Method Validation by Design to Support Formulation Development

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The authors describe a method-validation-by-design (MVbD) approach to validate a method over a range of formulations using both design-of-experiment and quality-by-design principles to define a design space that allows for formulation changes without revalidation. The approach provides the required International Conference on Harmonization validation elements as well as information on interactions, measurement uncertainty, control strategy, and continuous improvement. Despite being less resource intensive than the traditional validation approach, quality is not compromised. Additionally, through judicious planning, the MVbD approach can encompass early formulation design efforts so that a wide range of formulations is taken into consideration when defining the method-validation design space.

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Analytical method validation is a crucial part of formulation development. It is needed to ensure that methods provide accuracy and precision in detecting formulation differences in drug dissolution, stability, active, and impurity levels. Method validation is also key to meeting requirements of cGMP, compendia, and the International Conference on Harmonization (ICH) for testing and releasing clinical or commercial dosage forms. During early development, formulations are often changed to adapt to new preclinical and clinical data. Revalidating each of these new formulations is resource intensive and affects development timelines.

The ICH Q2 (R1) validation guidelines (1) do not provide crucial information such as how formulation changes affect method performance or what method critical validation parameters need to be monitored and controlled. An ICH approach may not provide a good understanding of a method-measurement uncertainty, which is needed to ensure that the overall process capabilities are met and that appropriate in-process controls and specifications are set. This sort of understanding is needed to meet FDA's process validation guidelines (2). Once the product acceptance criteria are established, the influence of assay variation can be determined relative to product acceptance rates.

This article describes a method-validation-by-design (MVbD) approach to validate a method over a range of formulations. It uses both design-of-experiment (DOE) and quality-by-design (QbD) principles to define a design space that allows for formulation changes without revalidation. The approach provides the required ICH validation elements as well as information on interactions, measurement uncertainty, control strategy, and continuous improvement. This approach is less resource intensive than the traditional validation approach without compromising quality. Additionally, through judicious planning, it can encompass early formulation design efforts so that a wide range of formulations can be used to define the method-validation design space.

MVbD enablers

MVbD is not specifically addressed in ICH; however, it is supported by the principles presented in ICH Q8, 9, 10, 11 (3–6) as well as ICH Q2 (R1). There are other industry guidelines (2), publications (7–9) and presentations (10–15) that support a QbD/DOE approach for analytical method development and validation, including FDA's 21st Century Quality Initiative, which was first presented in 2001 and later updated in 2005 (15), and the outcome from FDA's QbD pilot.

Table I: Hypothetical range of formulations. The API, preservative, and Excipient 3 are varied. Q.S. = Quant satis (quantity sufficient).

Ingredients (% w/w)	Placebo formulation	0.005% API formulation	0.01% API formulation	0.015% API formulation	0.02% API formulation	0.025% API formulation
API	0	0.005	0.01	0.015	0.02	0.025
Preservative	0.01 or 0.03	0.01 or 0.03	0.01 or 0.03	0.01 or 0.03	0.01 or 0.03	0.01 or 0.03
Excipient 1	0.3	0.3	0.3	0.3	0.3	0.3
Excipient 2	0.02	0.02	0.02	0.02	0.02	0.02
Excipient 3	0.5 or 0.8	0.5 or 0.8	0.5 or 0.8	0.5 or 0.8	0.5 or 0.8	0.5 or 0.8
Purified water	100% Q.S.	100% Q.S.	100% Q.S.	100% Q.S.	100% Q.S.	100% Q.S.

DOE application

The use of DOE is well established for determining method robustness, such as how much the mobile-phase composition, column temperature, and flow rate can vary. DOE is also well used in formulation screening. This approach broadens DOE to include method validation over a range of formulations.

QbD application

The MVbD approach applies to these QbD principles:

- *Risk management assessment* of the potential critical validation parameters. Systematic analysis (e.g., fishbone diagram) is done based on historical knowledge. The QbD/DOE output confirms those that are critical.
- *Analytical target profile (ATP).* Once the required process capability is known, the required method accuracy and precision acceptance criteria are defined in the ATP.
- *Control strategy.* DOE output defines those parameters that have the most impact on the method performance. Monitoring these parameters throughout development will help define acceptable ranges.
- *Continuous improvement.* MVbD provides the acceptance criteria that must be met to move to new technologies.
- *Knowledge management.* Knowledge gained is cycled backed to accelerate the next program.

Traditional versus DOE approaches

Traditionally, each new drug-product formulation requires validation to support clinical testing. In early development, the method is validated for linearity, accuracy, and precision. Method linearity is determined across five concentrations from 50% to 150% of the nominal (100%) concentrations. Accuracy and precision can be combined with the linearity study by doing six replicates at the 100% (nominal) concentration and three replicates at the other concentrations (50%, 75%, 125%, and 150%), which gives a total of 18 sample preparations. If there are five new formulations during development that require separate validation, a total of 90 sample preparations would be needed. This approach is resource intensive and affects project timelines. In this given example, only one formulation component is varied. As more components are varied, the workload increases proportionally.

The challenge with this approach is:

- It does not statistically define a design space
- It does not employ QbD principles

A QbD/DOE approach validates a method over a range of formulations within a defined design space. Movement within this design space does not require method revalidation with each new formulation. The DOE approach detects any excipient–API interactions and critical validation control parameters.

A DOE approach is not necessarily more labor intensive than the traditional method-validation approach and additional information gained is well worth the exercise. As described, the traditional approach requires at least 90 sample preparations to validate five formulations over the course of the development. Using DOE to validate across a broader range that includes these five formulations would require only three determinations for each of the five formulations, resulting in a total of 15 sample preparations.

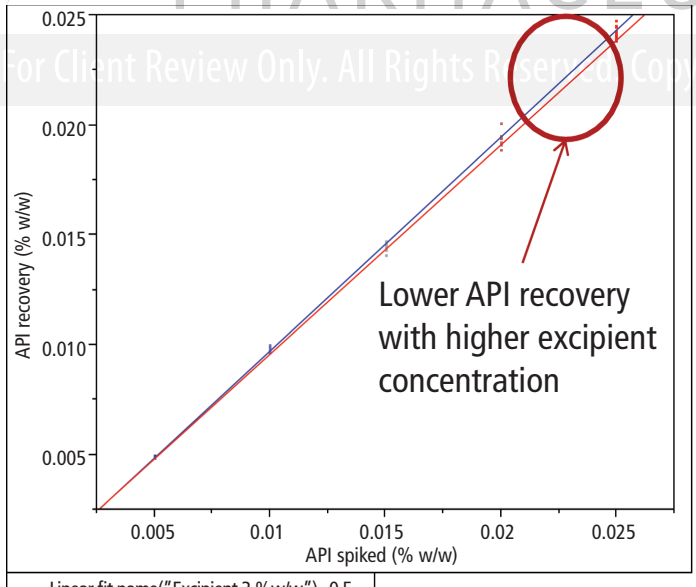
Steps to perform a MVbD

- *Step 1: Define the range of formulations and required process capability.* A formulation screening DOE may be used to determine all possible clinical formulations for the study. Manufacturing process capabilities need to be considered to define the ATP (i.e., the necessary accuracy and precision to ensure acceptable data). Full factorial, fractional factorial and/or custom (d-optimal) design all may be used in developing the study design.
- *Step 2: High-performance liquid chromatography (HPLC) method selection.* A method should be developed such that sample preparations of the different formulation can be diluted to the same API concentrations. The diluted sample from each formulation is injected onto the HPLC column under the same chromatography conditions. Standardizing these conditions will facilitate DOE and latter method-robustness studies.
- *Step 3: DOE design.* The set up of the DOE can be accomplished with or without using DOE software. Inputs to the DOE include a number of varying factors, such as the different API levels and/or excipient and preservative levels.
- *Step 4: ICH statistics and design space.* Data are analyzed for linearity, precision, and accuracy, including confidence intervals. Any interactions are reported and a design space

Figure 1: A design-of-experiment (DOE) approach for three factors showing the DOE pattern, API, preservative, Excipient 3 spiked concentrations along with simulated percent recovery data

	Pattern	API w/w%	Preservative w/w%	Excipient 3 w/w%	Percent recovery
2	1---	0.005	0.01	0.5	98.0
3	1---	0.005	0.01	0.5	97.7
4	1--+	0.005	0.01	0.8	95.6
5	1--+	0.005	0.01	0.8	96.2
6	1--+	0.005	0.01	0.8	96.6
7	1+--	0.005	0.03	0.5	97.4
8	1+--	0.005	0.03	0.5	97.1
9	1+--	0.005	0.03	0.5	97.8
10	1++	0.005	0.03	0.8	96.2
11	1++	0.005	0.03	0.8	96.6
12	1++	0.005	0.03	0.8	96.0
13	2---	0.01	0.01	0.5	99.6
14	2---	0.01	0.01	0.5	98.4
15	2---	0.01	0.01	0.5	97.6
16	2--+	0.01	0.01	0.8	96.3
17	2--+	0.01	0.01	0.8	95.7
18	2--+	0.01	0.01	0.8	97.4
19	2+--	0.01	0.03	0.5	97.4
20	2+--	0.01	0.03	0.5	97.2
21	2+--	0.01	0.03	0.5	97.3
22	2++	0.01	0.03	0.8	96.0
23	2++	0.01	0.03	0.8	96.9
24	2++	0.01	0.03	0.8	95.9
25	3---	0.015	0.01	0.5	95.5
26	3---	0.015	0.01	0.5	96.0
27	3---	0.015	0.01	0.5	96.5
28	3--+	0.015	0.01	0.8	93.5
29	3--+	0.015	0.01	0.8	96.8
30	3--+	0.015	0.01	0.8	95.8
31	3+--	0.015	0.03	0.5	96.6
32	3+--	0.015	0.03	0.5	97.6
33	3+--	0.015	0.03	0.5	97.1
34	3++	0.015	0.03	0.8	96.2
35	3++	0.015	0.03	0.8	95.6
36	3++	0.015	0.03	0.8	95.2
37	4---	0.02	0.01	0.5	100.2
38	4---	0.02	0.01	0.5	97.4
39	4---	0.02	0.01	0.5	96.9
40	4--+	0.02	0.01	0.8	94.1
41	4--+	0.02	0.01	0.8	95.2
42	4--+	0.02	0.01	0.8	95.2
43	4+--	0.02	0.03	0.5	96.7
44	4+--	0.02	0.03	0.5	97.2
45	4+--	0.02	0.03	0.5	96.6
46	4++	0.02	0.03	0.8	96.0
47	4++	0.02	0.03	0.8	95.8
48	4++	0.02	0.03	0.8	95.6
49	5---	0.025	0.01	0.5	98.9
50	5---	0.025	0.01	0.5	96.8
51	5---	0.025	0.01	0.5	96.6
52	5--+	0.025	0.01	0.8	95.6
53	5--+	0.025	0.01	0.8	95.8
54	5--+	0.025	0.01	0.8	95.2
55	5+--	0.025	0.03	0.5	97.9
56	5+--	0.025	0.03	0.5	97.0
57	5+--	0.025	0.03	0.5	97.6
58	5++	0.025	0.03	0.8	96.2
59	5++	0.025	0.03	0.8	95.6
60	5++	0.025	0.03	0.8	95.8

Figure 2: Linearity data for API recovered versus API spiked for formulations containing 0.05% and 0.08% (w/w) levels of Excipient 3



Linear fit name("Excipient 3 %w/w")=0.5

API recovery (%w/w) = -6.117 e-6 + 0.9739133*API spiked (% w/w)

Summary of fit

RSquare	0.999422
RSquare adjusted	0.999401
Root mean squre error (RSME)	0.000171
Mean of response	0.014603
Observations (or sum of weights)	30

RSME used to set specifications

is defined. The design space is defined by fitting a model to the factors used in the evaluation of assay concentrations and other key factors that may interfere or influence assay precision or bias. After fitting the model, the design space may be visualized using contour plot or profilers.

Case study

To illustrate the MVbD process, a simulated case study is presented where three factors are varied over a range of formulations (see **Table I**). The API concentrations are varied from 0.005%, 0.01%, 0.015%, 0.02%, and 0.025% (w/w). Preservative concentrations are either 0.01% or 0.03% (w/w) and concentrations of Excipient 3 are either 0.5% or 0.8% (w/w).

The DOE design is shown in **Figure 1**. The DOE pattern is presented as 1, 2, 3, 4 or 5 --, -+, +- or ++. The numbers 1 through 5 represent the five API concentrations and the "+" or "–" represent with or without the two possible preservative and excipient concentrations. Three replicates are done for each combination of API, preservative, and excipient concentration. A total of 60 sample preparations is required. The DOE typically suggests a randomized sample preparation to avoid any sample preparation variability; however, because the variability is low, a standard sample-preparation scheme can be followed.

Figure 1 also presents the percent recovery data. The required ICH statistics can be obtained from these data. For example, **Figure 2** shows linearity data along with typical linearity statistics. One key output is the root mean square error (RMSE); this parameter identifies method variability that is critical for understanding the method contribution to the overall process variability.

Figure 2 also shows linearity plots of percent recovery versus the two excipient concentrations. The difference in slopes indicates interaction (i.e., a lower API recovery at a higher level of Excipient 3 concentration), possibly due to interaction of the excipient with the API. Such information is important for the formulator and method developer to consider if higher preservative levels are needed.

Figure 3 presents the mean percent recovery for each combination of API, preservative, and excipient along with the corresponding confidence limits. In the

example describe, there was a somewhat lower API recovery with the higher excipient levels, which is further illustrated in **Figure 4** as discussed in the following.

Design space. A design space is illustrated in **Figure 4**. It is obtained by mathematically modeling the primary main effects and secondary two-factor interactions as well as possible polynomials using a DOE software. The operating range is the white space shown in **Figure 4**; it is where the method accuracy and precision meet the acceptance criteria. Adding formulations outside this range would require confirmatory testing.

System linearity, quantitation limits (QL), and detection limits (DL). To ensure the sample amount injected is within the linear range of the detector, a separate experiment can be done where the sample concentration is varied from 50% to 150% of the nominal (100%) concentration. If the potency method is also used to quantitate impurities, a wider linearity study can be performed from 100% to 0.03%. The QL and DL for the impurities can be determined by the ratio of RMSE to the slope, as described in ICH Q2 (R1).

Figure 3: Accuracy data with confidence intervals for varied levels of API, preservative and Excipient 3

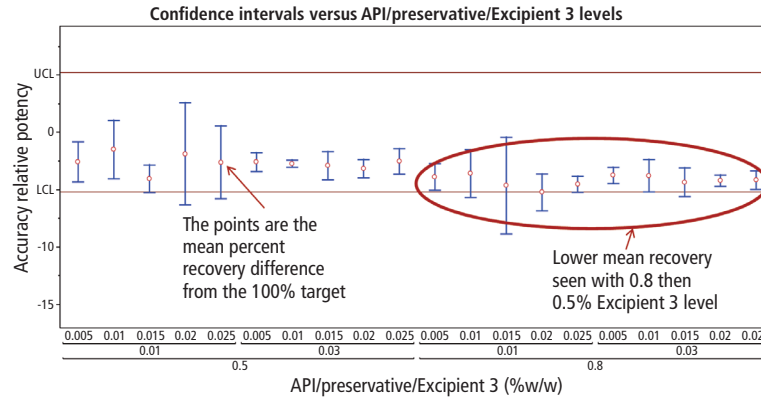
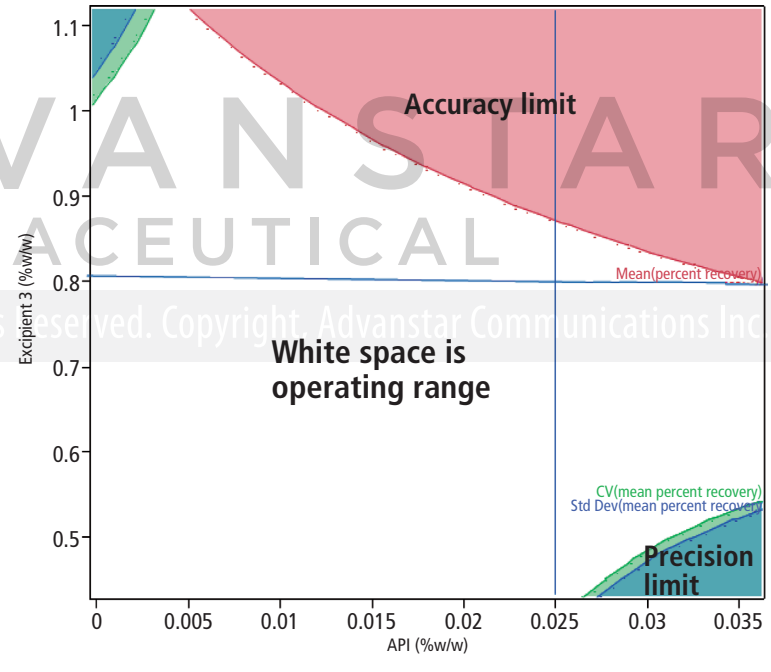


Figure 4: Design Space from the three-factor case study, API percent recovered versus the amount of Excipient 3 in the formulation



Other MVbD outcomes

Control strategy. Figure 4 shows there is less precision (i.e., higher standard deviation and confidence values) at the higher API levels for lower amounts of Excipient 3. This lower precision is approaching the acceptance limit. Similarly, accuracy is approaching the acceptance limit as the excipient and API levels increase. These parameters need to be monitored in case the method needs to be optimized prior to transfer to the final manufacturing site(s).

Continuous improvement. The contour profiler (top graph) in **Figure 5** shows a graph of the balance between precision and bias and its influence on product acceptance/failure rates based on the DOE data. Combinations of method precision and bias must fall within the designated white space to meet the acceptance criteria. The equation for how precision and bias influence product acceptance rates is as follows:

$$1-\text{Normal Distribution}\left[\frac{[LSL-|Bias+\text{Nominal}|]}{\text{Precision}}\right]-\left[1-\text{Normal Distribution}\left[\frac{[USL-|Bias+\text{Nominal}|]}{\text{Precision}}\right]\right]$$

LSL = lower acceptance criteria, USL = upper acceptance criteria, bias = mean percent recovery - 100%, nominal = 100%, and precision = mean standard deviation.

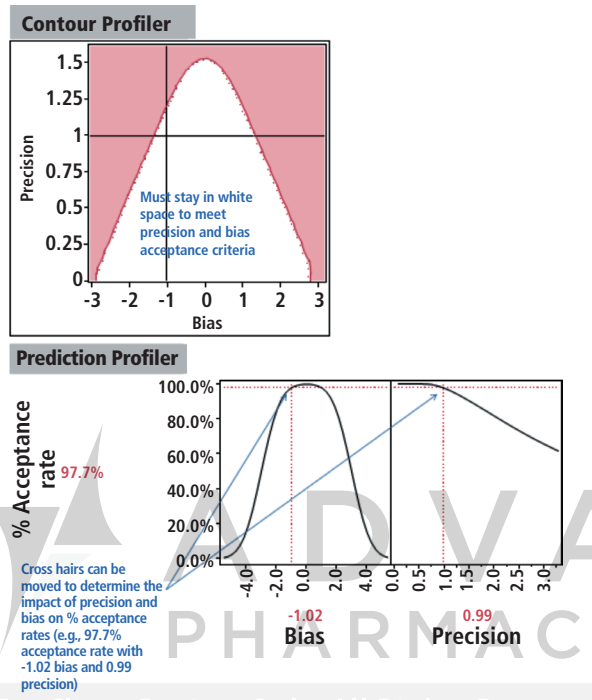
The prediction profiler (bottom graphs) in **Figure 5** allows the precision and bias to be varied to determine the impact on % acceptance rates. This tool can be used to justify moving to new technologies and new specifications by showing that the acceptance criteria will still be met.

Knowledge management. The lower recovery of API observed at high levels of excipient concentrations adds to the formulator's knowledge base. If higher excipient concentrations are needed in future formulations, this aspect needs to be considered.

Regulatory strategy and potential hurdles

Since the MVbD approach is not specified in ICH regulations, not

Figure 5: The top contour profile is a plot of precision (mean standard deviation) versus bias (mean percent recovery – 100%). Any combination of precision and bias that falls in the white space will give acceptable results. The bottom prediction profiler is a tool where bias and precision can be varied by moving the cross hairs to determine the impact on % acceptance rate.



all regulatory agencies may accept this approach. Some filing strategies are briefly discussed.

- **US:** The MVbD approach can be presented in an investigational new drug (IND) application and discussed at the end of Phase II chemistry, manufacturing, and controls (CMC) meeting. The details can be present in a MVbD design protocol along with a rationale for moving within the design space without revalidation. Moving outside the design space could be justified through the use of comparability protocol.
- **EMA and the rest of the world:** Submission of the MVbD rationale, justification, and protocol early in the development process is warranted since a face-to-face conversation may not be possible. Alternatively, this information can be supplied when responding to any agency questions.

Conclusions

The MVbD approach is statistically rigorous and scientifically defensible. It is in line with current regulatory thinking and allows movement to new formulations within the design space without revalidation. The MVbD approach provides a better understanding of the critical parameters of a method and allows greater flexibility and speed during formulation development, especially when time and resource are under constraints. Through judicious planning, MVbD can encompass early formulation design efforts so that a wide range of formulations can be used to define the method-validation design space. Once the ATP has been defined, movement to new analytical technologies and formulations is justified as long as the ATP criteria are met. This approach enables continuous improvement for efficiency and quality gains.

Control strategy, knowledge management, and measurement uncertainty are other key MVbD outputs. The DOE identifies critical validation parameters to monitor and control. The DOE also adds to the knowledge base that will help accelerate future programs. Knowing the method contribution to overall process variability enables setting appropriate in-process controls and product specifications.

Internal company alignment is needed to support a MVbD approach and define a global filing strategy. Early discussions and a presentation of a MVbD protocol to regulatory agencies can help avoid questions during regulatory submission review.

Additional studies can be done to expand the design space but not all changes may require additional studies, for example:

- Changing a grade or source of excipient (e.g., from one grade of lactose to another)
- Using a similar excipient
- Different drug substance process

Some of these and other changes may be justified based on the degree of interactions seen from the DOE data. For example, a source change in lactose may be justified if there are no interactions seen at higher lactose concentrations and historical data on other formulations has shown no impact.

References

1. ICH, Q2 (R1) *Validation of Analytical Procedures: Text and Methodology,* 2005.
2. ICH, Q8 (R2) *Pharmaceutical Development,* 2009.
3. ICH, Q9 *Quality Risk Management,* 2005.
4. ICH, Q10 *Quality System,* 2008.
5. ICH, Q11 *Development and Manufacture of Drug Substances,* 2012.
6. FDA, *Process Validation: General Principles and Practices*, (Rockville, MD, Jan. 2011).
7. P. Nethercote et al., *Pharm. Manufact.* 9 (10) 37-47 (2010).
8. P. Borman et al., *Pharm. Tech.* 31 (10) 142-152 (2007).
9. M. Schweitzer et al, *Pharm. Tech.* 34 (2) 52-59 (2010).
10. M. S. Alasandro, "Method Validation at Pre- and Post-Approval Stages Utilizing QbD Approaches," presentation at the AAPS Stability Workshop (Washington, DC, 2011).
11. J. Fleitman, "Method Validation by Design to Support Formulation Development," presentation at the Drug Development Formulation Summit (San Diego, CA, 2012).
12. B. Harrington, "Applications of QbD Principles to Analytical Methods (AQbD)," presentation at the AAPS Annual Meeting (Washington, DC, 2011).
13. T. Graul, "Applying Quality by Design Principles to the Development of Analytical Methods," presentation at the AAPS Annual Meeting (Washington, DC, 2011).
14. C. Moore, "An FDA Perspective on Drug Development and the Global Regulatory Landscape," presentation at the AAPS Annual Meeting (Washington, DC, 2011).
15. M. Nasr, "Pharmaceutical Quality Assessment - A Science and Risk-based CMC Approach in the 21st Century," presentation at the AAPS Workshop (Washington, DC, 2005). **PT**