Validation of analytical methods as an evaluation tool for research data reliability. Functional Ecology

Anne Jaulin, Elodie Ollivier, Nathalie Cheviron, Virginie Grondin, Amelie Trouve

▶ To cite this version:

Anne Jaulin, Elodie Ollivier, Nathalie Cheviron, Virginie Grondin, Amelie Trouve. Validation of analytical methods as an evaluation tool for research data reliability. Functional Ecology. Functional Ecology Conference - JEF AnaEE France, Dec 2018, Nancy, France. 2018. hal-02786058
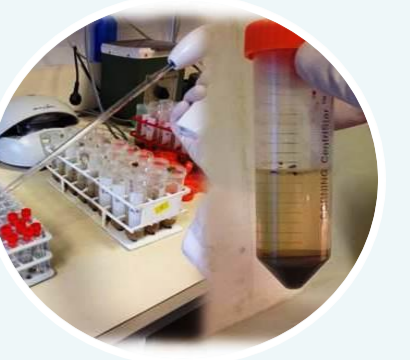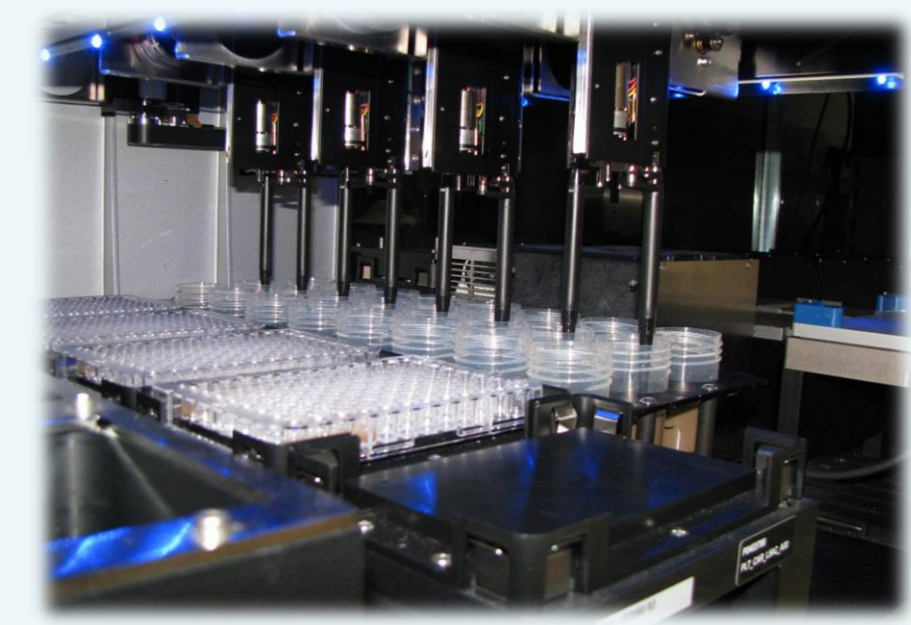
HAL Id: hal-02786058
https://hal.inrae.fr/hal-02786058v1

Submitted on 4 Jun 2020

BIOCHEM ENV
La Biochimie Environnementale
au service de la Recherche
www.biochemenv.fr
https://twitter.com/biochemenv

CERTIFIED LR ISO 9001

VALIDATION OF ANALYTICAL METHODS AS AN EVALUATION TOOL FOR RESEARCH DATA RELIABILITY

Anne JAULIN*, Elodie OLLIVIER, Nathalie CHEVIRON, Virginie GRONDIN, Amélie TROUVE

UMR ECOSYS INRA, AgroParisTech, Université Paris-Saclay, plateforme Biochem-Env, 78026, Versailles, France

* anne.jaulin@inra.fr   contact-biochemenv@inra.fr

Context

The platform Biochem-Env:
➤ Was created in 2012 by INRA (French National Institute for Agricultural Research) with the support of the ANR program "Investissements d'avenir" as a service of the infrastructure ANAEE-France,
➤ For the **biochemical characterization of natural environments** (soils and sediments) and **associated macrofauna** in research projects,
➤ By developing and validating methods in order to **provide traceable analytical data with high level of confidence.**

For intra-laboratory validation of quantitative analytical methods, the INRA's Quality Guidelines for research and experimental units (2013) recommends **"the accuracy profile" method** according to the **NF V03-110:2010 standard**.

Could we use a same internally developed method to quantify proteins in various biological models ?
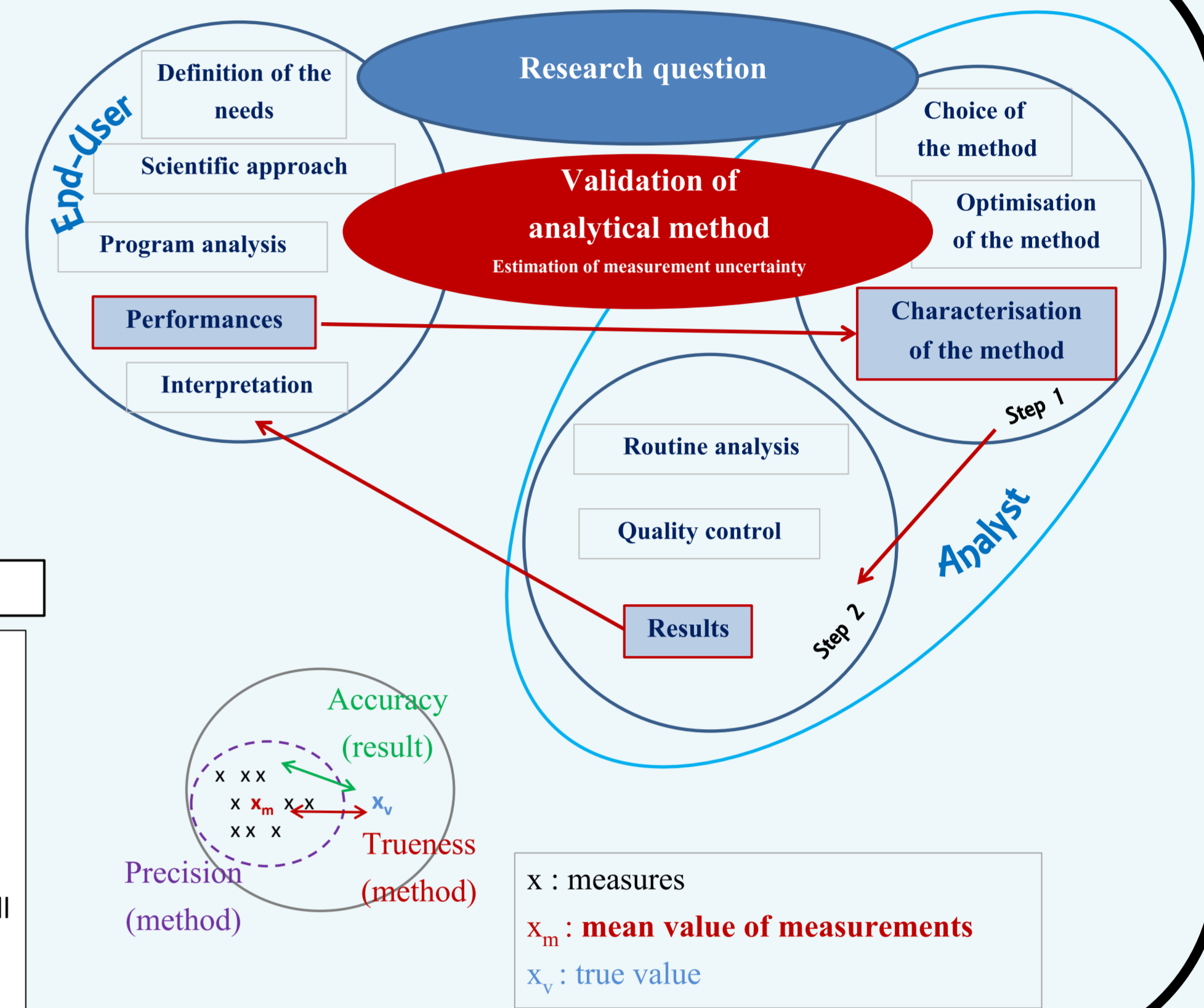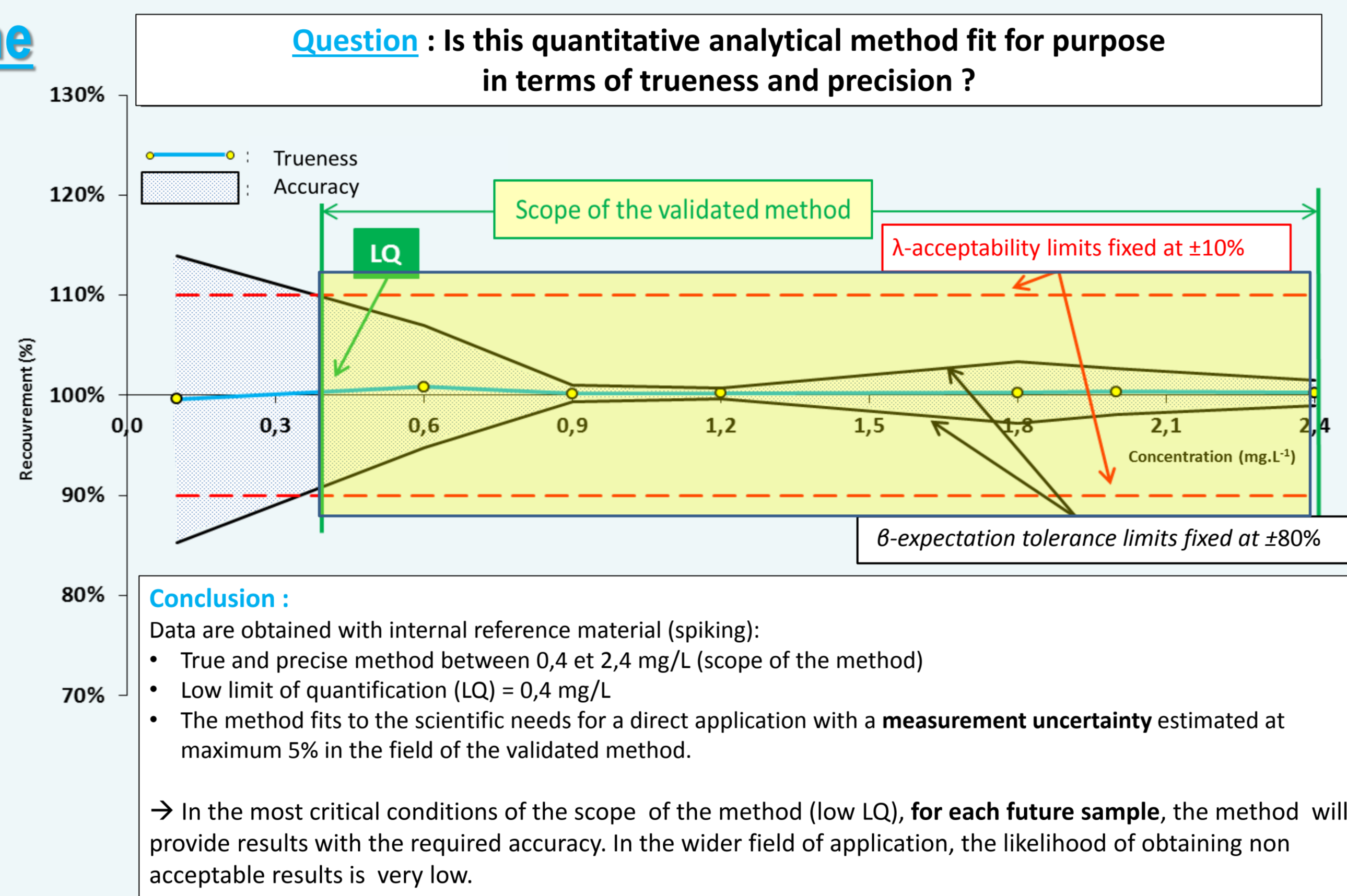
Validation of analytical methods by the accuracy profile approach

Purpose:
➤ To provide **guarantees on analytical results**, for the analyst and the end-user
➤ To demonstrate analytical **method fitting with the scientific objectives**
➤ To allow **laboratory recognition**
➤ To **improve** analysts **working practices**

Benefits of the accuracy profile approach:
➤ An **overall statistical method** combining **trueness and precision**
➤ A **standardized approach : NF V03-110:2010**
➤ **A simple and graphic interpretation** for a **rapid decision**
➤ The determination of the scope of the method
➤ The determination of quantification limits
➤ An estimation of measurement uncertainty

Question : Is this quantitative analytical method fit for purpose in terms of trueness and precision ?

Scope of the validated method — λ-acceptability limits fixed at ±10% — β-expectation tolerance limits fixed at ±80% — LQ

Conclusion :
Data are obtained with internal reference material (spiking):
• True and precise method between 0,4 et 2,4 mg/L (scope of the method)
• Low limit of quantification (LQ) = 0,4 mg/L
• The method fits to the scientific needs for a direct application with a **measurement uncertainty** estimated at maximum 5% in the field of the validated method.

→ In the most critical conditions of the scope of the method (low LQ), **for each future sample**, the method will provide results with the required accuracy. In the wider field of application, the likelihood of obtaining non acceptable results is very low.

x : measures
x_m : mean value of measurements
x_v : true value

Material and methods

Protein determination method by the Bicinchoninic acid
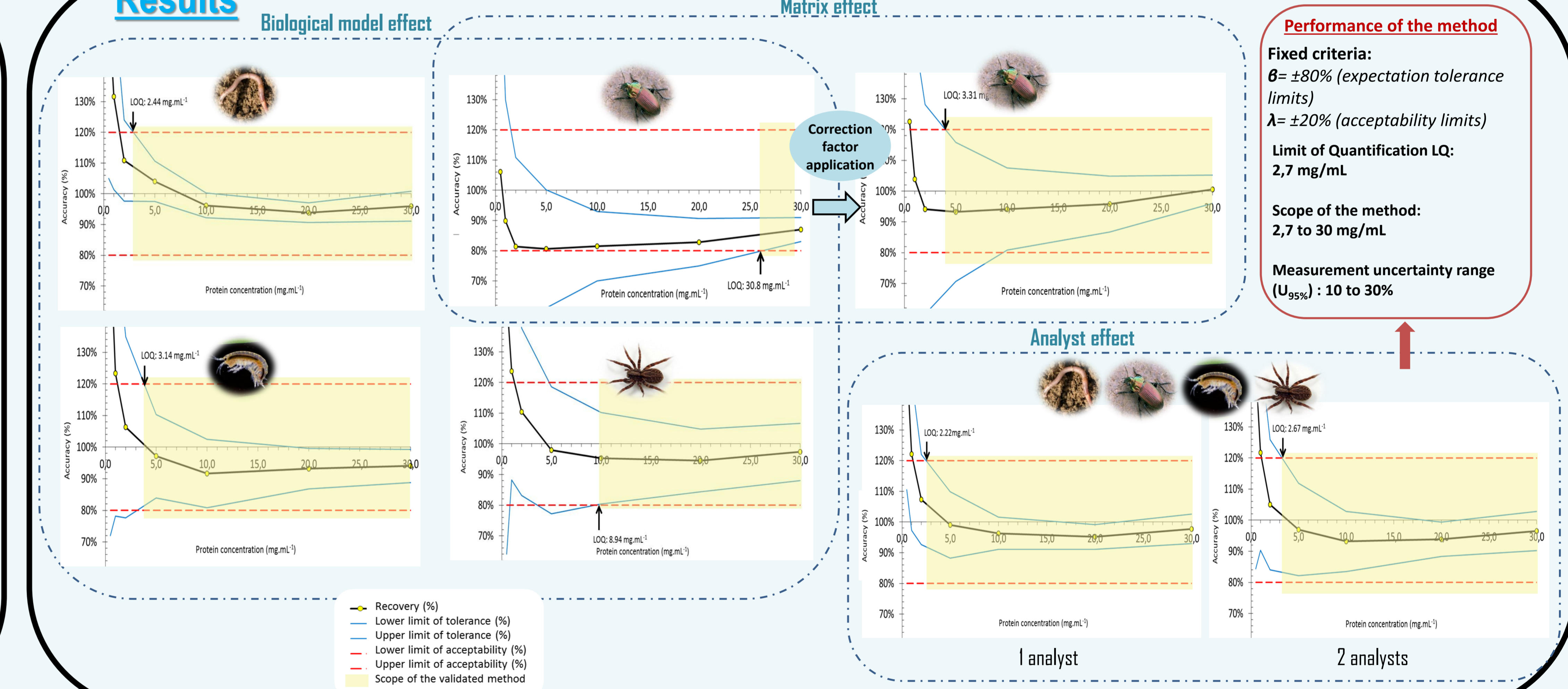*(Ref. QPBCA-1KT, QuantiPro™ BCA Assay Kit, Sigma-Aldrich)*
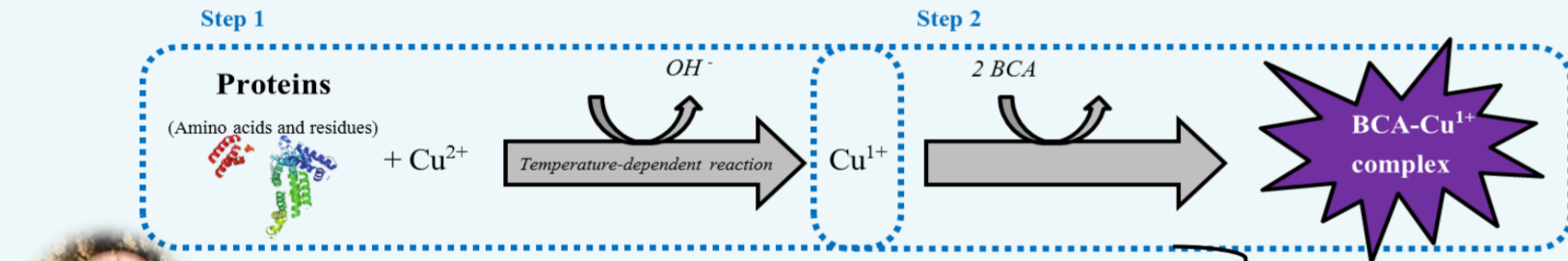
Aporrectodea icterica (earthworm)
Poecilus cupreus (beetle)
Gammarus pulex (gammarid)
Alopecosa pulverulenta (spider)

4 biological models

Validation method according to the « accuracy profile » approach (NF V03-110:2010 standard):
➤ Selection of validation samples:
- Spiked reference material: Bovine Serum Albumine
- 7 levels (0,5 ; 1 ; 2 ; 5 ; 10 ; 20 ; 30 mg.mL^{-1})
- 3 replicates (repeatability)
- 6 days of analysis *(intermediate precision)*
- 2 analysts *(intermediate precision)*
➤ Calibration (indirect quantitative analysis):
- 6 levels (0 ; 2 ; 4 ; 8 ; 16 ; 20 ; 30 ; 40 mg.mL^{-1})
- Regression model : polynomial

Results

Biological model effect — **Matrix effect** — **Analyst effect**

Correction factor application

Recovery (%)
Lower limit of tolerance (%)
Upper limit of tolerance (%)
Lower limit of acceptability (%)
Upper limit of acceptability (%)
Scope of the validated method

1 analyst — 2 analysts

Performance of the method

Fixed criteria:
β= ±80% (expectation tolerance limits)
λ= ±20% (acceptability limits)

Limit of Quantification LQ:
2,7 mg/mL

Scope of the method:
2,7 to 30 mg/mL

Measurement uncertainty range (U$_{95\%}$) : 10 to 30%

Conclusions

Accuracy profile approach:
- **Global statistical combination of trueness and precision** and pragmatic
- **Simple graphic interpretation**, allowing a clear and easy comparison between method performance and intended use and, a **rapid decision**
- No limit in the choice of the calibration model => **large scope range**
- Methods with very low variability can be validated (not rejected by a H0)
- **Diagnostic tool**, matrix effect taken into account
- Risks and guarantees managed for both end-users and laboratories
- **Estimation of measurement uncertainty**

The validation of this analytical method helped us to:
- Determine the performance of the method
- Improve the steps for sample preparation and analysis
- Assess the matrix effect
- Pointed out the importance of an experimented analyst

Decision on the fitness for purpose

➤ **Analytical method for protein quantification in 4 biological models was validated with a good accuracy considering scientific specification and needs.**

Perspectives :
- Adapt λ-acceptability values according to the concentration range
- Extend the method to other biological models and biomarkers (Lipid, glycogen…)

References

[1] Validation des méthodes d'analyse quantitative par le profil d'exactitude. Numéro spécial du Cahier des Techniques de l'INRA, 2010.

[2] Labo-Stat : Guide de validation des méthodes d'analyse, Feinberg M. Retirage 2012.

[3] Jaulin A. et Deschamps M. La validation de méthodes d'analyse quantitative, c'est l'affaire de tous ! La démarche Qualité au service des activités de recherche. Journées de la mesure et de la métrologie J2M 2012 du 8-11/10/2012, Le Croisic.

[4] Feinberg M. et Laurentie M. A global approach to method validation and measurement uncertainty. 2006. Accred. Qual. Assu. 11, 3-9.

[5] Feinberg M. et al. New advances in method validation and measurement uncertainty aimed at immroving the quality of chemical data. 2004. Anal. Bioanal. Chem. 380 : 502-514.

FUNCTIONAL ECOLOGY CONFERENCE NANCY 2018