Immuno-Assay Validation Report

Facility:
N°

Study director:

BIOMARKER / ANALYTE: *Precise the most common names of the analyte which will be measured by the assay.*

TYPE OF ASSAY:
☐ Microtiter-competitive ELISA
☐ Microtiter-sandwich ELISA
☐ Bead-based immunoassay
☐ Microtiter multiplex MSD

TYPE OF VALIDATION:
☐ Pre-exploratory validation *(for PoT studies)*
☐ Exploratory validation *(during PoT studies)*
☐ Pre-confirmatoryvalidation *(for PoP studies)*

REAGENTS:
☐ Kit……Source:……Batch number:……Date of production:……

☐ Antibody……Source:……Batch number:……Date of production:……

☐ Antigene……Source:……Batch number:……Date of production:……

☐ Standard……Source:……Batch number:……Date of production:……

Date of validation study completed:
Operator(s):
Date:

Validation procedure

See SOP: SAFE-T Standard Validation Procedure for Biomarker immunoassays.

Objectives / Intended use of the assay:

To demonstrate that the performance characteristics of the assay make it fit for the intended analytical application.
Define the intended use of the immune-assay (biological fluid, disease, etc).

Immuno-assay validation plan

Related to Immuno-Assay Validation Plan N°:
Immuno-Assay validation plan written by:

Validation results

Attach all data record forms and charts (table of all analytical runs with analysis dates).

Validation standards:

Description of the validation standards and matrix used for validation study.
The source and lot number, expiration date, certificates of analyses when available, and/or internally or externally generated evidence of identity and purity should be furnished for each reference standard.

Calibration curve

The calibration curve should be shown with the fitting model clearly indicated (e.g. 4PL, 5PL, etc...).
A table for observed and fitted data should be provided.

Assay dynamic range
LLOQ:
ULOQ:
Give a short reasoning for the determination of the LLOQ and ULOQ.

Sensitivity:
Limit of detection (LOD):
(minimum detectable dose).
Give a short overview of procedure used to determine the LOD.

Precision
Intra-assay precision:
The CV(%) of the middle concentration should be given.
Inter-assay precision:
A table with the mean value, the SD and CV for 5 samples with 3 different concentrations should be shown for inter-assay precision.
Operator-to-operator variability:
Show the effect of the operator on inter-assay variantion with at least 2 operators

Recovery: %
Mean recovery from triplicate measurements for 3 different concentrations covering the assay range

Selectivity / Specificity: % error
Potential interfering substance should be cited.

Parallelism
The parallelism curves and results should be shown.

Dilutional linearity
The dilutional linearity curve and results should be shown.

Robustness
- **Stability**

- *Freeze and Thaw Stability:*
- *Short-Term Temperature Stability*:
- *Long-Term Stability:*
- *Stock Solution Stability:*

- **Reliability**

If data are available

Suitable matrix for analyte detection
Give a short reasoning for suitable matrix types.

Validation evaluation: Acceptance criteria pass/fail
Compare results to acceptance criteria.

Criteria (See validation plan)	**Results**	**Pass/Fail**
Calibration curve:		
appropriate model		☐ / ☐
Assay dynamic range:		
Fitted to target range		☐ / ☐
LLOQ:		
lowest calibrator concentration of the back calculated concentrations that does not exceed a CV of 20% while the recovery is within 75-125%		☐ / ☐
ULOQ:		
highest calibrator concentration of the back calculated concentrations that does not exceed a CV of 20% while the recovery is within 75-125%		☐ / ☐
Sensitivity:		
LOD = 3 times the standard deviation of the background		☐ / ☐
Accuracy:		
		☐ / ☐
Precision:		
Intra-assay precision:		
15% CV for a sample in midrange		☐ / ☐
Inter-assay precision:		
20% CV for the VS samples except for the LLOQ < 30%		☐ / ☐
Recovery:		
100±20%		☐ / ☐
Selectivity / Specificity:		
%error within reasonable range		☐ / ☐
Parallelism:		
		☐ / ☐
Dilution linearity:		
Stability:		
Freeze and Thaw Stability:		
deviation should be within ±25%		☐ / ☐
Short-Term Temperature Stability:		
deviation should be within 15%		☐ / ☐
Long-Term Stability:		

deviation should be within ±25%_____ ☐ / ☐

Stock Solution Stability:

deviation should be within ±15%_____ ☐ / ☐

Validation conclusion

Is the assay validated?

Protocol deviations / descriptions and justifications

Additional data and information relevant to the validation study

All additional data needed for the validation report (Information about antibodies or reagent, Assay procedure, Calculations and Statistical Analyses, …)

Validation report approval

Study director Name:
Facility:

Validation report approved: ☐ **Yes** ☐ **No**

Comments:

Date:

Study director signature: