the Quantitation Limit of the assay and at or below the proposed specification limit.

Response: Work will be completed by 31 January 2014.

- Please provide data on the assessment of Quantitation Limit from analysis of representative samples of your product for which the assay is intended.

Response: Work will be completed by 31 January 2014.

- Please provide data, including your linear regression plots, to demonstrate parallelism between the linear regression fits for the final container product samples and the standard at different factor II concentrations.

Response: The linearity is evaluated using the standard only. The sponsor does not plan to study linearity of the sample and show parallelism between sample and the standard.

Review of Response: We do not think that this is not acceptable. DBSQC sent a follow-up IR on 03 December 2013.

- Please reevaluate the range of the assay based on your results of repeatability, accuracy and linearity data obtained using representative product samples and submit results.

Response: Work will be done

Review of Response: The sponsor provided no timeline. DBSQC sent a follow-up IR 03 December 2013 asking for the timeline.

- Please provide data to demonstrate appropriate robustness of the assay method using representative product samples for which this assay is intended. The data should demonstrate effect of small deliberate changes of critical method parameters, such as reagent concentration, incubation time, etc.

Response: Work will be completed by 31 January 2014.

#### Conclusion

Due to the pending IR, we can not complete review and conclude if the method can be approved as a lot release test for the drug product.

### **5. Determination of Factor IX ((b) (4) Assay)**

The test is adapted from the current (b) (4) method for the assay of human coagulation Factor IX (FIX), which is present as a product-related impurity. However, the sponsor has not (b) (4). The specification for the final container product is Not Greater Than (NGT) 1 IU/mL. In the initial submission, a summary of the procedure was provided. However, the information was insufficient to permit complete review. DBSQC submitted an IR for the SOP for the test procedure. In response, the sponsor submitted the SOP entitled (b) (4)

SOP

QCA/00149/15. This replaces the SOP number QCA/00149/14 which was referred to in the initial submission.

#### Method

(b) (4)



#### Method Validation



### Information Request and Review

The following IR was sent to the sponsor on October 9, 2013. The sponsor responses were received as amendment 125506/14.

- You have performed specificity study using factor IX (b) (4) concentration. However, your specification limit is NGT 1 IU/mL. Specificity should be evaluated at about the concentration of factor IX (analyte) at the specification limit. Please submit specificity data by analyzing samples containing factor IX at the specification limit to show that the results are not affected by the product matrix, including the presence of large concentration of factor X.

Response: Specificity was determined from a factor IX sample at (b) (4)

Review of the Response: The sponsor has demonstrated specificity of the assay for a factor IX product but not for the factor X final container product for which this assay is intended to be used. Specificity is the ability to assess unequivocally the analyte in the presence of components which may be expected to be present in the product. This cannot be achieved by demonstrating specificity of the assay using another product because the matrix of factor IX in the factor IX product is different from that in the factor X product. The ability to assess an analyte should be at the concentration at which the analyte is expected to be present in the product. Therefore, presenting results at concentrations (b) (4) fold higher than what is present in the actual product does not demonstrate specificity. The sponsor's response is not acceptable.

- You have demonstrated accuracy of the method by testing one standard (3rd International Standard) against another standard ((b) (4)). Please provide results of accuracy of your method using your product for which this assay is intended. We suggest you evaluate accuracy using spike-recovery method by analyzing unspiked samples at different concentrations and the same samples after spiking with known concentrations (IU/mL) of the standard in such a way that the total concentrations of factor IX in the samples are between Quantitation Limit and the proposed specification limit for the product.

Response: The sponsor will perform additional validation work using spiked samples as suggested, which will be completed by January 31, 2014.

- Please provide data obtained from representative samples by multiple analysts to demonstrate intermediate precision of this assay.

Response: The data provided in the BLA (reference 3.2.P.5.3.5.2.3) to demonstrate intermediate precision was obtained using a number of analysts, the number of analysts is recorded just beneath the results tables and as such may not have been obvious as to what it referred to.

Review of the Response: The information was provided in a small font as a foot-note to the table and no details was provided. The sponsor's response is acceptable and no further action is required.

- Please provide data on the assessment of Quantitation Limit from analysis of representative samples of your product for which the assay is intended.

Response: Quantitation Limit was determined from the standard as this provided a high degree of confidence in the potency obtained. Additional validation will be carried out using representative product samples. This validation work will be completed by the 31st January 2014.

- Please provide data, including your linear regression plots, to demonstrate parallelism between the linear regression fits for the final container product samples and the standard at different factor IX concentrations.

Response: BPL submitted a representative plot of the standard over the range (b) (4)

[REDACTED] . PBL did not give a time-line for when they will complete DBSQC requests.

Review of Response: An IR has been submitted asking the sponsor of the time-line.

- Please reassess the range of the assay based on your results of repeatability, accuracy and linearity data obtained using representative product samples and submit results.

Response: Range was determined from the standard as this provided a high degree of confidence in the potency obtained. Additional validation will be carried out using representative product samples. This validation work will be completed by the 31st January 2014.

- Please provide data to demonstrate appropriate robustness of the assay method using representative product samples for which this assay is intended. The data should demonstrate effect of small deliberate changes of critical method parameters, such as reagent concentration, incubation time, etc.

Response: BPL will carry out additional validation work to determine the robustness of the assay from normal variation of multiple assays runs using a final product. This validation work will be completed by January 31, 2014.

### Conclusion

Due to the pending IR, we can not make a decision if the method can be approved as a lot release test for the drug product.

## **6. Determination of Non-Activated Partial Thromboplastin Time (NAPTT)**

The test is adapted from the test for '(b) (4)' described in (b) (4). The specification for the final container product is (b) (4) of the product sample. The method is described in

the SOP number QCA/00008/16/SOP, which was not included with the initial submission but was submitted in response to our IR on 21 October 2013.

#### Method

(b) (4)

However, the SOP has several issues as listed below.

- It provided an identification number for the positive control but did not include a description.
- No information was provided as to how this positive control was calibrated (qualified).
- (b) (4)

An IR was submitted to address these deficiencies.

#### Method Validation

(b) (4)

#### Information Request and Review

The following IR was submitted to the sponsor on 9 October 2013:

- You have not validated NaPTT assay citing that it is a compendial procedure, taken from (b) (4). We do not consider the assays cited in (b) (4) as compendial assays. Furthermore, this is a test for the activated components of the coagulation mechanism, which is a critical test associated with product quality and safety. Consideration of such an assay is precluded without appropriate method validation. Please validate this assay using representative final container product samples and submit the validation report.

Response: In response, the sponsor proposed to perform specificity study only for method validation by showing changes to the clotting times after spiking activated factors into the final product. However, the sponsor did not provide any timeline when they would submit the validation data.

Review of Response: We do not agree with the sponsor's position. The method measures a safe level of thromboplastin based on the clotting time and is, therefore, a

quantitative method, in that the measurand is clotting time (and not concentration). The fact that the proposed specification is (b) (4) does not make it a qualitative (or semi-quantitative) method. Therefore, it should be validated as a quantitative method, however, keeping in mind that the measurand is time. A follow up IR was submitted on 13 December 2013.

- In response to the questions 5 and 6 of our previous IR regarding NaPTT and FCT assays, respectively, you responded that they are qualitative methods. Therefore, you will evaluate specificity only to validate these methods. We do not agree. You are assessing the impurity levels by measuring the clotting time in both assays – your reportable results are time for both assays. Therefore, the methods are quantitative. You need to validate both methods as quantitative methods. Thus, in addition to specificity, please provide data on evaluation of other applicable validation characteristics in terms of the reportable results.

We have not received the response from the sponsor prior to 31 January 2013.

In addition, the following IR was sent to the sponsor on 17 December 2013. The sponsor responded on 13 January 2014, which were reviewed as discussed below.

- In your SOP QCA/00008/16: Non Activated Partial Thromboplastin Time (NAPTT), a NAPTT control, (b) (4) is mentioned in sections 8.1.3 and 8.6. Please provide a detailed description of this control. Please provide data to show how this material is qualified as the control for this assay.

Response: The control was made in 2004 following SOP QCA/00089 Preparation of in-house Standards and Controls In Biochemistry. Please refer to Appendix II for copy of SOP.

(b) (4)

The calibration was carried out in January 2004 (Data presented in Appendix IV - Response 4 NAPTT control (b) (4) data).

A control is run with every assay, and performance continually monitored. The control is considered appropriate for use provided the overall mean remains within  $\pm 2$  standard deviations of the original mean.

Review of the Response: The description of the control material provided in the response is sufficient. However, based on our analysis of the (b) (4)

- In SOP QCA/00008/16, section 12 instructs the analysts to see the Biochemistry Manager/Deputy if the result from (b) (4) is not within (b) (4) standard deviation

from the mean. What is the implication if the result is outside of these limits? How will an analyst know if the results are within these limits?

Response: The operator will review the control chart and if the control result is not within (b) (4) standard deviations of the control chart mean, the assay would be considered invalid, and the results would not be used.

Review of the Response: In response to our previous IR comment, the sponsor responded that the control is considered appropriate for use provided the result for the control remain within (b) (4) standard deviations of the mean at each dilution. However, the sponsor responded here that the assay would be considered invalid if the control results are outside of (b) (4) standard deviations. These are contradictory statements. The sponsor needs revise the SOP to include appropriate and justifiable assay validity criteria in the SOP.

- Please revise your SOP QCA/00008/16: Non Activated Partial Thromboplastin Time (NAPTT) to include the assay validity criteria and submit for review.

Response: SOP QCA/00008 version19 (Date effective 02 Dec 13) is provided and states the assay validity criteria in the calculation section 11. Refer to Appendix V for copy of the revised SOP.

Review of the Response: The response refer to version 19 of the SOP, however, the SOP that the sponsor submitted has the number QCA/00008/18/SOP. Although the issue date on the document is 02 DEC 2013, it seems that the version number of this document is 18. The only assay validity criterion we found in document QCA/00008/18/SOP is, “The test is not valid unless the coagulation time measured for the (b) (4) .” It did not specify the (b) (4) the qualification data presented in Appendix IV of the submission. As discussed above, the assay validity criteria should be (b) (4) . The sponsor should revise the SOP to include the assay validity criteria based on the control calibration (qualification) results.

- Please explain the purpose of the (b) (4) dilution of the sample in the NaPTT test, as described in section 10.3 of your SOP QCA/00008/16. How will the result from this dilution be used? If this dilution has no meaningful purpose, please revise your SOP to delete this dilution.

Response: Performing the test at (b) (4) ) facilitates detection of activated clotting factors which may otherwise be masked, either by over-dilution or by the inhibitory effect of other components which can be reduced by dilution. The (b) (4) adopted by BPL for activated clotting factors specifies that for each of the dilutions, the coagulation time is (b) (4) . As a test of global haemostasis, this acceptance criterion has been adopted for FACTOR X.

The (b) (4) result is used for information purposes only.

Review of the Response: If (b) (4) are necessary to ensure that there is no masking, due to either over dilution or matrix inhibition, results from (b) (4) dilutions should be reportable results and included in the lot-release protocol.

- Please submit your SOP QCA/00071, which describes the procedure for (b) (4).

Response: The (b) (4) is mentioned in QCA00008/SOP as the SOP covers all the products tested for NAPTT ( IX, X and XI). (b) (4) is only carried out on samples (b) (4), which the Factor X does not. QCA/000071/SOP is the Factor XI Sampling and Testing SOP and is unrelated to Factor X.

For clarification, BPL will update QCA00008/SOP to state that Section 4.1 only refers to FXI concentrate.

Review of the Response: Section 4.1 of the SOP that the sponsor included in the response package (QCA/00008/18/SOP) does not reflect this revision. The SOP should be revised to include this clarification.

### Conclusion

Due to the pending IR, we can not make a decision if the method can be approved as a lot release test for the drug product.

## **7. Determination of Fibrinogen Clotting Time (FCT)**

The test measures the time required for thrombin (product-related impurity) in the test sample to clot a known concentration of fibrinogen. The specification for the final container product is (b) (4). The method is described in the SOP number QCA/00011/14/SOP, which was not included with the initial submission but was submitted in response to our IR.

### Method

The method is based on the method described in (b) (4). However, the sponsor introduced (b) (4)

However, the SOP does not include assay validity criteria.

### Method Validation

The sponsor felt that validation of the method was not necessary because they followed the method described in (b) (4). We do not agree that a method described in (b) (4) does not need validation. The method should be validated for the product.

### Information Request and Review

The following IR was submitted to the sponsor on 9 October 2013:

- You have not validated FCT assay citing that it is a compendial procedure, taken from (b) (4) . We do not consider the assays cited in (b) (4) as compendial assays. Please validate this assay using representative final container product samples and submit the validation report.

Response: In response, the sponsor proposed to perform specificity study only for method validation by showing changes to the clotting times after spiking activated factors into the final product. However, the sponsor did not provide any timeline when they would submit the validation data.

Review of Response: We do not agree with the sponsor's position. The method measures a safe impurity level based on the clotting time and is, therefore, a quantitative method, except that the measurand is clotting time (and not concentration). The fact that the proposed specification is (b) (4) hours does not make it a qualitative (or semi-quantitative) method. Therefore, it should be validated as a quantitative method keeping the perspective of time as the measurand in mind. A follow up IR was submitted 13 December 2013.

- In response to the questions 5 and 6 of our previous IR regarding NaPTT and FCT assays, respectively, you responded that they are qualitative methods. Therefore, you will evaluate specificity only to validate these methods. We do not agree. You are assessing the impurity levels by measuring the clotting time in both assays – your reportable results are time for both assays. Therefore, the methods are quantitative. You need to validate both methods as quantitative methods. Thus, in addition to specificity, please provide data on evaluation of other applicable validation characteristics in terms of the reportable results.

We have not received the response from the sponsor prior to 31 January 2013.

In addition, the following IR was sent to the sponsor on 17 December 2013.

- Please revise your SOP QCA/00011/15: The Fibrinogen Clotting Time Test to include the assay validity criteria and submit for review

Response: The sponsor responded that the SOP will be updated to include the assay validity criteria and submitted by 28th February 2014.

### Conclusion

Due to the pending IR, we can not make a decision if the method can be approved as a lot release test for the drug product.

## 8. Determination of (b) (4)

[Redacted content]



Sodium Chloride, Water for injections (bulk), (b) (4)

[Redacted]

Conclusion

The review of validation report could not be completed due to the pending validation data and information requests.

**10. Sucrose determination by (b) (4)**

The sucrose content in the final container drug product was measured using an (b) (4) method with (b) (4). The specification for sucrose in the drug product was set to be: (b) (4). The sponsor provided a brief description of test method, a brief description of the validation study, and a summary of the validation results in the submission.

Method

As described in the submission, sucrose in the final drug product was (b) (4). Although the principle of the assay was provided, no detailed information regarding the assay's procedures, system suitability/validity criteria, and result interpretation was provided in the submission.

Method Validation

(b) (4)

[Redacted text block]

Information Requests

- Please provide results obtained by multiple analysts on multiple days using representative final container product samples to demonstrate intermediate precision.

Review of the response In the sponsor’s response, it was clarified that the intermediate precision was validated by (b) (4) analysts over (b) (4) weeks period. Although the provided data table is not clear, we found that the described validation scheme for intermediate precision was acceptable and the pre-set validation criteria were met, and considered the intermediate precision of the assay was validated.

- Please provide a detailed description of composition of the Internal Quality Control sample used in robustness study of this assay? How is this sample related to the final container product? If this sample is not representative of the final container product, please provide data for robustness studies performed with representative final container product samples.

Review of the response: The sponsor provided the requested information requested. We found the information is adequate and acceptable. No further data is needed related to this IR.

- Please provide data, including your linear regression plots, to demonstrate parallelism between the linear regression fits for the final container product samples and the standard at different concentrations.
- Please provide data on the range of the assay based on your results of linearity, precision and accuracy evaluation using representative final container product samples.

In response to the last two IRs, the sponsor indicated further validation studies would be performed as suggested, and the new data will be completed by Jan 31, 2014.

Conclusion

Based on the limited information provided by the sponsor, the reviewer can not come to the conclusion that the assay has been validated for its intended use at this time.

**11. Determination of Citrate by (b) (4)**

Citrate is an excipient in the Factor X drug product. The specification in the drug product is (b) (4) for lot release.

Method

The citrate concentration in the final container product is determined by (b) (4) following the procedure described in the SOP, QAC/00402/11/SOP. In this method, (b) (4)

[Redacted]

Method Validation

(b) (4)

[Redacted]

(b) (4)

[REDACTED]

Information request

The following IR was submitted on 9 October 2013. The response by Bio products Laboratory of 18 October 2013, follows each request item.

- The accuracy is studied at concentrations much lower than the target concentration of (b) (4). Accuracy should be determined at (b) (4) of the target concentration. Please submit data with accuracy evaluated in this range.

Response: Additional validation as requested using the target concentration range of final product will be carried out.

- Please provide results obtained by multiple analysts on multiple days with representative final container product samples using more than one equipment, if you plan to use more than one equipment, to demonstrate intermediate precision.

Response: The data provided in the BLA to demonstrate intermediate precision was obtained using a number of analysts. Please see Table of Intermediate Precision results for Final Product by (b) (4) Analysts over (b) (4) weeks period on (b) (4) different (b) (4) (Table 3.2.P.5.3-T60)

Review of Response: The sponsor's response addresses our concern.

- The linearity of the method was evaluated in the range (b) (4) however the range of the method was determined to be (b) (4) based on the precision and accuracy results, which is different than the range in which linearity was studied. Please provide additional data for the linearity over the stated range of the assay or redefine your assay range that is supported by linearity, accuracy and precision results.

Response: Additional validation as requested for the linearity over the stated range of the assay will be carried out.

- The reduction of flow rate from (b) (4) did not meet the acceptance criteria. Section 3.2.P.5.3.4.3.7, states that "...as a preventive action from this observation, an assay acceptance limit has been applied to any (b) (4) of retention times within an assay set". Please revise your SOP to include this statement in the method description.

Response: This is stated in section 10.6 of current version of SOP QAC/00402 provided.

Review of Response: The SOP has been revised adequately, which addresses our concern.

After reviewing the response to our 1<sup>st</sup> IR and the method SOP submitted by the sponsor on 18 October 2013, a new IR was submitted.

- Section 4.3.3 Repeatability and Intermediate precision of Validation of procedure for determination of citrate: According to Repeatability and Intermediate precision studies, citrate concentration in the drug product is (b) (4). However, the batch data indicates that the citrate concentration is (b) (4). Please explain this difference. These studies should be performed using the representative final container samples at the target concentration of analysis.

We have not received response to our second information request prior to 31 January 2014.

#### Conclusion

The method is described in sufficient details, and incorporated appropriate assay validity criteria. However, review of validation report could not be completed due to pending information request.

## **12. Determination of Chloride**

This is a quantitative method for the determination of chloride, an excipient of the final container product, is according to the (b) (4) method described in (b) (4)

#### Method

The method involves (b) (4)

#### Method Validation

(b) (4)

Conclusion

The method and validation is satisfactory for the intended use.

**13. Determination of Phosphate**

Method

This quantitative method for the determination of phosphate, an excipient, is according to (b) (4) method described in (b) (4) In this method, (b) (4)

[Redacted]

Method Validation

(b) (4)

[Redacted]

Conclusion

The method and validation is satisfactory for the intended use.

**14. Sodium Content by (b) (4)**

Sodium is present as an excipient in the final container product. The quantity is determined following the method described in (b) (4). The SOP of the test method was not included in the original submission but submitted later in response to our IR (QAC/00319/12/SOP). The proposed specification of sodium (Na) in the final container product is (b) (4).

Method

Sodium in the final container product is quantitated by (b) (4)

Method Validation

(b) (4)

(b) (4)

### Information Request

The following IR was sent to Bio Products Laboratory Ltd. on August 27, 2013.

- Please provide data to show linearity of sodium response in the product matrix and parallelism between the standard and sample regression lines.

Response: Validation was previously carried out in accordance with ICH guidelines. As Control standards were used in the initial validation, additional validation as requested using final product will be carried out. This validation work will be completed by the 31st January 2014.

Review of response: This is acceptable.

- What material/compound was added to Factor X product for the accuracy study?

Response: (b) (4) Sodium standard (Lot (b) (4) from (b) (4) (b) (4)

Review of response: This is acceptable.

- Please provide results obtained by multiple analysts on multiple days using representative final container product samples to demonstrate intermediate precision.

Response: The data provided in the BLA (reference 3.2.P.5.3.4.5.3) to demonstrate intermediate precision was obtained using a number of analysts. Table of Intermediate Precision results for Final Product by (b) (4) Analysts over (b) (4) weeks period. BLA Reference 3.2.P.5.3.4.5.3 (Table 3.2.P.5.3-T77).

Review of response: This is acceptable.

- Please provide data for robustness studies performed with representative final container product samples to demonstrate effect of small variation of critical method parameters.

Response: Sodium method is carried out by (b) (4). There is no critical parameter in the test method that can be altered; hence the effect of small variation of the method parameters cannot be demonstrated. The method will be carried out on (b) (4) and the results reported to demonstrate the effect of the method using (b) (4).

Review of response: This is acceptable.

- Please provide your SOPs or detailed descriptions of the analytical procedures, including system suitability criteria and acceptance criteria for results for the test procedures listed below.

Response: SOP was submitted.

Review of response: This is acceptable.

Conclusion

Review of validation report could not be completed due to deficiencies in the validation report, as discussed above.

15. (b) (4) [Redacted]

[Redacted]

Method

(b) (4) [Redacted]

Method Validation

(b) (4) [Redacted]

[Redacted]

[Redacted]

[Redacted]

[Redacted]

[Redacted]

[Redacted]

[Redacted]



(b) (4)

Conclusion

The (b) (4) test is adequate as a lot release test of the final container drug product.

**17. Determination of Appearance**

The appearance of the freeze-dried material is examined for color and consistency and any abnormalities in vials, freeze-dried plug or vacuum by visual examination. The method was not validated. This is acceptable.

Conclusion

The Appearance test is adequate as a lot release test of the final container drug product.

**18. Determination of Solubility**

The lyophilized product is evaluated for the time necessary to dissolved in Water for Injections at room temperature, according to its reconstitution instructions. The characteristics of the solution is examined by visual examination for color, opalescence, and characteristics of any undissolved material. The proposed specification is colourless, clear or slightly opalescent solution. The method was not validated. Visual inspection is appropriate to verify solubility and the appearance of solution at room temperature and validation of this method is not necessary.

Conclusion

The Solubility test is adequate as a lot release test of the final container drug product.

**19. Determination of (b) (4)**

Method

(b) (4)

Method Validation

(b) (4)

(b) (4)

### Conclusion

The (b) (4) assay is adequate as a lot release test of the final container drug product.

### **CR Comments**

Based on the review of the methods for lot-release testing for the (b) (4) drug product and the validations of the methods for Coagulation Factor X (Human), STN: 125506 from Bio Products Laboratory Limited (BPL), Inc., several deficiencies have been identified. These deficiencies were brought to the attention of the sponsor in a few IRs. Only a few of the questions/comments were addressed so far. The information available at this point does not permit complete review of the application. The sponsor must address all outstanding issues and comments. The outstanding issues and comments are summarized below by test method.

#### **1. Determination of Factor X**

- a. Please revise SOP QCA/00179 to clearly state the assay validity (acceptance) criteria for the standard.
- b. Please describe clearly the details of the testing and calculation of potency in your SOP QCA/00089.
- c. You have not studied specificity of this assay citing that it is a (b) (4) procedure. However, evaluation of specificity is necessary to demonstrate that the method works for your product without interference from the product matrix. Please provide data to demonstrate specificity of this assay based on analysis of representative product samples and matrices.
- d. You have demonstrated accuracy of the method by testing one standard (3rd International Standard) against another standard (b) (4) ( ). Please provide results of accuracy of your method using your process intermediates and the final container product for which this assay is intended. We suggest you evaluate accuracy using spike-recovery method in which unspiked samples at different concentrations and the same samples after spiking with known concentrations (IU/mL) of the standard are analyzed.
- e. You evaluated linearity only using the (b) (4) ( ). please evaluate linearity at different dilution of the product (dilution linearity) and show that the linear regression line of the standard and that of the product are parallel within the proposed assay range to validate that interpolation from the standard regression line is appropriate for the determination of the potency of the product.
- f. Please provide data to establish the range of the assay based on your results of repeatability, accuracy and linearity studies obtained using representative process intermediate and product samples over the intended range of the assay.

- g. Please provide data to demonstrate appropriate robustness of the assay method using representative process intermediate and product samples for which this assay is intended. The data should demonstrate effect of small deliberate changes of critical method parameters, such as reagent concentration, incubation time, etc., as applicable.

## 2. Determination of Total Protein by (b) (4)

- a. Please provide data of the linearity study using representative final container product samples and to demonstrate parallelism between the linear regression fits for the final container product samples and the standard protein used in the linearity study.
- b. Please provide data to establish the range of the assay based on your results of repeatability, accuracy and linearity studies obtained using representative product samples over the intended range of the assay.
- c. We don't agree with that your Internal Quality Control (IQC) sample is representative of the Factor X final container product and that any variation in the method will have similar effect on both final container product and IQC because we found that the composition of IQC is significantly different from that of the product, including the fact that the average protein concentration of IQC is (b) (4) whereas the specification limit for the Factor X product is (b) (4). Thus, the IQC sample (b) (4)

(b) (4)  
Please provide data for robustness studies performed with representative final container product samples.

## 3. Moisture Determination in Freeze-Dried Products by (b) (4) Method

- a. For specificity study, you mentioned, "Specificity was not examined as substances known to interfere in the (b) (4) assay are not present in FACTOR X." Please provide data using representative product samples to substantiate your statement.
- b. You have conducted your method validation using (b) (4) standard but not the final container product for which the assay is intended to be used. Please provide you validation data using representative product samples over the intended range of the assay. The following characteristics should be addressed: specificity, accuracy (spike recovery), repeatability, intermediate precision (multiple analysts, multiple days), linearity, range, limit of quantitation and robustness of the assay. We suggest that you spike your sample with different known amount of (b) (4) and then assay both unspiked and spiked samples to calculate recovery.

## 4. (b) (4) Method for the Determination of Factor II ((b) (4) Assay)

- a. You have not studied specificity of this assay citing that the assay is performed as described in (b) (4). You need to perform specificity study to demonstrate that the method works for your product without interference from the product matrix.

Please provide data to demonstrate specificity of this assay based on analysis of representative product samples.

- b. You have demonstrated accuracy of the method by testing one standard (3rd International Standard) against another standard ((b) (4)). Please provide results of accuracy of your method using your product for which this assay is intended. We suggest you evaluate accuracy using spike-recovery method by analyzing unspiked samples at different concentrations and the same samples after spiking with known concentrations (IU/mL) of the standard in such a way that the total concentrations of factor II in the samples are between the Quantitation Limit of the assay and the proposed specification limit.
- c. Please provide data on the assessment of Quantitation Limit from analysis of representative samples of your product for which the assay is intended.
- d. You evaluated linearity only using the standard. (b) (4) please evaluate linearity at different dilution of the product (dilution linearity) and show that the linear regression line of the standard and that of the factor II content are parallel within the proposed assay range to validate that interpolation from the standard line is appropriate for the determination of factor II content of the product.
- e. Please reevaluate the range of the assay based on your results of repeatability, accuracy and linearity data obtained using representative product samples.
- f. Please provide data to demonstrate appropriate robustness of the assay method using representative product samples for which this assay is intended. The data should demonstrate effect of small deliberate changes of critical method parameters, such as reagent concentration, incubation time, etc.

#### 5. Determination of Factor IX ((b) (4) Assay)

- a. You performed specificity study using a factor IX product that contains (b) (4) IU/mL of FIX concentration. However, your specification limit is NGT 1 IU/mL. Specificity should be evaluated at the expected concentration at which the analyte (factor IX) is present in the product. You assessed specificity at the factor IX concentration of (b) (4), which is significantly higher than the concentration at which it is expected to be present in the product because your proposed specification limit is 1 IU/mL or less. Please submit specificity data by analyzing representative factor X product samples to show that the results are not affected by the matrix at the concentration at which they are expected to be present in the product.
- b. You have demonstrated accuracy of the method by testing one standard (b) (4) against another standard (b) (4). Please provide results of accuracy of your method using your product for which this assay is intended. We suggest you evaluate accuracy using spike-recovery method by analyzing unspiked samples at different concentrations and the same samples after spiking with known concentrations (IU/mL) of the standard in such a way that the total concentrations of factor IX in the samples are between Quantitation Limit and the proposed specification limit for the product.

- c. Please provide data on the assessment of Quantitation Limit from analysis of representative samples of your product for which the assay is intended.
- d. Please provide data, including your linear regression plots, to demonstrate parallelism between the linear regression fits for the final container product samples and the standard at different factor IX concentrations.
- e. Please reassess the range of the assay based on your results of repeatability, accuracy and linearity data obtained using representative product samples.
- f. Please provide data to demonstrate appropriate robustness of the assay method using representative product samples. The data should demonstrate effect of small deliberate changes of critical method parameters, such as reagent concentration, incubation time, etc.
- g. Please provide the SOPs QCA/00042 and QCA/00073. You referenced these two documents in your validation report but have not included them in your submission.

#### **6. Determination of Non-Activated Partial Thromboplastin Time (NAPTT)**

- a. In response to our IR, you responded that this is a qualitative method. Therefore, you will evaluate specificity only to validate these methods. We do not agree. You are assessing an impurity level that is related to safety of the product by measuring the clotting time. Thus, your reportable result is time. Therefore, the method is quantitative. You need to validate the method as a quantitative method with time as the measurand. Thus, in addition to specificity, please provide data on evaluation of other validation characteristics appropriate for a quantitative test for impurity in terms of the reportable result.
- b. Based on our analysis of the calibration (qualification) data for the control you submitted we found that the mean<sup>(b) (4)</sup> SD values are (b) (4)  

Please revise your SOP (QCA/00008) to include (b) (4)

  
as the assay validity criteria.
- c. You responded, “The operator will review the control chart and if the control result is not (b) (4), the assay would be considered invalid, and the results would not be used.” We cannot agree. The assay validity criteria should be mentioned in the SOP and the assay should be considered invalid, if the results do not meet the criteria. Please revise your SOP to include assay validity criteria, as discussed above.
- d. You mentioned that (b) (4) ) are necessary to ensure that there is no masking, due to either over dilution or matrix inhibition. In that case, results from (b) (4) should be your reportable results. Please revise your SOP (QCA/00008) accordingly.
- e. You indicated that (b) (4) step is not necessary for the factor X product. Please revise your SOP (QCA/00008) to include this clarification.

#### **7. Determination of Fibrinogen Clotting Time (FCT)**

- a. In response to our IR, you responded that this is a qualitative method. Therefore, you will evaluate specificity only to validate these methods. We do not agree. You are assessing the impurity level by measuring the clotting time. Thus, your reportable result is time. Therefore, the method is quantitative. You need to validate the method as a quantitative method. Thus, in addition to specificity, please provide data on evaluation of other applicable validation characteristics for a quantitative test for impurity in terms of the reportable result.
- b. Please revise your SOP QCA/00011/15: The Fibrinogen Clotting Time Test to include appropriate and justifiable the assay validity criteria and submit with your justification.

**8. Determination of (b) (4)**

- a. (b) (4)
- [Redacted text block]

**9. Determination of (b) (4)**

- a. (b) (4)
- [Redacted text block]

(b) (4)

[Redacted]

**10. Sucrose determination by (b) (4)**

- a. Please provide data, including your linear regression plots, to demonstrate parallelism between the linear regression fits for the final container product samples and the standard at different concentrations.
- b. Please provide data to establish the range of the assay based on your results of linearity, precision and accuracy evaluation using representative samples of final container product.

**11. Determination of Citrate by (b) (4)**

- a. The accuracy, repeatability and intermediate precision were studied at concentrations much lower than the target concentration of (b) (4). Please evaluate these validation characteristics over the actual assay range.
- b. The linearity of the method was evaluated in the range (b) (4) however the range of the method was determined to be (b) (4) based on the precision and accuracy results, which is different than the range in which linearity was studied. Please provide additional data for the linearity over the stated range of the assay or redefine your assay range that is supported by linearity, accuracy and precision results.

**12. Sodium Content by (b) (4)**

- a. Please provide data to show linearity and accuracy of sodium response using final container product. Also, show parallelism between the standard and sample regression lines to demonstrate assay linearity.

**13. (b) (4) Determination by (b) (4)**

- a. (b) (4)

[Redacted]

[Redacted]

(b) (4)

A large, irregularly shaped grey redaction box covers the majority of the page's content. The text "(b) (4)" is visible at the top left of this redacted area. The redaction obscures all text and graphics that would otherwise be present in the main body of the document.