

TECHNOLOGY PREVIEW 03/2020 – This report has been generated with the upcoming Dose-Response-Analysis Package for PLA 3.0.5 (or higher). The package has not been released and has not been finally verified for production use. Contact Stegmann Systems (<http://support.bioassay.de>) to report issues or suggest additional features.

4 PLF - Interpolation

Document-Id: Document-27 (Revision: 1)
Document type: Dose-response analysis/1
Last modified: 25.03.2020 07:55:41
Database: DRA build 65
Database GUID: 741ac16e-3f79-46e6-a8a7-5cb4ad388293

Calculation information

Calculation performed: 24.03.2020 14:15:39, odin.kleuser@stegmannsystems.com (PLA 3.0.4 Build 762, nbersatz02, 10014)
Messages (warning): [W5228] The fit of sample STD has a reduced accuracy.
Report generated: 25.03.2020 08:09:42, Administrator (PLA 3.0.4 Build 762, NBAFR01, 10014)

Signatures

.....
Responsibility

.....
Review

.....
Approval

Table of contents

STD.....	3
TST1.....	6
TST2 - Source Material with Predilution.....	8
TST3 - Transformaiton with Predilution.....	10
TST4 - Mixed Response Data Handling with Stock Solution.....	12

Documentation

Date 31.07.2019 13:22:55



DOCUMENT-27



741AC16E-3F79-46E6-A8A7-5CB4AD388293

TECHNOLOGY PREVIEW - NOT FOR PRODUCTION USE

Assay overview

Assay elements	Summary
<ul style="list-style-type: none"> ■ STD ■ TST1 ■ TST2 - Source Material with Pr... ■ TST3 - Transformaiton with Pre... ■ TST4 - Mixed Response Data Han... 	<p>Reference standard, multi-dose (10 dilution steps, 3 replicates)</p> <p>Test sample, single-dose (3 replicates)</p> <p>Test sample, single-dose (3 replicates), defined concentration: 80.0 IU/L</p> <p>Test sample, single-dose (3 replicates)</p> <p>Test sample, single-dose (3 replicates), defined concentration: 3.0 IU/L</p>

Property	Value
<i>Response data processing*</i> Response normalization Response adjustment Response transformation <i>Analytical model</i> Model Dose transformation	<p>Normalization by fixed value: 2 (TST4 - Mixed Re...)</p> <p>Adjustment by fixed value: 100 (TST4 - Mixed Re...)</p> <p>Square root response transformation (TST3 - Transfor...)</p> <p>4 parameter logistic fit</p> <p>Logarithmic: binary logarithm (base 2)</p>

* The response data processing steps are executed in the reported order.

Assay suitability tests

	Passed	Failed (rejected)	Failed (warning)	Info
Assay suitability tests	3	0	0	0
Overall test result	Passed			

■ STD F-Test (hypothesis test): Model Scope: Standard only; Severity level: rejection		Passed
Calculated F-value	1052.3393	
F _{critical} (95%)	0.11587	
■ STD Equivalence test (parameter estimate), Parameter: B parameter Scope: Standard only; Severity level: warning		Passed
Calculated value	1.38702	
Limits	1.15491 to 1.61913	
Margins	1.00000 to 2.00000	
■ STD Test: Sum of squares (ANOVA): Lack of fit Scope: Standard only; Severity level: warning		Passed
Calculated value	14.04239	
Upper margin	20.00000	

Interpolation results overview

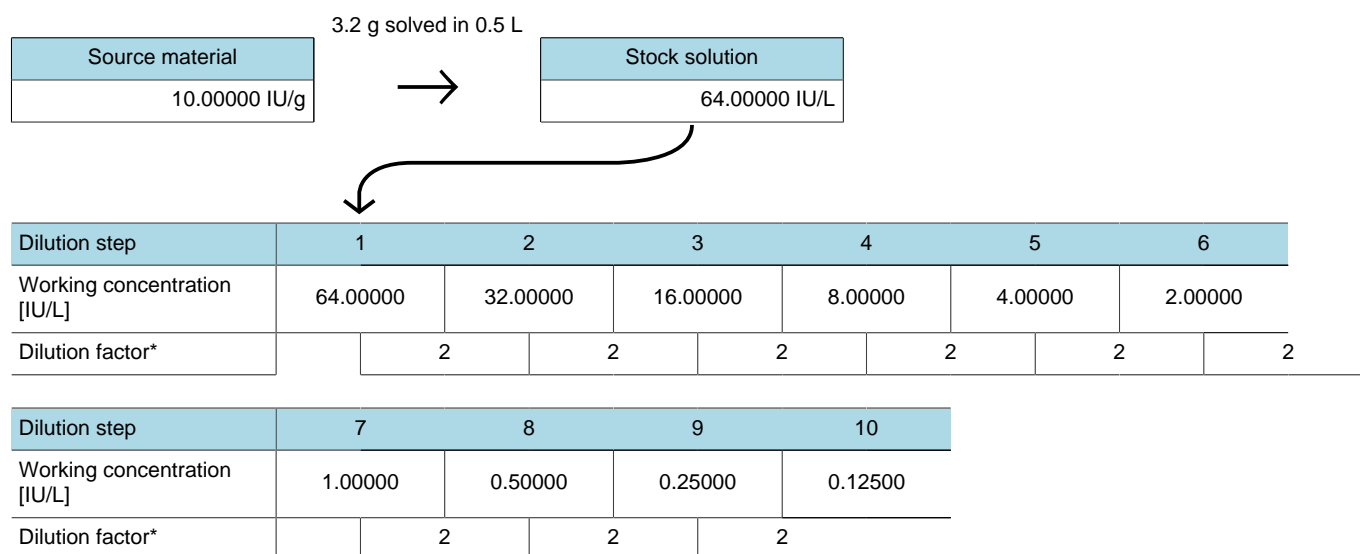
Assay elements	Calculated working concentration	Calculated absolute concentration	Recovery	Suitability test status
TST1	1.64458 [IU/L]	1.64458 [IU/L]	N/A	Passed
TST2 - Source Material with Predilution	8.68768 [IU/L]	6.25513 [IU/g]	97.74%	Failed (warning)
TST3 - Transformaiton with Predilution	1.38885 [IU/L]	12.49967 [IU/L]	N/A	Passed
TST4 - Mixed Response Data Handling with Stock Solution	2.54936 [IU/L]	2.54936 [IU/L]	84.98%	Passed

TECHNOLOGY PREVIEW - NOT FOR PRODUCTION USE

STD: Setup

Property	Value
Concentration: Defined by source material	10.00000 IU/g
Preparation scheme	MultiDoseScheme-1
Number of replicates	3
Geometric Sequence	2-fold sequence
Number of steps	10
Data selection scheme	FullSequenceScheme
Outlier detection	No outlier detection
Response range	Not restricted

Working concentration



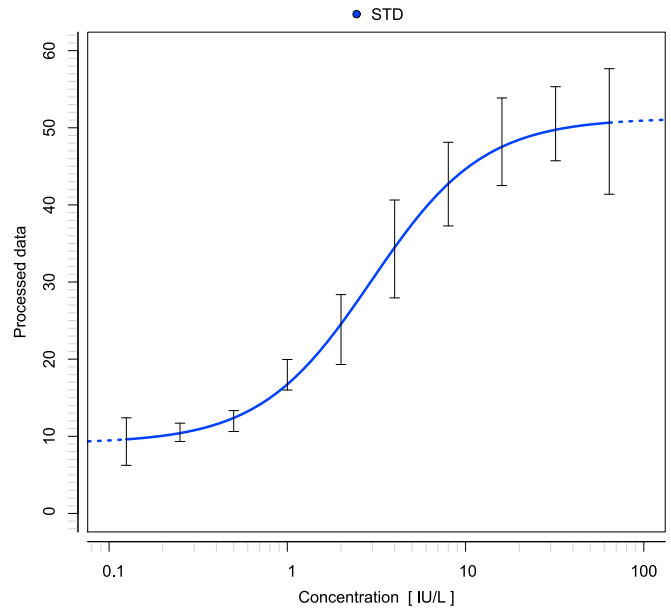
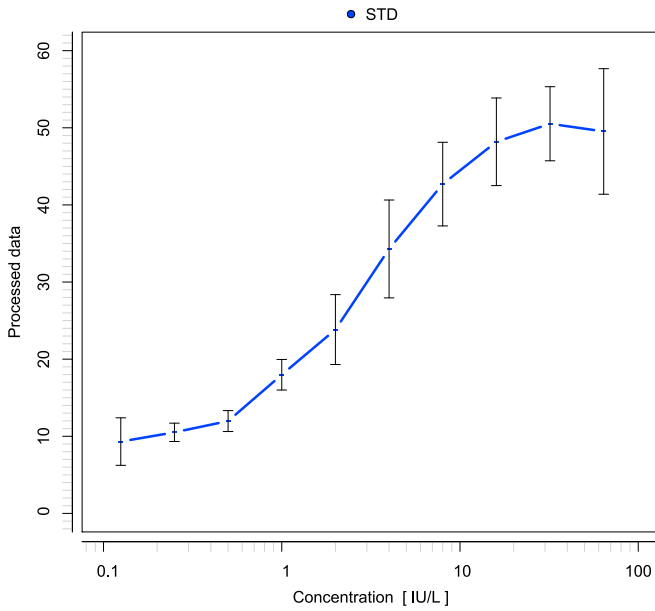
* Dilution factors are defined by the user.

TECHNOLOGY PREVIEW - NOT FOR PRODUCTION USE

STD: Result

Data plot

Regression plot



Response data

Dilution step	1	2	3	4	5	6
Working concentration [IU/L]	64.00000	32.00000	16.00000	8.00000	4.00000	2.00000
Response	48.50084 52.60127 47.47226	48.76707 51.89881 50.91054	48.83845 46.05329 49.66799	44.77683 41.83346 41.48714	31.88864 35.87165 35.11109	24.75387 22.09473 24.66664
Mean response	49.52479	50.52547	48.18658	42.69915	34.29046	23.83841
SD	2.71350	1.60098	1.89347	1.80764	2.11451	1.51070
CV	5.48%	3.17%	3.93%	4.23%	6.17%	6.34%

Dilution step	7	8	9	10
Working concentration [IU/L]	1.00000	0.50000	0.25000	0.12500
Response	18.25924 18.45370 17.22557	11.53347 11.95265 12.43647	10.68759 10.06012 10.79260	10.49572 8.63454 8.81395
Mean response	17.97950	11.97420	10.51343	9.31473
SD	0.66013	0.45189	0.39608	1.02669
CV	3.67%	3.77%	3.77%	11.02%

Analysis of variance (ANOVA)

Source of variation	d.f.	Sum of squares	Mean squares	F-ratio	Probability
Model	3	7869.3605	2623.1202	1052.3393	3.004 E-27
Residual error	26	64.80907	2.49266		
Lack of fit	6	14.04239	2.34040	0.92202	0.50005
Pure error	20	50.76668	2.53833		
Total	29	7934.1695	273.59205		

TECHNOLOGY PREVIEW - NOT FOR PRODUCTION USE

Regression results

Parameter	Estimate	Standard error	Confidence interval (95%)	Width of CI
A upper asymptote	51.24777	0.82313	49.55580 to 52.93974	6.60%
B parameter	1.38702	0.11292	1.15491 to 1.61913	33.47%
C parameter	1.56808	0.08598	1.39135 to 1.74482	22.54%
D lower asymptote	9.08670	0.79349	7.45566 to 10.71773	35.90%
<i>Curve characteristics</i>				
A/D ratio of asymptotes	5.63987	1.33109	-3.67457 to 15.35440	337.40%
A-D difference of asymptotes	42.16107	1.33109	39.42498 to 44.89716	12.98%
EC50	2.96511	N/A	2.62324 to 3.35153	24.56%

	Quality of regression
r ²	0.99183
r ² adjusted	0.99089
Iterations	32
	Reduced accuracy
Final tolerance	1.490 E-08
Reason for termination	Relative error in the sum of squares is at most `ftol'.

Effective concentration (EC)

	25%	50%	75%
Percentile			
EC [IU/L]	1.34292	2.96511	6.54682
Response at EC	19.62697	30.16724	40.70750

Sample suitability tests

	Passed	Failed (rejected)	Failed (warning)	Info
Assay suitability tests	3	0	0	0
Sample suitability tests	1	0	0	0
Overall test result	Passed			

<p>■ STD Test: Mean squares (ANOVA): Pure Error Scope: all assay elements; Severity level: warning</p>		Passed
Calculated value	2.53833	
Upper margin	25.00000	

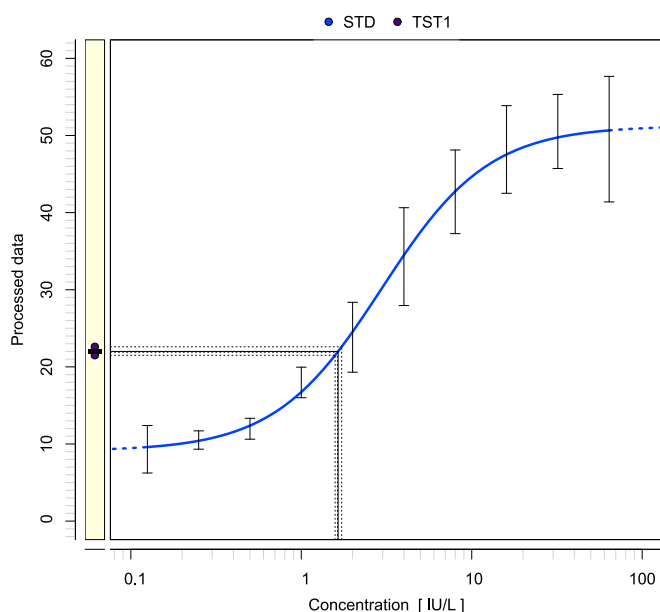
TECHNOLOGY PREVIEW - NOT FOR PRODUCTION USE

TST1: Setup

Property	Value
Preparation scheme	SingleDoseScheme-1
Number of replicates	3
Data selection scheme	FullSequenceScheme
Outlier detection	No outlier detection
Response range	Not restricted

TST1: Result

Interpolation plot



Response data

	Raw Data
Response(s)	21.90000 21.50000 22.60000
Mean response	22.00000
SD	0.55678
CV	2.53%

Interpolation results

Property	Value
Mean response	22.00000
Calculated concentration of the working sample [IU/L]	1.64458
Calculated concentration of the sample stock solution [IU/L]	1.64458

Sample suitability tests

	Passed	Failed (rejected)	Failed (warning)	Info
Assay suitability tests	3	0	0	0
Sample suitability tests	2	0	0	0
Overall test result	Passed			

■ TST1 Test: Interpolation interval: Working concentration Scope: Test samples only; Severity level: warning		Passed
Calculated value	1.57901 ; 1.72491	
Margins	1.00000 to 10.00000	

TECHNOLOGY PREVIEW - NOT FOR PRODUCTION USE

■ TST1 Test: Interpolation interval: Working concentration Scope: all assay elements; Severity level: rejection		Passed
Calculated value	1.57901 ; 1.72491	
Margins	0.10000 to 100.00000	

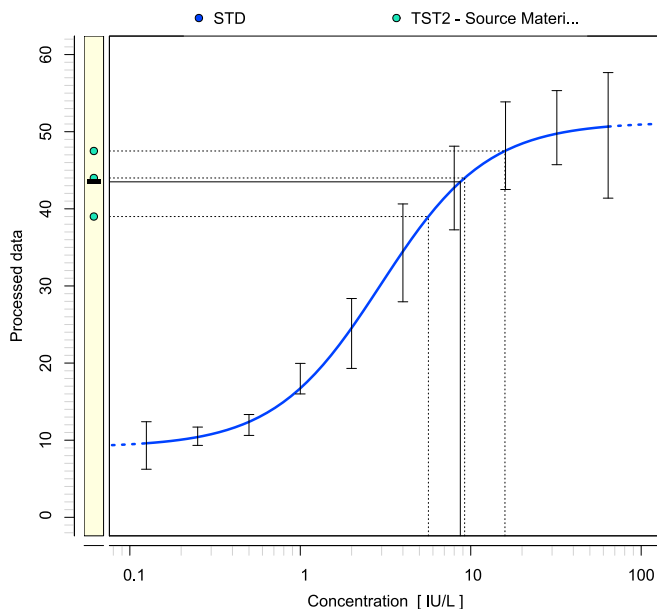
TECHNOLOGY PREVIEW - NOT FOR PRODUCTION USE

TST2 - Source Material with Predilution: Setup

Property	Value
Concentration: Defined by source material	6.40000 IU/g
Preparation scheme	SingleDoseScheme-2
Number of replicates	3
Predilution factors	9
Data selection scheme	FullSequenceScheme
Outlier detection	No outlier detection
Response range	Not restricted

TST2 - Source Material with Predilution: Result

Interpolation plot



Response data

	Raw Data
Response(s)	39.00000 47.50000 44.00000
Mean response	43.50000
SD	4.27200
CV	9.82%

Interpolation results

Property	Value
Mean response	43.50000
Calculated concentration of the working sample [IU/L]	8.68768
Calculated concentration of the sample stock solution [IU/L]	78.18913
Calculated concentration of the sample source material [IU/g]	6.25513
Defined concentration of the sample source material [IU/g]	6.40000
Recovery	97.74%

Sample suitability tests

	Passed	Failed (rejected)	Failed (warning)	Info
Assay suitability tests	3	0	0	0
Sample suitability tests	1	0	1	0
Overall test result	Failed (warning)			

TECHNOLOGY PREVIEW - NOT FOR PRODUCTION USE

■ TST2 - Source Material with Predilution Test: Interpolation interval: Working concentration Scope: Test samples only; Severity level: warning		Failed
Calculated value	5.64466 ; 15.87561	
Margins	1.00000 to 10.00000	Passed
■ TST2 - Source Material with Predilution Test: Interpolation interval: Working concentration Scope: all assay elements; Severity level: rejection		
Calculated value	5.64466 ; 15.87561	
Margins	0.10000 to 100.00000	

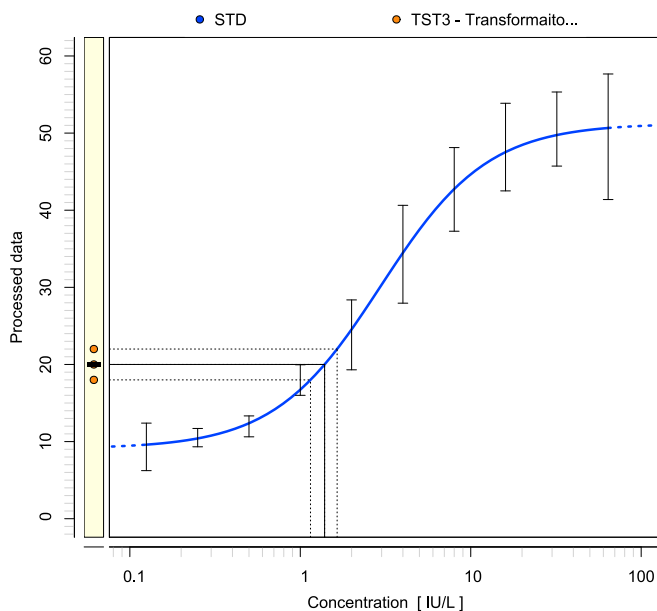
TECHNOLOGY PREVIEW - NOT FOR PRODUCTION USE

TST3 - Transformaiton with Predilution: Setup

Property	Value
Preparation scheme	SingleDoseScheme-2
Number of replicates	3
Predilution factors	9
Data selection scheme	FullSequenceScheme
Outlier detection	No outlier detection
Response range	Not restricted

TST3 - Transformaiton with Predilution: Result

Interpolation plot



Response data

	Raw Data
Response(s)	484.00000 324.00000 400.00000
Mean response	402.66667
SD	80.03333
CV	19.88%

Interpolation results

Property	Value
Mean response (processed data)	20.00000
Calculated concentration of the working sample [IU/L]	1.38885
Calculated concentration of the sample stock solution [IU/L]	12.49967

Sample suitability tests

	Passed	Failed (rejected)	Failed (warning)	Info
Assay suitability tests	3	0	0	0
Sample suitability tests	2	0	0	0
Overall test result	Passed			

TST3 - Transformaiton with Predilution		Passed
Test: Interpolation interval: Working concentration Scope: Test samples only; Severity level: warning		
Calculated value	1.14774 ; 1.64458	
Margins	1.00000 to 10.00000	

TECHNOLOGY PREVIEW - NOT FOR PRODUCTION USE

■ **TST3 - Transformaiton with Predilution**

Test: Interpolation interval: Working concentration
Scope: all assay elements; Severity level: rejection

Passed

Calculated value	1.14774 ; 1.64458
Margins	0.10000 to 100.00000

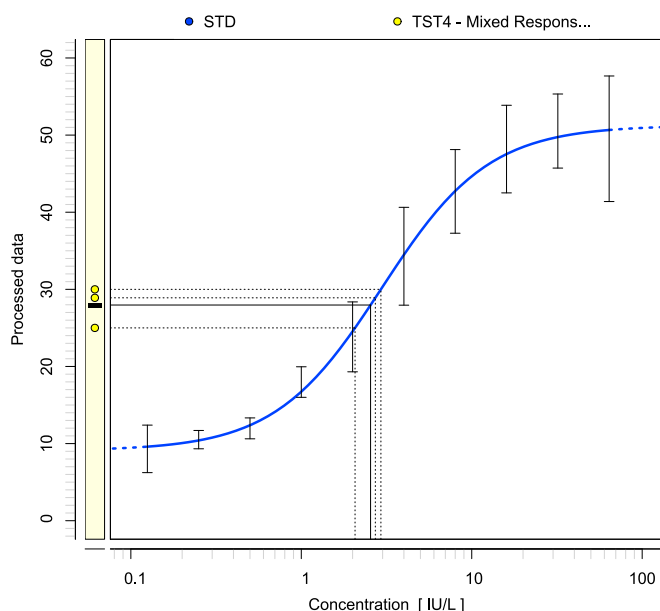
TECHNOLOGY PREVIEW - NOT FOR PRODUCTION USE

TST4 - Mixed Response Data Handling with Stock Solution: Setup

Property	Value
Concentration: Defined by stock solution	3.00000 IU/L
Preparation scheme	SingleDoseScheme-1
Number of replicates	3
Data selection scheme	FullSequenceScheme
Outlier detection	No outlier detection
Response range	Not restricted

TST4 - Mixed Response Data Handling with Stock Solution: Result

Interpolation plot



Response data

	Raw Data
Response(s)	260.00000 250.00000 257.80000
Mean response	255.93333
SD	5.25484
CV	2.05%

Interpolation results

Property	Value
Mean response (processed data)	27.96667
Calculated concentration of the working sample [IU/L]	2.54936
Calculated concentration of the sample stock solution [IU/L]	2.54936
Defined concentration of the sample stock solution [IU/L]	3.00000
Recovery	84.98%

Sample suitability tests

	Passed	Failed (rejected)	Failed (warning)	Info
Assay suitability tests	3	0	0	0
Sample suitability tests	2	0	0	0
Overall test result	Passed			

TST4 - Mixed Response Data Handling with Stock Solution		Passed
Test: Interpolation interval: Working concentration Scope: Test samples only; Severity level: warning		
Calculated value	2.06705 ; 2.93138	
Margins	1.00000 to 10.00000	

TECHNOLOGY PREVIEW - NOT FOR PRODUCTION USE

■ TST4 - Mixed Response Data Handling with Stock Solution

Test: Interpolation interval: Working concentration

Scope: all assay elements; Severity level: rejection

Passed

Calculated value	2.06705 ; 2.93138
Margins	0.10000 to 100.00000