

**CENTER FOR DRUG EVALUATION AND
RESEARCH**

APPLICATION NUMBER:

22-041

STATISTICAL REVIEW(S)



STATISTICAL REVIEW AND EVALUATION

Stability Studies

| | |
|---------------------------------|----------------------------------------------|
| NDA/SERIAL NO.: | 22-041/N_000 |
| DRUG NAME: | Cyanokit (hydroxocobalamin), 2.5 g Vials |
| INDICATION: | Known or Suspected Cyanide Poisoning |
| SPONSOR: | EMD Pharms |
| DATE RECEIVED BY CENTER: | October 27, 2006, via email |
| REVIEW PRIORITY: | Priority, Orphan Drug, Animal Rule |
| DOCUMENTS REVIEWED: | Stability Data and Report |
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Keywords: Stability, Shelf life estimation.

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2.4 The Stability Study

2.4.1 Sponsor's Analyses, Results, and Conclusions

The sponsor reported that all results of the three primary stability batches were within specifications when stored at 25°C/60%RH or at 30°C/65%RH for 24 months or at 40°C/65%RH for six months with the exception of the sterility test for one batch at 12 months when stored under the intermediate condition. The origin of the contamination could not be identified, but the sponsor stated that the renovation of the production area and the preventive actions implemented in the production and the OC laboratory should ensure that the identified risks have been appropriately addressed, eliminated, or minimized.

In Section 3.2.P.8.3.6 'Statistical Analysis of Stability Studies Results', the sponsor reported the results from the statistical analyses using an FDA SAS program. The shelf life estimate based on assay of hydroxocobalamin data from the three primary batches stored at 25°C/60%RH was 55 months. The sponsor performed the same analyses on the data for total impurities and major impurity when the product was stored at 25°C/60%RH, but apparently did not integrate these findings into their conclusions. They reported that an increase in the major impurity was observed over the first 12 months, followed by a leveling off, but did not state that the major impurity content estimated a shelf life of only 31 months. Instead, the sponsor concluded that the stability data supported a 31-month expiry and that Cyanokit may be stored at temperatures up to 40°C with brief exposures to temperatures up to 40°C. As far as the reviewer could determine, no statistical analyses of the 30°C/65%RH data were submitted.

b(4)

2.4.2 Reviewer's Analyses, Results, and Conclusions

The three primary batches (# 2079, 2080, and 2081) were of commercial size and contained 10 vials each. Stability results under the 25°C/60%RH and 30°C/65%RH conditions had data through 24 months and were evaluated by the reviewer. The reviewing chemist requested shelf life estimates for assay using 88%LC and 90%LC as the lower specification limits. Total impurities were tested against upper specification limits of 0.5% and 1.0%, major impurity against 0.5% and 1.0%, unknown impurity against 0.5% and 1.0%, and unknown impurity against upper limits of 0.5% and 1.0%.

b(4)

The sponsor had performed statistical analyses for the assay, total impurities, and major impurity (specifications of 0.5%LC, 1.0%, and 1.0%, respectively) when the product was stored at 25°C/60%RH. The reviewer confirmed the appropriateness and correctness of these findings. To investigate the sponsor's claim that the product can be stored at the intermediate condition, the reviewer also analyzed the data collected under 30°C/65%RH. In addition, the data from the unknown impurities 0.5% and 1.0% were analyzed when the product was stored at either the room temperature or intermediate condition. In each case, the data from the three batches pooled to a common regression

1.3.4 Statistical Issues

The sponsor's analyses were appropriate and correct. However, the reviewer does not agree with the sponsor's conclusion which is based on results from assay alone. Furthermore, the sponsor's recommended storage condition would allow for storage under the intermediate condition. Hence the data collected under 30°C/65%RH needed to be evaluated as well.

The sponsor mentioned that the data for the major impurity showed an initial steep increase and a subsequent leveling off, but submitted only the results from the linear analysis. They did not incorporate these findings into their conclusions. The reviewer concluded after investigation that the linear model was adequate for these data.

The observed amount of impurity _____ declined over time. As the impurity has only an upper specification limit and the confidence limit was close to the regression line, there was no intersection between the confidence limit and the specification limit.

b(4)

2. STATISTICAL REVIEW AND EVALUATION OF EVIDENCE

2.1 Introduction and Background

Dr. M. Salazar Driver, the reviewing chemist, asked the Division of Biometrics 6 to evaluate the sponsor's stability data. Quick analyses of the 18 month data had confirmed her concerns that the requested shelf life of _____ months was not supported. The sponsor was requested to submit the 24 month stability update. These data and the sponsor's reports arrived via email to the project manager, Mr. M. Sullivan, on October 27, 2006. On October 30, 2006, the SAS Transport data files for assay, major impurity and total impurities were filed in the EDR.

b(4)

2.2 Overview of the Stability Program and Studies Reviewed

The sponsor submitted 24 month stability data for three primary stability batches and 36 month stability data for three supportive stability batches. The three primary batches constitute the main stability program and will be tested again when they reach 30 and 36 months (Sponsor's Table 3.2.P.8.1-15).

2.3 Data Analyzed and Sources

The sponsor submitted the 24 month stability data from the three primary batches as PDF attachments to their email of October 27, 2006. The data for assay, total impurities, and major impurity were also submitted as SAS transport files, which the reviewer used. The data for the unknown impurities _____ and _____ were copied from the sponsor's PDF file 'Cyanokit - Revised 2.3.P.8 Stability 27 Oct 2006'.

b(4)

The reviewer analyzed the stability data for assay, total impurities, major impurity and the unknown impurities _____ and _____ from the three primary batches when stored both at room temperature and the intermediate condition. Varying specification limits were applied to these attributes to explore the product's stability performance.

b(4) Table 1 shows the extrapolated shelf life estimates based on the reviewer's analyses. Shelf life estimates shorter than 36 months are bolded.

Table 1: Shelf Life Estimates by Attribute and Storage Condition

| VARIABLE | SPECIFICATION | CONDITION | EXTRAPOLATED EXPIRY |
|------------|---------------|------------|---------------------|
| Assay | 88%LC | 25°C/60%RH | 55 mos |
| Assay | 90%LC | 25°C/60%RH | 47 mos |
| Total Imp | 1% | 25°C/60%RH | 51 mos |
| Total Imp | 1% | 25°C/60%RH | 34 mos |
| Major Imp | 1/2% | 25°C/60%RH | 31 mos |
| Major Imp | 1/2% | 25°C/60%RH | 25 mos |
| Unk: _____ | 1/2% | 25°C/60%RH | 46 mos |
| Unk: _____ | 1/2% | 25°C/60%RH | 26 mos |
| Unk: _____ | 5% | 25°C/60%RH | 200 mos* |
| Unk: _____ | 10% | 25°C/60%RH | 200 mos* |
| Assay | 88%LC | 30°C/65%RH | 37 mos |
| Assay | 90%LC | 30°C/65%RH | 31 mos |
| Total Imp | 1% | 30°C/65%RH | 36 mos |
| Total Imp | 1% | 30°C/65%RH | 23 mos |
| Major Imp | 1/2% | 30°C/65%RH | 22 mos |
| Major Imp | 1/2% | 30°C/65%RH | 16 mos |
| Unk: _____ | 1/2% | 30°C/65%RH | 36 mos |
| Unk: _____ | 1/2% | 30°C/65%RH | 20 mos |
| Unk: _____ | 5% | 30°C/65%RH | 200 mos* |
| Unk: _____ | 10% | 30°C/65%RH | 200 mos* |

* Confidence limit did not intersect with specification limit.

b(4) **1.3.3 Extent of Evidence in Support of Requested Shelf Life**

The sponsor submitted stability data for 24 months when the product was stored under the 25°C/60%RH and 30°C/65%RH conditions. This amount of data would allow for an extrapolated shelf life of _____ months if supported by the results of the statistical analyses.

1. EXECUTIVE SUMMARY

1.1 Conclusions and Recommendations

The sponsor submitted the 24 month stability update via email attachments to the project manager. The reviewing chemist requested the Division of Biometrics 6 to confirm whether shelf life estimation based on assay, total impurities, major impurity and two unknown impurities of interest supported the requested expiry of —months. b(4)

The sponsor's statistical analyses of the stability data collected at 25°C/60%RH appear appropriate and correct. However, the reviewer does not agree with the sponsor's conclusion that the data support an extrapolated shelf life of — months, as the data from the major impurity estimated an extrapolated expiry of only —months. The sponsor had estimated the same expiry for this attribute but ignored the finding in their conclusion. b(4)

The expiry appropriate for this product will depend on which storage conditions are relevant and which specification limits are chosen for the attributes.

1.2 Overview of the Submission

A 10/27/06 email from the sponsor to the project manager, Mr. Mathew Sullivan, contained attachments of updated stability data and the sponsor's analyses and conclusions. These data were used in the reviewer's analyses.

1.3 Principal Findings

1.3.1 Sponsor's Results and Conclusions

The sponsor proposed a shelf life of — months based on 24 month stability data from three primary batches and — month stability data from three supportive batches. In particular, the sponsor relied on the results of the hydroxocobalamin assay which estimated an expiry of 55 months when the three primary batches were stored at 25°C/60%RH. The recommended storage conditions allowed for temperatures up to 30°C and brief exposures up to 40°C.

1.3.2 Reviewer's Results and Conclusions b(4)

The sponsor's analyses results were correct but were limited to the 25°C/60%RH storage condition and their conclusions appeared to be based only on the assay results. Further, the sponsor recommended storage conditions of up to — C (with excursions up to —C) without submitting any analyses of the data collected under 30°C/65%RH.

b(4) line, which usually gives the longest shelf life estimate (as compared to estimating the shelf life based on individual lines). Under the 25°C/60%RH storage condition, the major impurity and the unknown impurity proved stability limiting depending on which specification limits were applied. The major impurity supported only a 25 month expiry when an upper specification limit of 0.95% was used. The unknown impurity estimated a slightly longer shelf life (26 months) when 0.90% was used as specification limit. When the product was stored 30°C/65%RH, assay, total impurities, major impurity, and unknown impurity could all become stability limiting depending on which specification limits were appropriate. The details of these analyses are given in the Appendix.

b(4) The sponsor noted the steep initial increase in the major impurity data followed by a leveling off, but submitted only the results of the linear regression. They did not, however, incorporate these results into their conclusions. The reviewer considers the results from the linear analyses adequate and hence its shelf life estimate relevant. She based her conclusion on the R^2 statistic which measures the goodness of the linear fit. It gives the proportion of the total variance that is explained by the linear model. For the major impurity data collected under the 25°C/60%RH storage condition, R^2 was slightly greater than 0.59, i.e. almost 60% of the total variation was explained by the linear model. This can be considered as adequate. For comparison, the R^2 for assay reflected a very good fit in that the linear model explained 74% of the total variation. The R^2 for the unknown impurity on the other hand was only 0.51, but visually the linear fit was better than the one for the major impurity. The reviewer also applied transformations to the data (log, square root) and re-fitted the linear model, but the R^2 's were poorer than for the non-transformed data. The shelf life estimation based on a true non-linear model (versus a linear model on transformed data) is hampered by the difficulty of estimating the 95% confidence limits around this model, which are necessary to take the variation due to future batches into account. Again, the reviewer considered the linear fit to the data of the major impurity adequate and hence, considers this attribute as potentially stability limiting.

b(4) The reviewer observed that the values of the unknown impurity were identical across the three primary batches when stored at 25°C/60%RH. Upon inquiry, the sponsor explained that there had been small differences but when the results were rounded to one decimal place, they proved to be identical across batches. This impurity appeared to degrade into other impurities over time and hence showed a negative slope. The confidence limit fit closely around the regression line and did not intersect with the upper specification of 0.95% or 0.90%, which is reflected in the long shelf life estimate. At 30°C/65%RH storage there was somewhat greater variation between the batches, but a similar model led to the same conclusions.

If the product will be labeled for only the 25°C/60%RH storage condition, the appropriate shelf life may be as low as 25 months if 0.95% is the relevant specification limit for the major impurity. All shelf life estimates obtained under the 30°C/65%RH storage condition are shorter than the respective shelf life estimates obtained under the

room temperature condition. The product is clearly sensitive to increased temperatures and humidity.

2.5 Statistical and Technical Issues

The sponsor used a SAS stability program which was given to sponsors years ago. Though the program is no longer made available to sponsor due to potential technical problems (the program is not locked), the sponsor apparently used the program correctly. The reviewer obtained the identical analysis results when using the same data and specification limits as the sponsor had.

The sponsor had performed the same linear analysis on assay, major impurity and total impurities. They implied that the data for the major impurity followed a non-linear pattern, but did not submit or propose any non-linear analyses. The reviewer considered the linear fit to the data of the major impurity adequate based on an $R^2=0.59$.

b(4) The sponsor explained that the data reported for impurity _____ were valid when the product was stored at 25°C/60%RH. The data had shown small variation across batches but became identical when rounded to one decimal place. In addition, this impurity appeared to degrade itself over time, which was reflected in the negative slope of the model. For these data there was no intersection between the confidence limit and the upper specification limit resulting in an unusually long shelf life estimate of 200 months.

b(4) The reviewer considered the sponsor's conclusion of a ← month shelf life for Cyanokit, as inappropriate, because they had obtained a shorter shelf life based on one of the attributes (major impurity with ←% as specification). It is illogical to propose an expiry longer than was estimated by any of the attributes. This is the only inconsistency observed by the reviewer. The sponsor had not applied the more stringent specification limits.

It seemed that the sponsor considered the product equally stable at 30°C/65%RH, but the reviewer could not locate any analyses of these data. As can be seen from Table 1, all shelf life estimates are shorter when based on data from the 30°C/65%RH condition as compared to the 25°C/60%RH condition.

2.6 Statistical Evaluation of Collective Evidence

b(4) The reviewer evaluated the data from the batches of the primary stability study. These were production size lots and appropriate to estimate the shelf life. In addition, 24 month data would allow for a potential extrapolation to the requested ← months. The reviewer included the findings of all important attributes, as identified by the reviewing chemist, into her conclusion. The sponsor had based their conclusions only on assay results.

The reviewer's evaluation of the data collected under the 30°C/65%RH condition provides relevant information when deciding whether the sponsor's proposal of storing the product up to —C can be permitted. In the files submitted, the sponsor had not addressed this issue.

2.7 Conclusions and Recommendations

b(4)

The shelf life estimate will depend on which storage conditions are appropriate for the product and which is the appropriate specification limit for each attribute. With the 24 month stability data submitted, the expiry cannot be more than 31 months. If the product will be labeled for the 25°C/60%RH storage condition and if —% is the relevant upper limit for major impurity, an expiry of only 25 months is supported. If 30°C/65%RH will be an acceptable storage condition, a shelf life of 22 months is possible if the most liberal specification limits apply to all attributes. Otherwise a shelf life as short as 16 months may be applicable.

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3. APPENDIX: ANALYSIS RESULTS

3.1 Shelf Life Estimation under 25°C/60%RH.

b(4)

The analyses results showed that each shelf life estimate is based on the pooled data from the three production batches 2079, 2080, and 2081. The expiry was estimated by the intersection of the one-sided 95% confidence limit for the mean around the regression line with the specification limit. For assay the lower 90%LC and 88%LC was used as specification, which translated into --- g and --- g respectively. Total impurities were tested against an upper limit of ---\% and ---\% . The major impurity had an upper specification of ---\% or ---\% , and the unknown impurity --- was tested against both --- and --- . Finally the unknown impurity --- had upper limits of ---\% and ---\% .

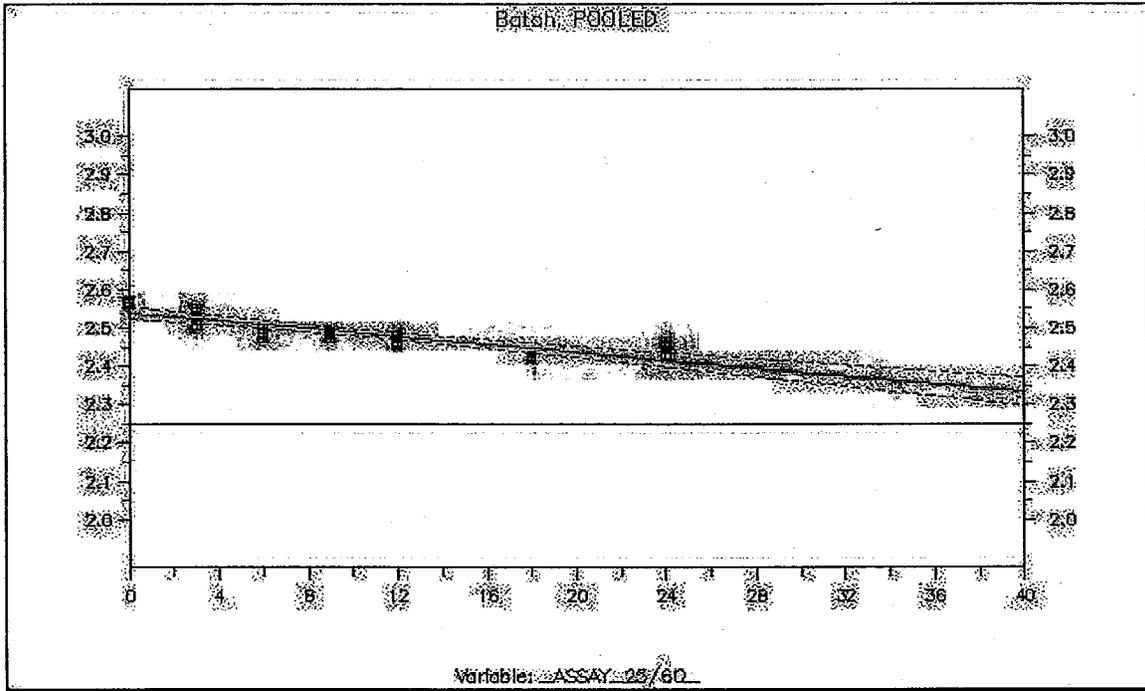
Table 2: Shelf Life Estimate for Assay under 25°C/60%RH

| Batch | SS | df | MS | F-Statistic | F _{0.95,df} |
|----------|--------|----|--------|-------------|----------------------|
| A | 0.0014 | 4 | 0.0004 | 0.5148 | 0.7261 |
| B | 0.0014 | 2 | 0.0007 | 0.9853 | 0.3962 |
| C | 0.0001 | 2 | 0.0000 | 0.0442 | 0.9569 |
| RESIDUAL | 0.0103 | 15 | 0.0007 | | |

| Regression | R-Square | F-Stat | Estimator Expiry Point Spec = 92%LC = 2.76 g |
|----------------------------|----------|--------|-------------------------------------------------|
| Y = 2.5403 - 0.0051 x Time | 0.7434 | POOLED | 55 |
| | | -MIN- | 55 |

| Regression | R-Square | F-Stat | Estimator Expiry Point Spec = 88%LC = 2.67 g |
|----------------------------|----------|--------|-------------------------------------------------|
| Y = 2.5403 - 0.0051 x Time | 0.7434 | POOLED | 47 |
| | | -MIN- | 47 |

Figure 1: Shelf Life Estimate for Assay under 25°C/60%RH



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Table 3: Shelf Life Estimate for Major Impurity under 25°C/60%RH

| Source | SS | DF | MS | F-Statistic | P-Value |
|----------|--------|----|--------|-------------|---------|
| A | 0.0088 | 2 | 0.0022 | 0.0310 | 0.9979 |
| B | 0.0088 | 2 | 0.0019 | 0.0269 | 0.9735 |
| C | 0.0050 | 2 | 0.0025 | 0.0352 | 0.9655 |
| RESIDUAL | 0.668 | 15 | 0.0707 | | |

| Model | R-Square | Batch | Distribution | Number of Data Points |
|------------------------------------------|----------|--------|--------------|-----------------------|
| $Y = 2.1373 + 0.0348 \times \text{time}$ | 0.5932 | POOLED | | 31 |
| | | MIN | | 31 |
| | | | | |
| $Y = 2.1373 + 0.0348 \times \text{time}$ | 0.5932 | POOLED | | 25 |
| | | MIN | | 25 |

b(4)

Figure 2: Shelf Life Estimate for Major Impurity under 25°C/60%RH

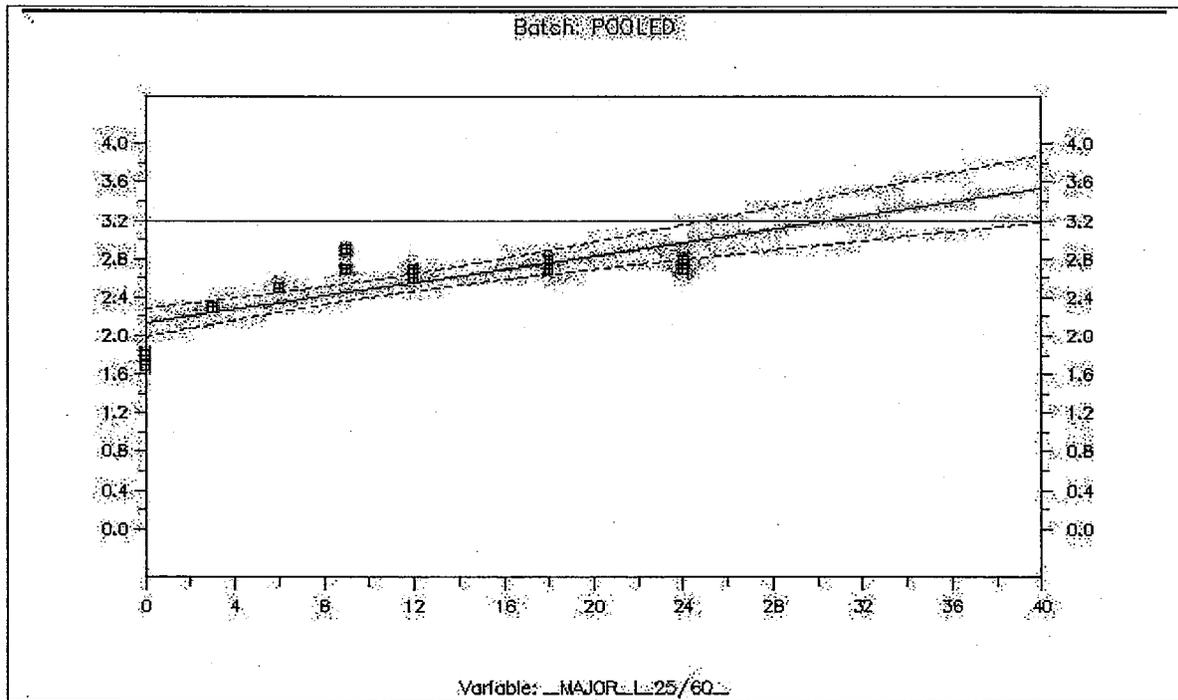


Table 4: Shelf Life Estimate for Total Impurities under 25°C/60%RH

| Source | SS | DF | MS | F-Statistic | P-Value |
|----------|--------|----|--------|-------------|---------|
| A | 0.9363 | 4 | 0.0841 | 0.2956 | 0.8763 |
| B | 0.2552 | 2 | 0.1276 | 0.4487 | 0.6468 |
| C | 0.0810 | 2 | 0.0405 | 0.1424 | 0.8684 |
| RESIDUAL | 1.2665 | 15 | 0.2841 | | |

| Model | R-Square | Stat | Estimated Value (Time) |
|------------------------------------------|----------|--------|------------------------|
| $Y = 6.8529 + 0.0805 \times \text{time}$ | 0.6446 | POOLED | 51 |
| | | MIN | 51 |
| | | | |
| $Y = 6.8529 + 0.0805 \times \text{time}$ | 0.6446 | POOLED | 34 |
| | | MIN | 34 |

b(4)

Figure 3: Shelf Life Estimate for Total Impurities under 25°C/60%RH

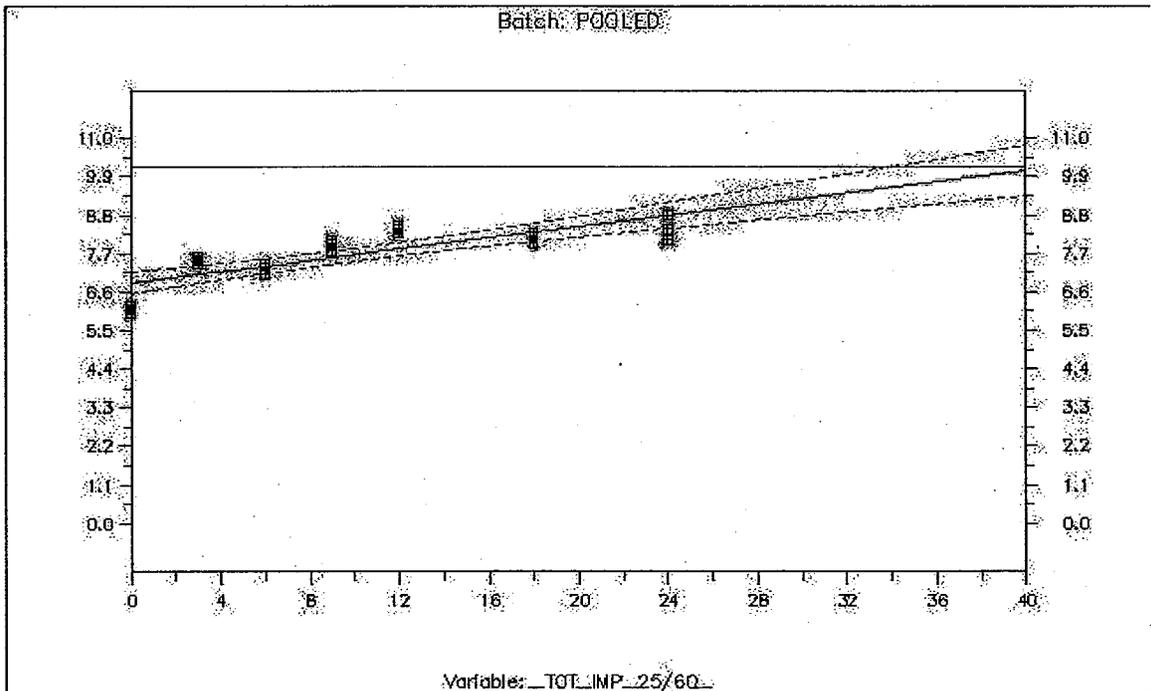


Table 5: Shelf Life Estimate for Unknown Impurity under 25°C/60%RH

| Source | SS | DF | MS | F-Statistic | p-value |
|----------|--------|----|--------|-------------|---------|
| A | 0.0121 | 2 | 0.0030 | 0.9479 | 0.8414 |
| B | 0.0067 | 2 | 0.0033 | 0.9829 | 0.6884 |
| C | 0.0054 | 2 | 0.0027 | 0.9129 | 0.7360 |
| RESIDUAL | 0.1306 | 15 | 0.0087 | | |

| Model | R-Square | Batch | Estimated Total Sample Size |
|------------------------------------------|----------|--------|-----------------------------|
| $Y = 0.0415 + 0.0108 \times \text{Time}$ | 0.5119 | POOLED | 46 |
| | | MIN | 46 |
| Model | R-Square | Batch | Estimated Total Sample Size |
| $Y = 0.0415 + 0.0108 \times \text{Time}$ | 0.5119 | POOLED | 26 |
| | | MIN | 26 |

b(4)

Figure 4: Shelf Life Estimate for Unknown Impurity under 25°C/60%RH

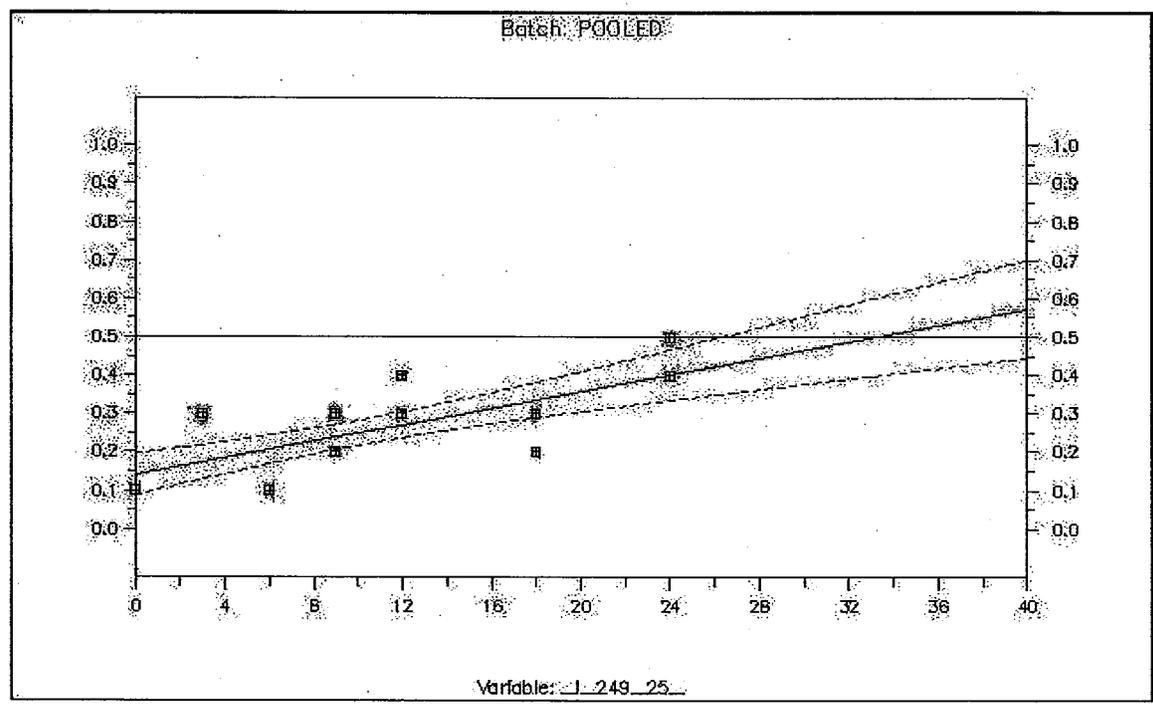


Table 6: Shelf Life Estimate for Unknown Impurity under 25°C/60%RH

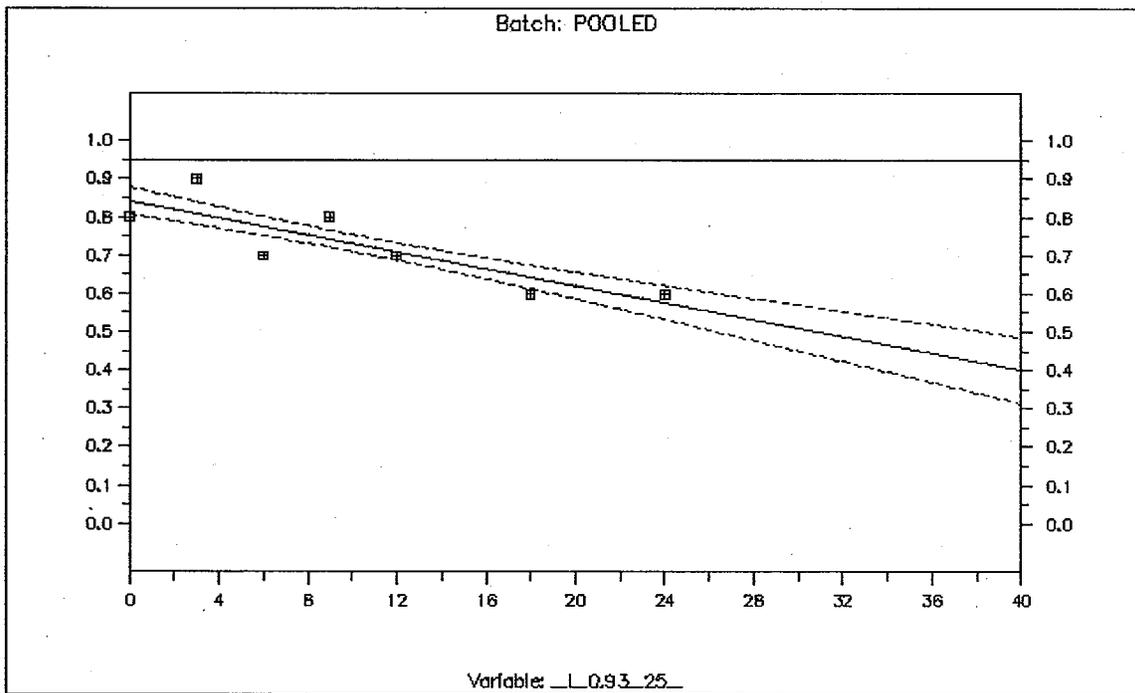
| Source | SS | DF | MS | F-Statistic | P-Value |
|----------|--------|----|--------|-------------|---------|
| A | 0.0000 | 4 | 0.0000 | 0.0000 | 1.0000 |
| B | 0.0000 | 2 | 0.0000 | 0.0000 | 1.0000 |
| C | 0.0000 | 2 | 0.0000 | 0.0000 | 1.0000 |
| RESIDUAL | 0.0648 | 15 | 0.0043 | | |

| Model | R-Square | Factor | Estimated Total Sample Size |
|------------------------------------------|----------|--------|-----------------------------|
| $Y = 0.8425 - 0.0111 \times \text{time}$ | 0.7094 | POOLED | 200 |
| | | MIN | 200 |

* Data were identical across batches.

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Figure 5: Shelf Life Estimate for Unknown Impurity under 25°C/60%RH



3.2 Shelf Life Estimation under 30°C/65%RH.

The analyses results showed that each shelf life estimate is based on the pooled data from the three production batches 2079, 2080, and 2081. As before, the expiry was estimated by the intersection of the one-sided 95% confidence limit for the mean around the regression line with the specification limit. For assay the lower 88%LC and 90%LC was used as specification, which translated into --- g and --- g respectively. Total impurities were tested against an upper limit of ---\% or of ---\% . The major impurity had an upper specification of ---\% or of ---\% , and the unknown impurity --- was tested against both ---\% and --- . Finally the unknown impurity --- had upper limits of --- and ---\% .

b(4)

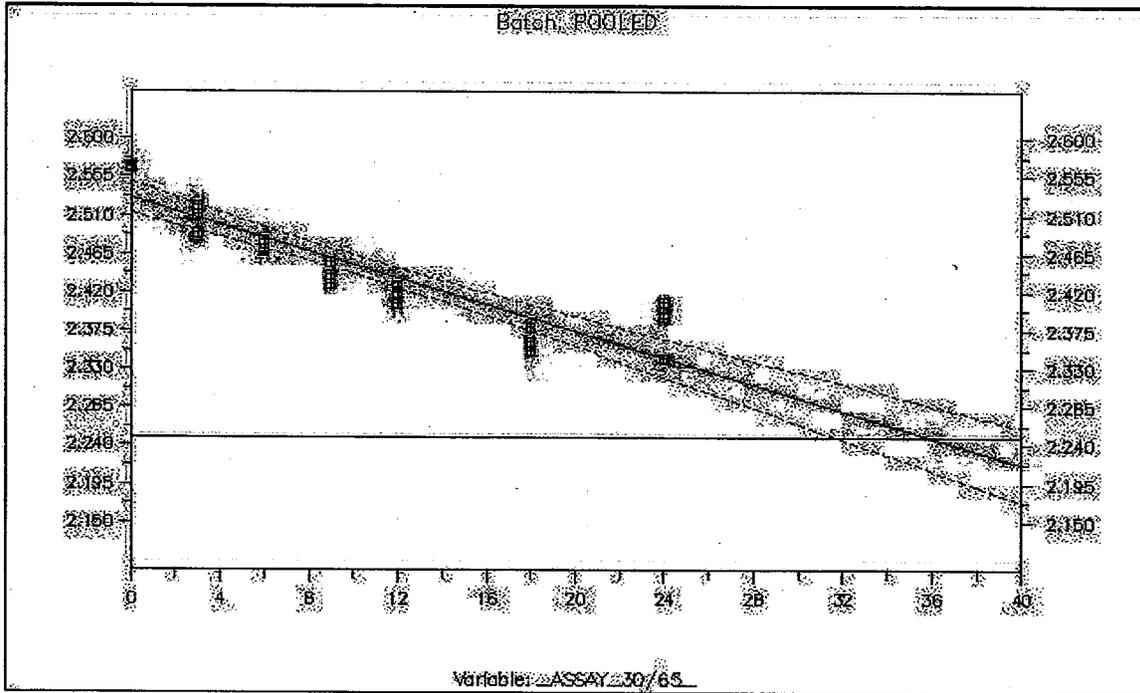
Table 7: Shelf Life Estimate for Assay under 30°C/65%RH

| Sample | S _e | DF | t _{0.95} | Residual | Expiry |
|----------|----------------|----|-------------------|----------|--------|
| A | 0.0028 | 4 | 0.0007 | 0.7640 | 0.5649 |
| B | 0.0022 | 2 | 0.0011 | 1.1866 | 0.3323 |
| C | 0.0006 | 2 | 0.0003 | 0.3414 | 0.7162 |
| RESIDUAL | 0.0138 | 15 | 0.0009 | | |

| Fitted Line | R-Square | Batch | Estimated Expiry Period Spec = 88%LC |
|----------------------------|----------|--------|-----------------------------------------|
| Y = 2.5310 - 0.0079 x Time | 0.8268 | POOLED | 37 |
| | | -MIN- | 37 |

| Fitted Line | R-Square | Batch | Estimated Expiry Period Spec = 90%LC |
|----------------------------|----------|--------|-----------------------------------------|
| Y = 2.5310 - 0.0079 x Time | 0.8268 | POOLED | 31 |
| | | -MIN- | 31 |

Figure 6: Shelf Life Estimate for Assay under 30°C/65%RH



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Table 8: Shelf Life Estimate for Major Impurity under 30°C/65%RH

| Source | SS | DF | MS | F-Statistic | P-Value |
|----------|--------|----|--------|-------------|---------|
| A | 0.0379 | 4 | 0.0095 | 0.0598 | 0.9927 |
| B | 0.0352 | 2 | 0.0176 | 0.1113 | 0.8954 |
| C | 0.0026 | 2 | 0.0013 | 0.0089 | 0.9917 |
| RESIDUAL | 2.3737 | 15 | 0.1582 | | |

| Model | R-Square | Batch | Number of Observations |
|------------------------------------------|----------|--------|------------------------|
| $Y = 2.3499 + 0.0405 \times \text{time}$ | 0.4673 | POOLED | 22 |
| | | MIN | 22 |
| Model | R-Square | Batch | Number of Observations |
| $Y = 2.3499 + 0.0405 \times \text{time}$ | 0.4673 | POOLED | 16 |
| | | MIN | 16 |

b(4)

Figure 7: Shelf Life Estimate for Major Impurity under 30°C/65%RH

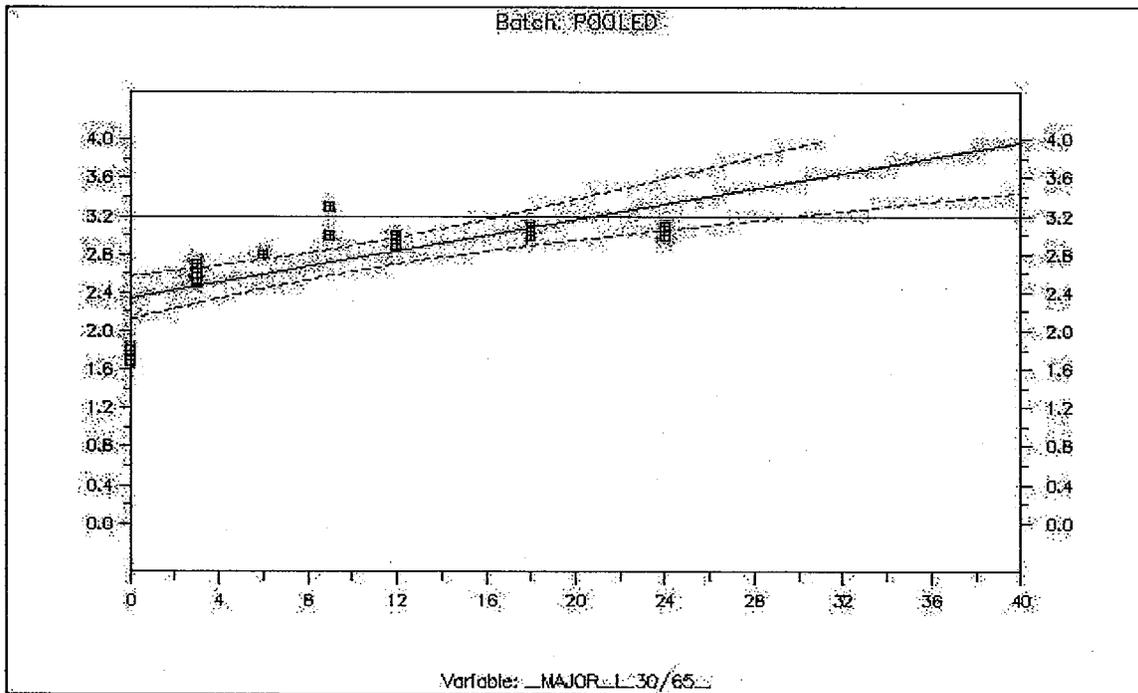


Table 9: Shelf Life Estimate for Total Impurities under 30°C/65%RH

| Source | SS | DF | MS | Statistic | P-value |
|----------|--------|----|--------|-----------|---------|
| A | 0.7708 | 4 | 0.1927 | 0.3171 | 0.8621 |
| B | 0.7400 | 2 | 0.3700 | 0.6089 | 0.5569 |
| C | 0.0308 | 2 | 0.0154 | 0.0253 | 0.9750 |
| RESIDUAL | 9.1450 | 15 | 0.6077 | | |

| Model | R-Square | F-Value | Probability > F |
|----------------------------|----------|---------|-----------------|
| Y = 7.2156 + 0.1068 x Time | 0.5979 | POOLED | 36 |
| | | MIN | 36 |

| Model | R-Square | F-Value | Probability > F |
|----------------------------|----------|---------|-----------------|
| Y = 7.2156 + 0.1068 x Time | 0.5979 | POOLED | 28 |
| | | MIN | 23 |

b(4)

Figure 8: Shelf Life Estimate for Total Impurities under 30°C/65%RH

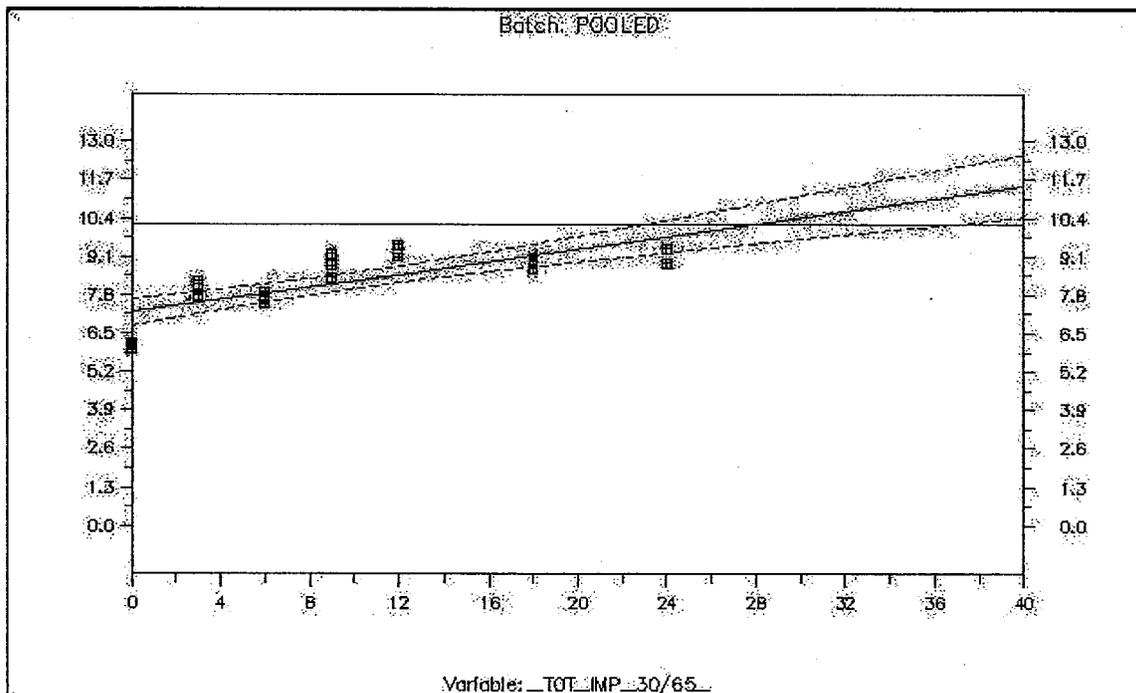


Table 10: Shelf Life Estimate for Unknown Impurity under 30°C/65%RH

| Source | SS | df | MS | F-Statistic | P-Value |
|----------|--------|----|--------|-------------|---------|
| A | 0.0044 | 4 | 0.0011 | 0.0994 | 0.9810 |
| B | 0.0038 | 2 | 0.0019 | 0.1717 | 0.8438 |
| C | 0.0006 | 2 | 0.0003 | 0.0271 | 0.9743 |
| RESIDUAL | 0.1664 | 15 | 0.0111 | | |

| Initial Conc | F-Statistic | Batch | Batched Error Variance |
|------------------------------------------|-------------|--------|------------------------|
| $Y = 0.1423 + 0.0144 \times \text{Time}$ | 0.6102 | POOLED | 36 |
| | | MIN | 36 |
| | | | |
| $Y = 0.1423 + 0.0144 \times \text{Time}$ | 0.6102 | POOLED | 20 |
| | | MIN | 20 |

b(4)

Figure 9: Shelf Life Estimate for Unknown Impurity under 30°C/65%RH

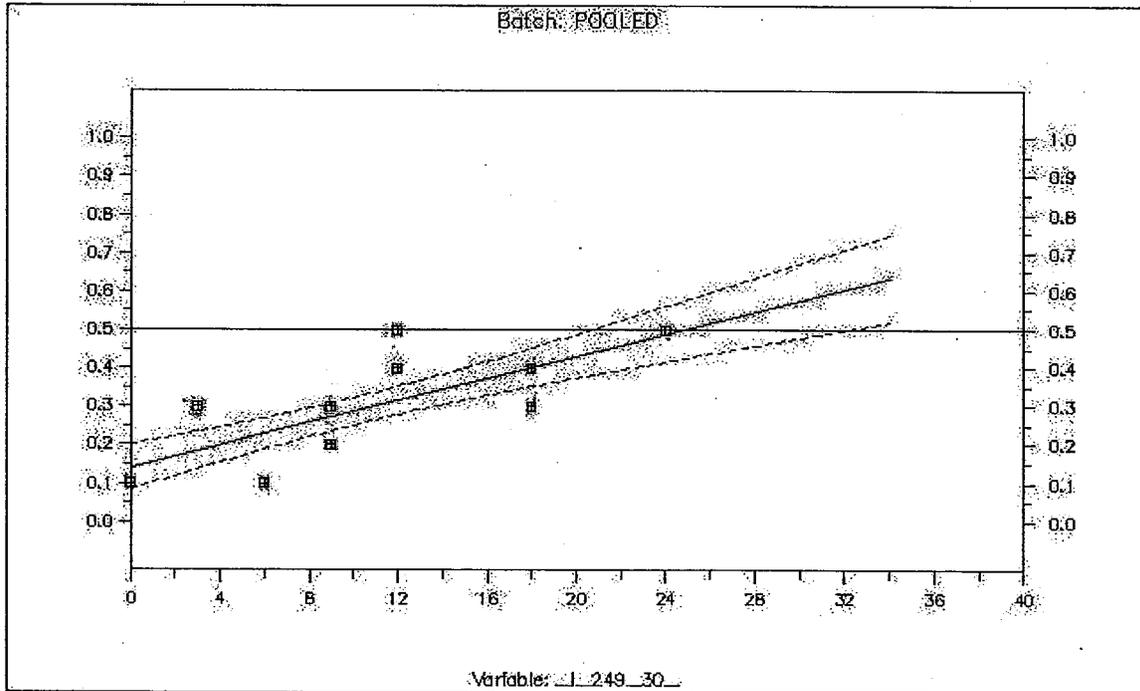


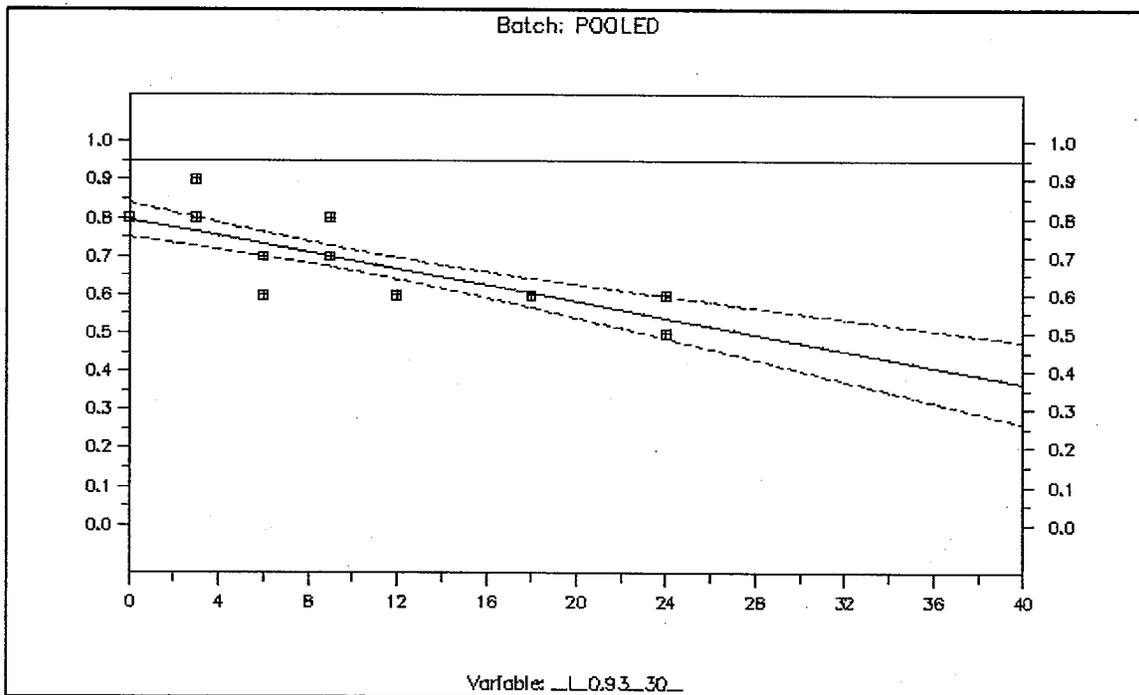
Table 11: Shelf Life Estimate for Unknown Impurity under 30°C/65%RH

| Source | SS | df | MS | F-Statistic | P-Value |
|----------|--------|----|--------|-------------|---------|
| A | 0.0094 | 4 | 0.0023 | 0.9885 | 0.8136 |
| B | 0.0086 | 2 | 0.0043 | 0.7114 | 0.5068 |
| C | 0.0008 | 2 | 0.0004 | 0.0656 | 0.9368 |
| RESIDUAL | 0.0004 | 15 | 0.0000 | | |

| Model | R-Statistic | Batch | Distribution |
|------------------------------------------|-------------|--------|--------------|
| $Y = 0.7952 - 0.0106 \times \text{Time}$ | 0.5942 | POOLED | 200 |
| | | MIN | 200 |

b(4)

Figure 10: Shelf Life Estimate for Unknown Impurity under 30°C/65%RH



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U.S. Department of Health and Human Services
Food and Drug Administration
Center for Drug Evaluation and Research
Office of Translational Sciences
Office of Biostatistics

STATISTICAL REVIEW AND EVALUATION

CLINICAL STUDIES

NDA/Serial Number: 22,041/N000

Drug Name: Cyanokit (hydroxocobalamin)

Indication(s): Treatment of known or suspected cyanide poisoning

Applicant: EMD Pharmaceuticals

Date(s): Paper submission dated 06/16/2006

Review Priority: Priority

Biometrics Division: Division of Biometrics II

Statistical Reviewer: James Gebert, Ph.D.

Concurring Reviewers: Dionne Price, Ph.D., Acting Statistics Team Leader
Thomas Permutt, Ph.D., Acting Division Director

Medical Division: Division of Anesthesia, Analgesia and Rheumatology Products

Clinical Team: Arthur Simone, M.D.
Sharon Hertz, M.D.

Project Manager: Matthew Sullivan

Keywords: Clinical studies, NDA review

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1. EXECUTIVE SUMMARY

1.1 Conclusions and Recommendations

EMD Pharmaceuticals has conducted an animal efficacy study in dogs to evaluate Cyanokit (hydroxocobalamin) for the treatment of known or suspected cyanide poisoning. Efficacy of Cyanokit compared to vehicle (0.9% saline) is seen in the survival rates of dogs treated with both the 75 mg/kg and 150 mg/kg doses of Cyanokit. These doses correspond approximately to 5 g and 10 g in humans, respectively.

1.2 Brief Overview of Clinical Studies

My review will focus on the animal efficacy study (N106342) since it was the only controlled study and the only study that a statistical review would be helpful. It would be very difficult to estimate what survival rates might be expected in the uncontrolled studies if cyanokit was not used. [This is the reason that an animal efficacy study was needed.] The animal efficacy study was a randomized, parallel group, vehicle-controlled, single center study of 14 days duration conducted in beagle dogs.

My review will not discuss the uncontrolled human studies from the literature submitted by the sponsor nor will it discuss the safety study in humans.

1.3 Statistical Issues and Findings

The animal efficacy rule states that a single animal species is adequate if it represents a sufficiently well-characterized animal model (meaning the model has been adequately evaluated for its responsiveness) for predicting the response in humans. The medical division believes that this is true for the study in beagles. According to the sponsor, the mechanism of action is identical in beagles and humans. Namely, hydroxocobalamin binds with cyanide to form cyanocobalamin. This action was demonstrated by the reduction of cyanide and the increase in cyanocobalamin in the cyanokit treated beagles. The effect was additionally seen in the highly significant differences in survival between the cyanokit treated beagles and the vehicle treated beagles.

2. INTRODUCTION

2.1 Overview

Cyanokit is used to treat known or suspected cyanide poisoning. Cyanokit was approved in France in May 1996. Cyanokit received Orphan Drug Designation status for this indication on November 25, 2003. Cyanokit contains two vials of lyophilized powder for i.v. infusion containing 2.5 g lyophilized hydroxocobalamin. Hydroxocobalamin binds with cyanide to form cyanocobalamin, a stable, nontoxic compound that is excreted in the urine. The sponsor states, "Hydroxocobalamin is prepared by r _____"

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Each vial of
Cyanokit is recommended to be reconstituted with 100 ml of sterile saline (0.9% NaCl).

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One of the main causes of cyanide poisoning is smoke inhalation. Hydrogen cyanide is produced by the incomplete combustion of natural fibers (such as wool and silk) and synthetic polymers (such as polyurethane and nylon) widely used in insulation, cushioning, carpets and other building materials and home furnishings. Because of the nature of exposure and the life-threatening toxicity of cyanide, it is unethical to do controlled human clinical trials of Cyanokit. Some uncontrolled studies are reported in the literature and discussed in the submission. However, these studies will not be discussed in my review. The sponsor was told at an industry meeting on April 29, 2003 that it was likely that at least one efficacy study in an appropriate animal model and performed under GLP conditions would be needed to support approval.

Under 21 CFR part 314, subpart I (the animal efficacy rule), the FDA can rely on the evidence from animal studies to provide substantial evidence of the effectiveness when:

- 1. There is a reasonably well-understood pathophysiological mechanism for the toxicity of the chemical, biological, radiological, or nuclear substance and its amelioration or prevention by the product;*
- 2. The effect is demonstrated in more than one animal species expected to react with a response predictive for humans, unless the effect is demonstrated in a single animal species that represents a sufficiently well-characterized animal model (meaning the model has been adequately evaluated for its responsiveness) for predicting the response in humans;*
- 3. The animal study endpoint is clearly related to the desired benefit in humans, which is generally the enhancement of survival or prevention of major morbidity; and*
- 4. The data or information on the pharmacokinetics and pharmacodynamics of the product or other relevant data or information in animals and humans is sufficiently well understood to allow selection of an effective dose in humans, and it is therefore reasonable to expect the effectiveness of the product in animals to be a reliable indicator of its effect in humans.*

The sponsor submitted a special protocol for an animal efficacy study in dogs on October 30, 2003. A review of the protocol led to a change of the primary efficacy variable to survival at Day 14.

2.1.1 Study N106342

Study N106342 was a randomized, parallel group, single center study of 14 days' duration in beagle dogs. The dogs were anesthetized and an indwelling catheter was placed in the femoral vein for blood cyanide, cyanocobalamin, and total cobalamins III sample collections. Each dog was administered an intravenous dose of potassium cyanide (KCN) at 0.4 mg/kg/min until the first apnea was observed. Apnea was defined as complete cessation of breathing or a reduction in tidal volume to less than 4 mL/kg body weight. After the first apnea, the KCN infusion continued for an additional three minutes. Immediately after the conclusion of the KCN, either the test treatments or vehicle was infused over 7.5 minutes. The treatments were 75 mg/kg cyanokit i.v., 150 mg/kg cyanokit i.v. or vehicle. Concurrently at the end of KCN infusion, the dogs were mechanically ventilated. The dogs that survived were later weaned from ventilation. Observations for morbidity and mortality were made twice daily. Information was recorded whether mortality occurred by 1 hour, 4 hours, 7 days, or 14 days following the start of dosing. The primary endpoint of the study was 14 day survival.

2.2 Data Sources

The sponsor provided only stability and adverse event data files for this submission. Data derived from the animal efficacy study was contained in data listings in the clinical reports.

3. STATISTICAL EVALUATION

3.1 Evaluation of Efficacy

A total of 56 (27 male and 29 female) dogs were used in study N106342. Two female dogs did not receive the proper doses and were removed from the study and replaced by additional dogs. The study report did not specify what was not dosed properly, KCN or cyanokit. There was no data for these dogs in the study report. The dogs were 7 to 8 months of age at the time of dosing. There were 17 dogs in the vehicle group, 19 dogs in the low-dose Cyanokit group and 18 dogs in the high dose Cyanokit group. The treatment groups were comparable in mean body weight and mean total KCN received.

At the end of the Cyanokit infusion, cyanide concentration had been decreased to 30-40 nmol/mL, while the vehicle group cyanide concentrations remained at levels of about 70 nmol/mL. Formation of considerable amounts of cyanocobalamin was observed in the Cyanokit groups. No cyanocobalamin was observed in the vehicle group. The mean Cmax of cyanocobalamin was 27% higher in dogs receiving the high-dose Cyanokit compared to the dogs receiving the low-dose.

The sponsor's table 2 (located in section 4.2.1.1/ page 45 of — Study Number N106342 of the study report), below provides the number and percentage of dead dogs in the treatment groups at the four protocol specified evaluation times. I do not know why the sponsor's table uses Day 8 and 15 rather than Days 7 and 14 discussed earlier. The sponsor states that Fisher's exact test was used both for the overall significance among the three groups and the comparisons with the vehicle group. Since Fisher's exact test is used to compare differences between two treatment groups, I do not know how the sponsor used the test to compare the three groups, overall. The sponsor may have possibly used an exact test extended to account for several groups. The same conclusions can be drawn if a Chi-square test is used to compare the three groups. The difference between the low-dose and high-dose Cyanokit groups at days 8 and 15 is not significant ($p=0.105$) using Fisher's Exact test.

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Table 2 Mortality Rate Analysis in Beagle Dogs Receiving Vehicle or Cyanokit (75 or 150 mg/kg i.v.) as a Treatment after KCN Poisoning.

| Timepoint | Vehicle (n=8 male, 9 female) | Low dose (75mg/kg) (n=10 male, 9 female) | High Dose (150 mg/kg) (n=9 male, 9 female) | Overall Significance among Dose Groups |
|-----------|------------------------------------|---------------------------------------------------|-----------------------------------------------------|-------------------------------------------------|
| Hour 1 | No Dead (%) 3 (17.6 %) | No. Dead (%) 1 (5.3 %) | No. Dead (%) 0 (0 %) | No |
| Hour 4 | 10 (58.8 %) | 1 (5.3 %)* | 0 (0 %)* | Yes |
| Day 8 | 14 (82.4 %) | 4 (21.1%)* | 0 (0 %)* | Yes |
| Day 15 | 14 (82.4 %) | 4 (21.1%)* | 0 (0 %)* | Yes |

* Significant at the 0.025 level compared to vehicle using Fisher's exact test.

The sponsor has corrected for multiplicity within each timepoint by using the 0.025 level for the Fisher's exact test. No multiplicity adjustment is needed for different timepoints

3.2 Evaluation of Safety

Safety is not the focus of my review. See the review of the medical officer, Dr. Arthur Simone, for the safety evaluation.

4. FINDINGS IN SPECIAL/SUBGROUP POPULATIONS

4.1 Gender, Race and Age

Due to the small sample size, formal subgroup analyses were not conducted. The three vehicle dogs that survived were females. One male and three females did not survive out to day 15 in the low-dose cyanokit group. The cyanokit doses appeared to be effective in male and female dogs.

The efficacy in human subgroups will be discussed in the medical officer's review.

4.2 Other Special/Subgroup Populations

The efficacy and safety in subjects in uncontrolled smoke-inhalation studies will be discussed in Dr. Simone's review.

5. SUMMARY AND CONCLUSIONS

5.1 Statistical Issues and Collective Evidence

Although the sponsor conducted only one animal efficacy study, I believe that it alone is adequate for demonstrating efficacy for the following reasons: both doses of Cyanokit were highly significantly different from vehicle in survival at day 15, the mechanism of action was identical in dogs and humans, and the study demonstrated the reduction in cyanide and the increase in cyanocobalamin in the Cyanokit groups.

The sponsor has also submitted uncontrolled supportive studies from the literature.

5.2 Conclusions and Recommendations

The efficacy of Cyanokit compared to vehicle was evident in the survival rates of dogs for both the 75 mg/kg and 150 mg/kg doses of Cyanokit. The doses correspond approximately to 5 g and 10 g in humans, respectively.

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