Analytical risk assessment from bioanalytical method validation results

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INTRODUCTION

During a bioanalytical method validation [1], accuracy and precision are determined from QC samples at different concentration levels (the accuracy mean bias should be within 15% and precision RSD should not exceed 15% of the nominal value). During routine analysis, the QC samples provide the basis of accepting or rejecting the run. The aim of this poster is to present tools for anticipating the risk of rejecting a run according to validation results.

MATERIALS AND METHODS

Tool 1 = Operating characteristic curves (OCC)

This graphic tool is adapted from the OCC concept [2] to determine the probability to have one QC sample out of the acceptance criteria (+/- 15% of nominal concentration). In order to evaluate risks on real case studies and understand the use of the OCC tool, 5 examples of QC results with different bias and RSD from during validation studies are presented in Figure 1.

This tool uses the following Excel formula:
RESULT = NORMDIST(USL, BIAS, RSD,TRUE) - NORMDIST(LSL, BIAS, SD, TRUE)
with :
- *USL* and *LSL* = Upper and Lower Specification Limits (-15% and +15%, respectively);
- *BIAS* = percentage bias estimated during accuracy step of method validation [1] (= 100 – mean recovery) ;
- *RSD* = Relative Standard Deviation estimated during validation [1];
- *TRUE* = logical operator.

The corresponding *RESULT* is the probability to get an individual QC sample within the 15% limits and *(1-RESULT)* is then the risk to reject this QC sample.

Setting levels of risk from low (0.01%) to high (50%) allowed to draw the relationship between RSD and BIAS to reach the selected risk value and to draw an OCC for each one, as presented in Figure 2.

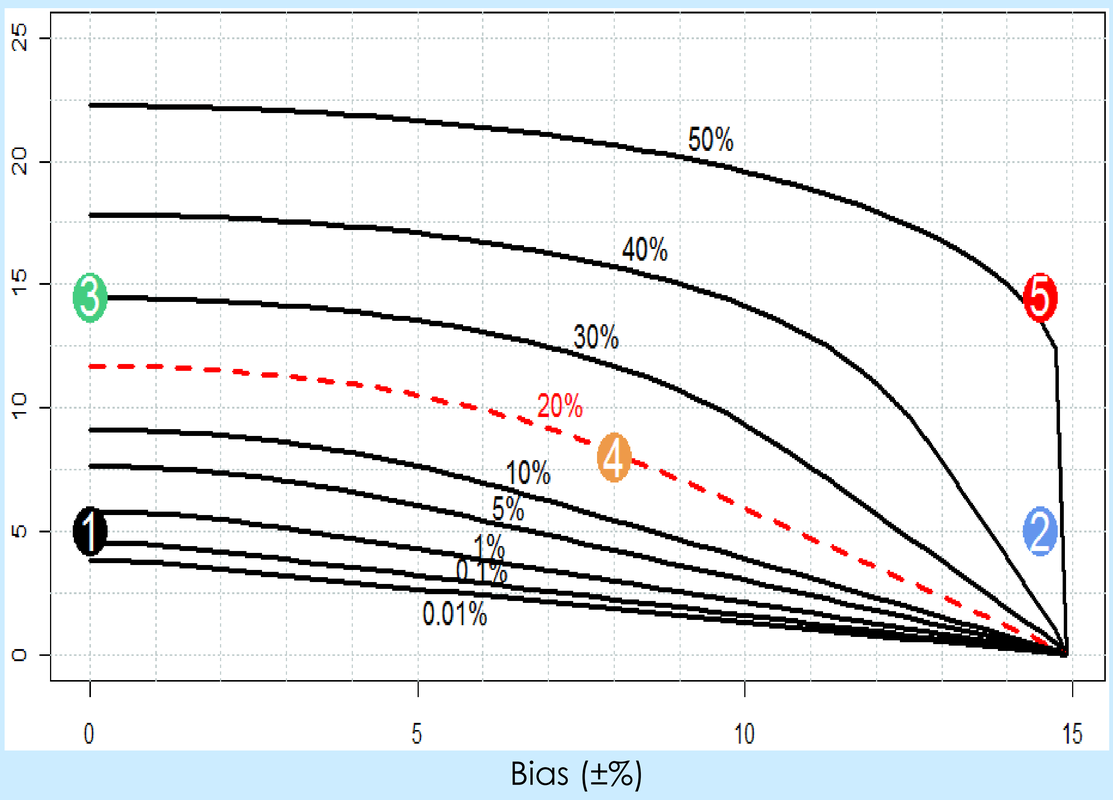

Figure 2: Assessment of risk levels of having a single QC value out of +/- 15% limits, according to method performances in terms of accuracy (BIAS) and precision (RSD). The dotted line corresponds to an acceptable risk estimated at 20% by the authors.

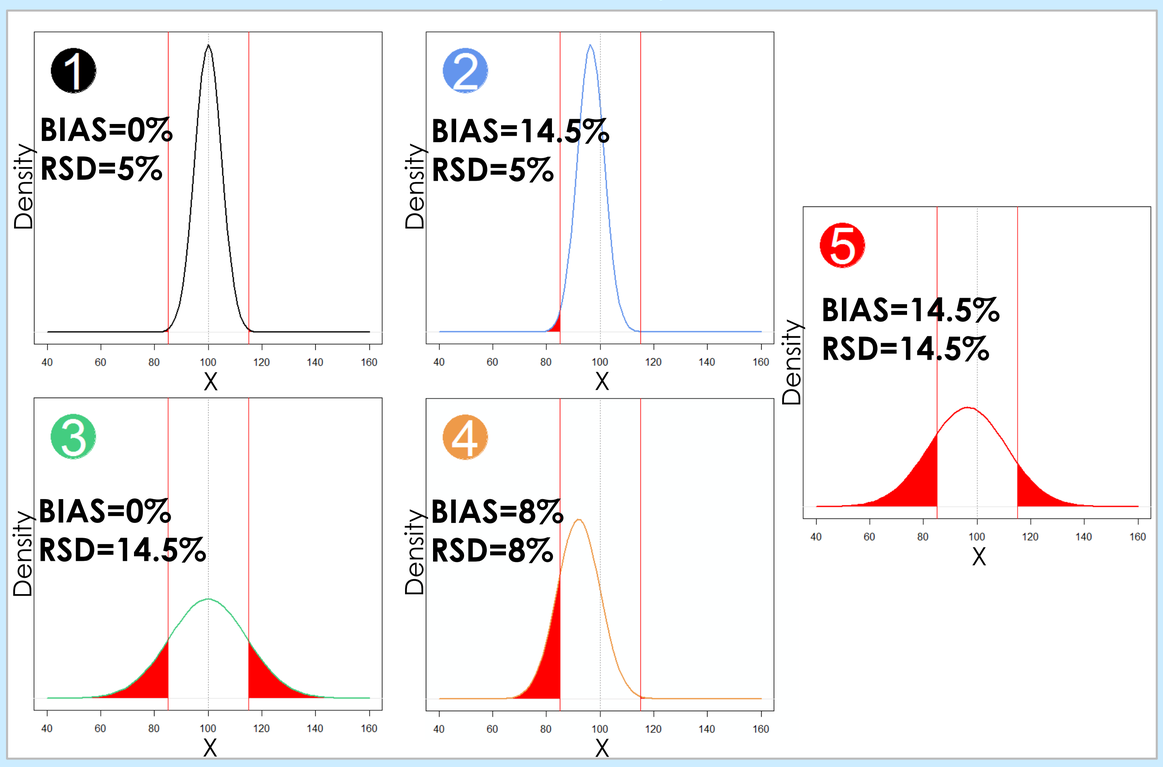

Figure 1: Normal distribution and its probability to be out of acceptance criteria (area in red) for 5 different examples of QC results from validation study

Tool 2 = Probability to reject an analytical run

The total probability to reject a run is the sum of the probability to have both QC samples at the same concentration level out of +/-15% limits and the probability to have more than 1/3 of QC samples out of +/-15% limits.

The total probability can be a decision tool to assess if the method provides acceptable results (low probability to reject analytical runs) or not, depending on the use of the method.

If large sets of samples for clinical studies require several analytical runs, and if the risk is high, you may reconsider the method and try to improve it. It can be noticed that the risk of having poor results with incurred samples was not calculated here, and is therefore not taken into account.

RESULTS

Different scenarios for QC results presented in Figure 1 are combined in order to have 3 QC levels with different risks, as for a bioanalytical run with QCs at low, medium and high concentration levels. The combination tested and the corresponding total probability are described in the table below.

	Low QC	Medium QC	High QC	Total Probability
Scenario 1	1	2	3	30.3%
Scenario 2	1	2	5	47.0%
Scenario 3	1	2	4	24.9%
Scenario 4	2	3	4	36.6%
Scenario 5	1	3	4	12.8%
Scenario 6	1	1	1	0.0%
Scenario 7	3	4	5	41.4%
Scenario 8	5	5	5	90.2%

Even if each QC samples is always within acceptance criteria, the total probability varies from a "no risk scenario" (scenario 6) to a "very high risk level" (scenario 8).
This high variability in the risk levels shows that validating a method is not sufficient to ensure a smooth routine testing. A in-depth review of validation results can help for a better evaluation of such risks.

Note that the worst case of QC samples 5 has a strong influence on the total probability calculation (High QC for scenario 2, for example). As shown in Figure 2, this QC performance induces a rejection risk for a single QC sample above 50%. Such a risk level should indeed be avoided and a method optimization should be considered.

CONCLUSION

The Operating Characteristic Curves (OCC) combined with the total probability approach are useful tools for method development analysts in order to check if the developed method will be able enough to generate acceptable analytical runs. Transfer of Knowledge about method performances between development laboratories and analysts involved in routine testing of clinical trials may provide a sound scientific argumentation regarding results obtained out of the predefined limits. As the risks you can accept may decrease during the development of your product, the same approach can also be used as an evolving tool for method performance evaluation during its life cycle management.

These tools are currently used in the Bioanalysis Laboratory of Amatsigroup through different methods/types of studies. A review will then be done to evaluate the extent of benefit of such tools.

REFERENCES

[1] FDA Draft Guidance "Bioanalytical Method Validation", Revision 1, September 2013
[2] Small Molecules Collaborative Group: « Performance based monographs », Pharm. Forum 35 (3), 2009