REVISION
https://doi.org/10.22239/2317-269X.02162

Definitions and scope of Analytical Quality by Design (AQbD) key elements for method development in the pharmaceutical industry

Definições e escopo dos elementos-chave do *Analytical Quality by Design* (AQbD) para o desenvolvimento de métodos na indústria farmacêutica

Rosemberg Bernardez Moure[I]

Flávia Furtado de Mendonça de Sousa[I]

Diogo Dibo do Nascimento[I]

Jorge Lima de Magalhães[II]

Camila Areias de Oliveira[I]

Livia Deris Prado[I,*]

[I] Laboratório de Desenvolvimento e Validação Analítica, Instituto de Tecnologia em Fármacos, Farmanguinhos, Fundação Oswaldo Cruz (Fiocruz), Rio de Janeiro, RJ, Brasil

[II] Núcleo de Inovação Tecnológica, Instituto de Tecnologia em Fármacos, Farmanguinhos, Fundação Oswaldo Cruz (Fiocruz), Rio de Janeiro, RJ, Brasil

* **E-mail:** liviaderis@gmail.com

Received: Mar 21, 2023
Approved: Jul 31, 2023

How to cite: Moure RB, Sousa FFM, Nascimento DD, Magalhães JL, Oliveira CA, Prado LD. Definitions and scope of Analytical Quality by Design (AQbD) key elements for method development in the pharmaceutical industry. Vigil Sanit Debate, Rio de Janeiro, 2023, v.11: e02162. https://doi.org/10.22239/2317-269X.02162

ABSTRACT

Introduction: The pharmaceutical sector is constantly evolving and is highly regulated with rules to ensure efficacy, safety and quality of medicines. Quality by Design (QbD) is a systematic approach to pharmaceutical development, grounded in scientific knowledge and risk management associated with the manufacturing process. Applied to the development of analytical procedures, QbD has been called Analytical Quality by Design (AQbD) and becomes a process for outlining more robust procedures, applicable throughout the life cycle of the product with a reduction in the incidence of out of trend results or out of specification, related to the method. **Objective:** Clarify the concepts of AQbD and the scope of its key elements for alignment within the pharmaceutical industry. **Method:** Search in databases of scientific articles, as well as national and international guides on the subject. **Results:** AQbD elements include: (1) analytical target profile; (2) identification of critical attributes and critical parameters of the analytical procedure; (3) development, optimization and understanding of the analytical procedure; (4) robustness and definition of the method operable design region; (5) control strategy that includes specifications as well as necessary controls. AQbD values prior knowledge, applies risk assessment and experiment planning during the design of the analytical procedure. **Conclusions:** As the pharmaceutical industry moves towards the implementation of AQbD, a common terminology, understanding of concepts and expectations are needed, which will facilitate better communication between those involved in drug development, including regulatory agencies.

KEYWORDS: Analytical Quality by Design; Analytical Procedure Life Cycle; Analytical Development

RESUMO

Introdução: O setor farmacêutico está em constante evolução e é altamente regulado com normas para garantir eficácia, segurança e qualidade dos medicamentos. O *Quality by Design* (QbD) é uma abordagem sistemática para o desenvolvimento farmacêutico, fundamentada no conhecimento científico e no gerenciamento do risco associado ao processo de fabricação. Aplicado ao desenvolvimento de procedimentos analíticos, o QbD vem sendo denominado de *Analytical Quality by Design* (AQbD) e torna-se um processo de delineamento de procedimentos mais robustos, aplicáveis ao longo do ciclo de vida do produto com redução da incidência de resultados fora da tendência ou fora de especificação relacionados ao método. **Objetivo:** Esclarecer os conceitos de AQbD e o escopo de seus elementos-chave para alinhamento dentro da indústria farmacêutica. **Método:** Busca em bases de dados de artigos científicos, além de guias e diretrizes nacionais e internacionais acerca do tema. **Resultados:** Os elementos AQbD incluem: (1) perfil analítico alvo; (2) identificação dos atributos e parâmetros críticos do

procedimento analítico; (3) desenvolvimento, otimização e compreensão do procedimento analítico; (4) robustez e definição da região de concepção operacional do método; (5) estratégia de controle que inclua especificações, bem como controles necessários. O AQbD valoriza o conhecimento prévio, aplica avaliação de risco e planejamento de experimentos durante o delineamento do procedimento analítico. **Conclusões:** À medida que a indústria farmacêutica avança em direção à implementação do AQbD, uma terminologia comum, compreensão de conceitos e expectativas são necessárias, o que facilitará uma melhor comunicação entre os envolvidos no desenvolvimento de medicamentos, incluindo as agências regulatórias.

PALAVRAS-CHAVE: *Analytical Quality by Design*; Ciclo de Vida do Procedimento Analítico; Desenvolvimento Analítico

INTRODUCTION

The production of medicines includes various pharmaceutical processes, characterized by a sequence of unit operations that transform raw materials in order to generate products with efficacy, safety, and quality[1]. These pharmaceutical processes, established during the development of medicines, must be approved by regulatory bodies in order to be registered, which makes the pharmaceutical industry one of the most regulated. Registration ensures that the company has proven it can consistently supply quality products and protects the health and well-being of the population[2].

Traditionally, pharmaceutical product quality has been guaranteed by tests carried out on the final product, with limited understanding of the process and critical process parameters. As a result, regulatory bodies are focusing on implementing *Quality by Design* (QbD) as a scientific and systematic approach that improves process understanding, reducing variation, and enabling control strategies[3]. The concept of QbD was outlined by Joseph Moses Juran, who believed that most problems concerning a product are related to the way its quality was planned in the first place[4]. In recent years, QbD has been increasingly applied by the pharmaceutical industry, following guidelines from the *International Conference on Harmonization* (ICH)[5,6,7].

As part of drug development, analytical procedures are critical elements, as they are used in the pharmaceutical industries to conduct research and development and to control manufacturing inputs and outputs. These procedures must continuously provide quality data to support decisions while managing risk and uncertainty[8]. Analytical procedures that are not suited to the product can lead to inaccurate results, generating incorrect information that can jeopardize the quality of medicines and the health of the population and generate rework and unnecessary costs for the company. According to the *Food and Drug Administration* (FDA), analytical procedures play an essential role in the QbD philosophy, since implementing QbD requires a high degree of robustness, product quality, and understanding of the analytical procedure[9].

Several researchers have reported that there are similar opportunities for applying QbD to analytical methods as there are for manufacturing processes[10,11,12]. As a result, QbD in the development of *analytical* procedures has been adopted and referred to as *Analytical Quality by Design* (AQbD) to provide a systematic process for obtaining robust and applicable procedures in the product life cycle[13,14,15,16,17]. AQbD is rooted in the ICH guidelines Q86 and Q97, which have been translated into the analytical space through articles[15,16,17,18,19], as well as the U.S. Pharmacopeia (USP) proposition documents chapter <1220>[20] and the ICH guide Q14[21].

AQbD incorporates considerations of scientific and regulatory knowledge, as well as quality control needs, in order to achieve regulatory flexibility and a high degree of robustness and to reduce out-of-trend and out-of-specification results. AQbD exploits scientific understanding in the implementation sequences of the procedure and starts from the identification of the *critical* quality attributes (CQA) of the product[17].

Despite progress in recent years, the interpretation of AQbD concepts and the scope of its key elements is still an ongoing process and will require further clarification and alignment within the international and national pharmaceutical industry. Most applications of AQbD focus on the use of *design of experiments* (DoE) without encompassing other essential elements of this approach. In addition, many articles are conflicting with regard to knowledge about the target analytical profile, method performance characteristics, risk assessment, choice of DoE tool, and obtaining the method's operational design region. This reflects inadequate knowledge and use of AQbD terms. Thus, much effort is still needed to perfect AQbD procedures and to push the concept forward for all analytical procedures that attest to the quality of a drug. This review was proposed with the aim of clarifying the concepts of AQbD and the scope of its key elements for alignment within the pharmaceutical industry.

METHOD

An exploratory descriptive study of the narrative literature review type was carried out, which makes it possible to update knowledge and identify gaps to be explored on a subject in a non-systematized way. The study consisted of the stages of searching and analyzing the literature and the personal interpretation and analysis of the researchers involved[22].

The guiding question was: what are the definitions and key stages of AQbD for implementing the approach in the pharmaceutical industry? Electronic searches were conducted between January 10, 2022, and February 28, 2023, in the *Web*

of Science, Scopus, PubMed, *ScienceDirect*, and *Google Scholar* databases. The searches were carried out using a combination of the following terms: "*Quality by Design*", "*Analytical Quality by Design*", "*fármaco*" *(drug)*, "medicamento" (drug product), "método analítico" (analytical method), "procedimento analítico" (analytical procedure). The terms were used in Portuguese and English.

Scientific articles considered relevant to the topic were selected. The inclusion criteria were: being available electronically, addressing the subject under study, in Portuguese and English and without delimiting the period of publication or the source of information. Articles and technical publications on subjects of no interest to the review were excluded, i.e., those that did not cover AQbD for analytical procedures in the pharmaceutical industry.

This review also used books on the subject of pharmaceutical product development, as well as national and international guides and guidelines.

RESULTS AND DISCUSSION

Regulatory aspects of AQbD

The management of the drug's life cycle is set out in guide Q10 of the ICH[5], which deals with the pharmaceutical quality system. This quality management model requires a harmonized combination of the concepts described in other ICH guides, such as Q86 and Q97, as well as those described in the good manufacturing practices (GMP) legislation of the Brazilian National Health Surveillance Agency (Anvisa)[23]. Implementing the guidelines of the ICH Q10 guide throughout the product life cycle promotes innovation and continuous improvement in pharmaceutical development[5]. The stages of a drug's life cycle begin with pharmaceutical development, which encompasses pharmacotechnical development, process development, analytical procedures and scale-up, through to technology transfer, industrial-scale manufacturing and product discontinuation[24].

With reference to the pharmaceutical quality system, analytical procedures are a fundamental part of the control strategy[15]. Although GMP regulations have been in force for some years, the significant number of quality control-related warnings issued by the FDA has shown that companies have difficulties with risk management in analytical procedures[15]. Do Carmo et al.[25] reported that aspects related to quality control were the main causes of rejection of generic and similar drug registrations by Anvisa. The problems involved a lack of selectivity, linearity, and precision in dissolution tests and methods for detecting impurities. This highlights the need to develop more adequate analytical procedures in terms of performance characteristics or validation.

The principles established in the ICH Q2 guide[26] and in Anvisa's Collegiate Board Resolution (RDC) No. 166 of July 24, 2017[27] govern the validation of the analytical procedure. The revision of the ICH Q2 (R2) guideline[26] made it possible to expand the concepts of validation beyond a single controlled study. This guideline is closely linked to the ICH Q14 concept paper on the development of analytical procedures[21], since it is not only validation data but also information on the development of the procedure that can demonstrate its suitability for the intended purpose.

According to the forthcoming ICH Q14 guideline, analytical procedure development should emphasize understanding and controlling method parameters to obtain the desired results according to specifications and intended use. In addition, it is described that the systematic approach of ICH Q8 and Q9 can be applied to the lifecycle management of analytical procedures[21]. The new ICH guidelines highlight the importance of a comprehensive strategy in the development of analytical procedures, involving a multivariate approach and risk assessment and, although the term AQbD is not used, the researchers refer to the approach described in ICH Q14[21] as AQbD, covering the same steps and using the same tools[16,17].

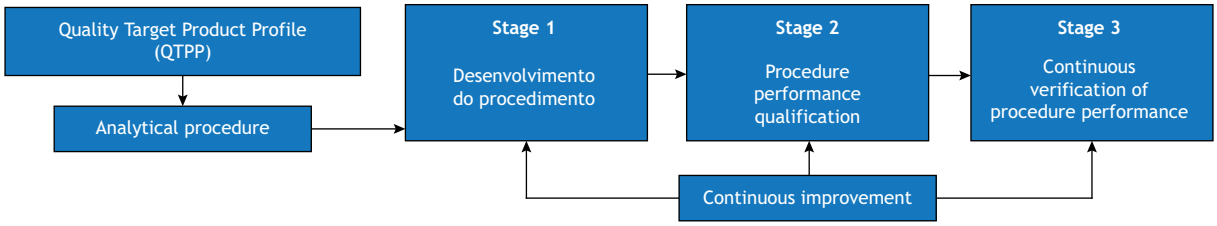
Similarly and recently, experts from the *United States Pharmacopeia* (USP) proposed a new general chapter <1220> "Analytical Procedure Life Cycle"[20], in which the term AQbD is not used, but a comprehensive view of the method and its risk management is integrated to ensure valid data and quality improvement of the procedure at all stages of its life cycle. The principles described in USP <1220>[20] are an AQbD approach to the development, validation and continuous monitoring of analytical procedures. The procedure life cycle advocated by USP <1220> consists of three stages: (1) analytical procedure development; (2) analytical procedure performance qualification (procedure validation); and (3) analytical procedure performance verification (Figure 1). In the proposed general chapter, there is greater emphasis on the initial phases of the life cycle of an analytical procedure, such as defining the procedure specification in a target analytical profile.

The biggest difference between the ICH Q2 (R2)[26] and Q14[21] guides and the USP <1220>[20] chapter is that the ICH guides do not consider the continuous monitoring of the analytical procedure in any specific way (Table 1). In addition, ICH Q14[21] focuses on the operational steps of method development, as described in stage 1 in the USP <1220>[20] chapter.

Although it is not currently considered obligatory to adopt AQbD when developing a procedure, it is likely that regulatory agencies will soon encourage the adoption of AQbD principles for new submissions, given the movement of bodies to draw up documents on the subject. This enhances the role of the analytical area in the product development cycle to measure critical product quality attributes during development, process, control, and in the continuous verification of the process and to monitor trends in product quality.

Essential definitions for implementing AQbD

Attribute of the analytical procedure: a specific property that must be within an appropriate limit, range or distribution to guarantee the desired quality of the measured result. For example:



Source: Prepared by the authors, 2023.
QTPP: Quality Target Product Profile.

Figure 1. Stages of the analytical procedure life cycle inserted in the context of Quality by Design.

Table 1. Comparison of regulatory and compendial documents on the life cycle of the analytical procedure.

Phase of the procedure's life cycle	ICH	USP[20]
Development	ICH Q14[21]	<1220> Stage 1
Validation	ICH Q2 (R2)[26]	<1220> Stage 2
Monitoring	-	<1220> Stage 3

Source: Prepared by the authors, 2023.
ICH: *International Conference on Harmonization*; USP: *United States Pharmacopeia*.

attributes for chromatography measurements can include peak symmetry and resolution[21].

Critical quality attributes or CQA: physical, chemical, or microbiological properties or characteristics that must be within appropriate limits, ranges, or distributions to guarantee the desirable quality of the product[6].

Risk assessment: systematic process of organizing information to support a risk decision to be made within a risk management process. It consists of identifying hazards and analyzing and evaluating the risks associated with exposure to these hazards[7]. Risk assessment tools can be used to identify the parameters of the analytical procedure (factors and operational steps) with a potential impact on its performance and to identify and prioritize the analytical parameters to be investigated experimentally[21].

Performance characteristic: characteristic to guarantee the quality of the measured result, such as accuracy, precision, selectivity, and range. These are characteristics that, in an analytical development without the systematic AQbD approach, are called validation characteristics[26].

Performance criterion: acceptance criterion that describes a numerical range, limit, or desired state to guarantee the quality of the measured result[21].

Total analytical error (TAE): represents the overall error in a result that is attributed to imprecision and inaccuracy. It is the combination of systematic procedural error and random measurement error[21].

Analytical procedure control strategy: planned set of controls derived from the understanding of the analytical procedure that guarantees its performance and the quality of the measured result[21].

Proven acceptable *range for analytical procedures* (PAR): characterized range of an analytical procedure parameter for which operation within that range, other parameters remaining constant, will result in the analytical measurement meeting the performance criteria[21].

Continuous monitoring: collecting and evaluating the performance data of the analytical procedure to ensure the quality of the measured results throughout the life cycle of the analytical procedure[21].

Analytical procedure parameter: any factor (including reagent quality) or operational step of the analytical procedure that can vary continuously (e.g., flow rate) or be specified at single, controllable levels[21].

Quality target product profile (QTPP): a prospective summary of the qualitative characteristics of a product that should ideally be achieved to guarantee desirable quality, considering the safety and efficacy of the product[6].

Analytical target profile (ATP): prospective summary of performance characteristics describing the intended purpose and anticipated performance criteria of an analytical measurement[21].

Design of experiments (DoE): a structured and organized method for determining the relationship between the factors affecting a process and the output of that process[6].

Analytical quality by design (AQbD): systematic approach to analytical development that starts with pre-defined objectives and emphasizes the understanding of the analytical procedure and procedure control, based on sound scientific data and quality risk management.

Quality by design (QbD): a systematic approach to development that starts with pre-defined objectives and emphasizes product and process understanding and process control, based on sound scientific data and quality risk management[6].

Method operable design region (MODR): combination of analytical procedure parameter ranges within which the performance criteria of the analytical procedure are met and the quality of the measured result is guaranteed[21].

Robustness: a measure of the analytical procedure's ability to meet expected performance requirements during normal use. Robustness is tested by small, deliberate variations in the parameters of the analytical procedure[21]. This evaluation allows the determination of robust operating regions for procedure parameters[20,21].

System suitability test (SST): tests developed and used to verify that the measurement system and the analytical operations associated with the analytical procedure are suitable for the intended analysis and increase the detectability of possible faults[21].

Flow of analytical procedure development following the AQbD approach

The implementation of AQbD is a parallel process to QbD for product development. From the construction of the QTPP, which encompasses aspects of pharmaceutical development as a whole, the AQCs are defined, which can include: content, impurities, dissolution, among others. Once the CQAs have been defined, it is necessary to determine control strategies so that they remain within the specified limits. With this, analytical procedures must be developed for application throughout the life cycle. The stages in the development of the analytical procedure using the AQbD approach are shown in Figure 2 and consist of the steps described in ICH Q14[21] and stage 1 of USP <1220>[20].

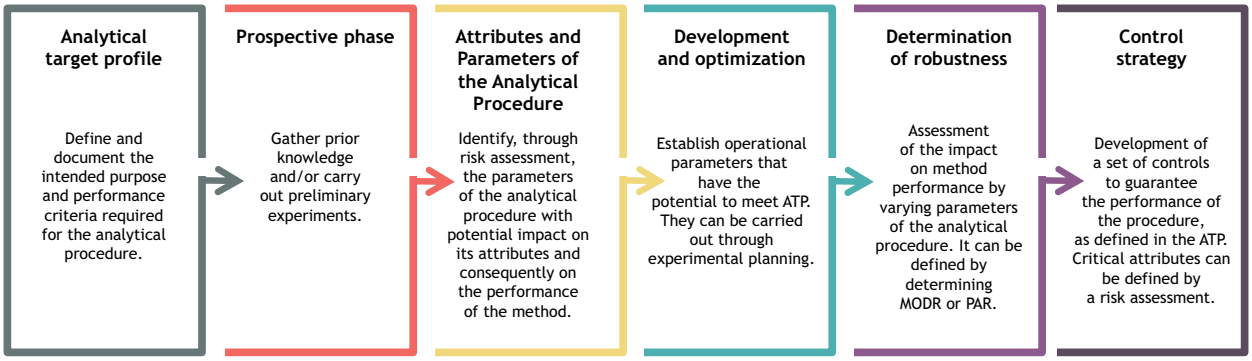
The first stage of AQbD is to define the ATP requirements. Once these have been defined, an analytical technique capable of meeting the ATP requirements must be selected and initial tests can be carried out to verify the technique's suitability[28]. Thus, a prospective phase begins in order to gather prior knowledge about the procedure[12,29]. The next step is to select the attributes of the analytical procedure, which are the responses measured to control the procedure's performance. The parameters of the analytical procedure are also defined, which are factors or operational steps that can impact on the attributes of the procedure. This is followed by the method development and optimization phase, in which univariate or multivariate experiments can be carried out. The robustness of the procedure is then determined and an initial control strategy is defined[21,30].

Analytical target profile

A well-defined ATP is fundamental to the successful application of AQbD, as it ensures that the procedure developed is fit for the purpose, provides the criteria for validation of the procedure and a mechanism for flexibility of the method within the control strategy during its lifecycle. The ATP defines the purpose of the test and the quality requirements for the reportable result (usually associated with a CQA), aligned with the QTPP, and is linked to the attribute to be tested and not to a specific analytical procedure[18]. In other words, the ATP focuses the goals of the development of an analytical procedure, guides the choice of analytical technology, serves as the basis for the procedure's performance qualification criteria and provides a guide for continuous monitoring during its life cycle[31].

The ATP can be defined in various ways, but the general focus of having a procedure with acceptable accuracy and precision should be part of the ATP. In addition, other performance characteristics can be established, such as limit of detection, limit of quantification and selectivity. The robustness of the procedure technically can also be delimited in the ATP, but it is usually more appropriate for it to be derived from the development stage, with the construction of the MODR's operational design region. The performance criteria (acceptance criteria) should also be described and determined based on a number of factors, including: the criticality of the CQA being measured; the risk that an error may occur; the acceptance range of the specification for the CQA; the potential clinical impact on safety or efficacy (if known) that an analytical error may have[17].

The definition of the TAE can be an alternative approach to the individual assessment of accuracy and precision described in the ATP[21]. Table 2 presents a hypothetical example of an ATP, where the objective of the procedure is the quantification of



Source: Prepared by the authors, 2023.
MODR: Method Operable Design Region; PAR: Proven Acceptable Range.

Figure 2. Analytical procedure development flow following the Analytical Quality by Design (AQbD) approach.

an active pharmaceutical ingredient (API) in a drug, using the technique of high-performance liquid chromatography. The ATP in this example captures the performance expectations that are driven by the product, such as selectivity and the TAE. Once the TAE has been defined, accuracy and precision can also be included in the ATP, depending on the specific needs of the test.

Changes to analytical procedures can occur throughout the product's life cycle and may involve modifying existing procedures or replacing them completely. Important changes in performance characteristics or additional information on attributes can, in certain cases, lead to a reassessment of the ATP[15]. ATP can be applied prospectively to new procedures and retrospectively to existing procedures[17].

Prospective phase

Once the ATP has been drawn up, relevant information must be collected before development activities begin, such as: chemical structures and their properties, reference chemicals, reagents, instrumentation and any other linked to operational requirements. In other words, in the prospective phase, prior knowledge must be gathered to assist in the development activities of the analytical procedure. This prior knowledge can be internal company knowledge and analytical experience and/or external knowledge, such as scientific and technical references or established scientific principles[19].

Once the analytical technology has been selected, preliminary experiments can be carried out to initially select parameters that bring the procedure closer to the ATP. In the prospective phase, in separation procedures, for example, the stationary phases, mobile phases and elution mode, as well as the sample preparation, are usually quickly evaluated or selected[32]. The prospective phase deserves attention because it deals with the general conditions of analysis, which will be extensively investigated and optimized[33].

Analytical procedure attributes and Analytical procedure parameters

The analytical procedure attributes must be identified prior to the definition of the analytical procedure parameters and after the preparation of the ATP, selection of the analytical technology and the prospective phase. The attributes of the analytical procedure must be within limits, ranges, or distributions to guarantee the desired quality of the measured result[21]. Attributes for chromatography measurements, for example, can include resolution, peak symmetry factor, and number of theoretical plates. The limits of the procedure's attributes are usually defined through prior knowledge, compendia, literature and/or experiments carried out in the prospective phase. In fact, these attributes are represented by key responses directly correlated to a mathematical representation of the quality of the method's performance and, therefore, to the quality of the analytical data[32]. In the literature, there are different terms for an attribute of the analytical procedure, as described in ICH document Q14[21], such as: critical *method* attributes (CMA)[28,34,35] and CQA[36,37], the latter being confused with CQA as an element of the QbD for product development and not the analytical procedure.

The analytical procedure parameters should be identified by risk assessment tools supported by prior knowledge, compendia, literature and/or initial experiments. During the risk assessment, the parameters of the procedure are screened and investigated to identify those that could potentially affect one or more attributes of the method[19]. Among the main risk assessment tools used are flowcharts, the Ishikawa diagram, cause and effect matrix and others. The USP, in the proposed chapter <1220> suggests the use of *heat* maps to support qualitative risk assessment, as it provides a visual indication of which procedure parameters are considered to have a potentially high, medium, or low impact on procedure performance[20]. In the literature, there are different terms for analytical procedure parameters, a term described in ICH document Q14[21] as critical *method* parameters (CMP)[28,34,35].

Table 2. Example of a analytical target profile (ATP) for determining the content of an active pharmaceutical ingredient (API) in a drug.

ATP		
Quantify the API content to guarantee the quality of the product batch by batch, as well as assessing stability.		
Correlation with the CQA		**Justification**
The procedure must be able to measure the API content from 90% to 110%		Effectiveness of the drug product
Method performance	**Target**	**Justification**
Accuracy Precision	Standard uncertainty = 1.25%	To ensure that the analytical variation around the estimated mean is within the widest specification range
Selectivity	No interference from impurities or matrix components in the API peak	Ensure that the API is dosed unequivocally, with adequate selectivity (minimum resolution of 1.5) in the presence of impurities and matrix components.
Reportable range	90.0% to 110.0% of declared	Range established in the product monograph in an official compendium

Source: Prepared by the authors, 2023.
ATP: Analytical target profile; API: Active pharmaceutical ingredient; CQA: *Critical quality attributes*.

Figure 3 shows a hypothetical Ishikawa diagram with the parameters of the high-performance liquid chromatography analytical procedure for determining the content of an API in the presence of its impurities. Initially, the parameters with a potential effect on the selectivity of the method were defined, and consequently on the resolutions between the peaks (an attribute of the procedure). With the Ishikawa diagram established, it is possible to identify the parameters with the greatest potential to impact on the attributes of the procedure using an integrated approach, whether it be *brainstorming* or a cause and effect matrix, for example. These should be extensively investigated in the next stage, especially using DoE tools[32].

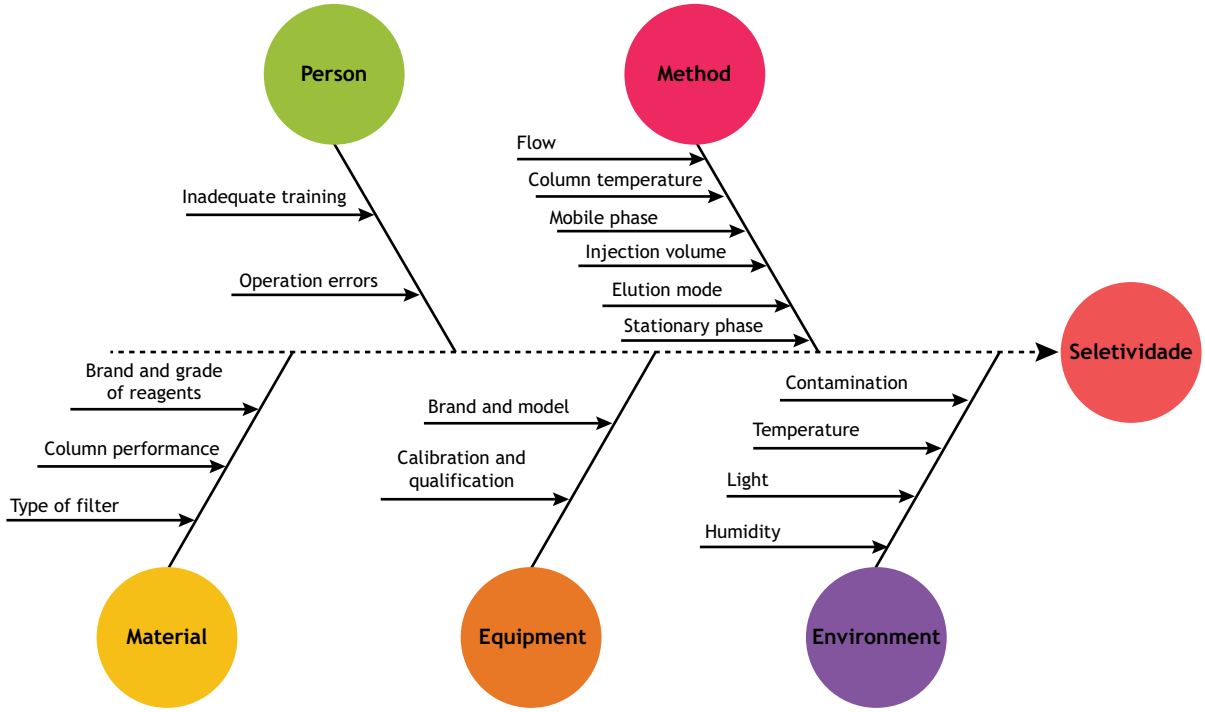
Development and optimization of the analytical procedure

The main objectives of developing analytical procedures are to identify conditions that minimize bias, optimize variability, and establish robust operating parameters to meet the ATP[20]. To this end, the relevant analytical procedure parameters can be investigated in univariate or multivariate experiments using DoE[21]. To better understand the influence of the analytical procedure parameters (inputs or factors) on the analytical procedure attributes (outputs or responses) and their impact (directly or indirectly) on the ATP, the use of DoE tools is recommended.

DoE is a systematic approach that integrates multifactorial experimentation, mitigation of the impact of variability and response modeling to maximize the information obtained[38]. The application of DoE is justified due to the many variables (parameters) that affect the results (attributes) of the method. In addition, adopting an appropriate DoE protocol allows for the identification of MODR and, consequently, a high degree of understanding of the analytical procedure[21]. The procedure parameters considered as variables in the DoE should be selected based on the risk assessment in the previous step.

The decision on the selection of the DoE tool should be made based on the number of variables, knowledge of the parameters and scientific understanding between result and variable[39]. For example: if the effect of all input variables and their interactions are to be measured, DoE can be applied and then considered and optimized with the response surface methodology. When many variables are studied without the need to evaluate interaction effects, the Plackett-Burman method can be used[15].

Different strategies for analytical development using DoE can be used. In the literature, DoE is applied in two phases of development, called screening and optimization[15,28,32]. In these cases, screening DoE is useful for studying the effects of qualitative and/or quantitative parameters on the attributes of the procedure and with a low number of experiments. Screening experiments make it possible to identify parameters that have no influence on the attributes of the procedure and to set their values, as well as to identify parameters for which the results indicate the optimum values, which can also be set. Another advantage is the possibility of moving the experimental domain to the optimization DoE, leading to the best results. The Plackett-Burman design and fractional factorial designs are the most commonly used in screening[32]. Moreira



Source: Prepared by the authors, 2023.

Figure 3. Example of Ishikawa diagram for risk assessment to define analytical procedure parameters with potential impact on selectivity.

and Lourenço applied the Plackett-Burman matrix in screening experiments for the chromatographic separation of verapamil hydrochloride and its impurities with 13 analytical variables (type of buffer in the mobile phase, concentration of the buffer, pH of the buffer, type of organic solvent in the mobile phase, concentration of ammonium hydroxide in the mobile phase, type of C18 column, column temperature, mobile phase flow rate, injection volume, elution time of the first gradient, elution time of the second gradient, proportion of organic solvent in the mobile phase during gradient elution and isocratic elution time). According to the effects observed in the responses of interest, the factors pH of the buffer, concentration of ammonium hydroxide and injection volume were selected for the optimization phase[40].

It should be noted that the screening phase is not mandatory and can be avoided on the basis of preliminary knowledge and/or univariate experiments, provided that the information available allows for rational planning of the subsequent optimization.

The optimization phase generally consists of applying the response surface methodology to estimate the main interaction and/or quadratic effects of the parameters on the attributes of the analytical procedure. In this case, at least three levels can be studied for each parameter in order to assess the presence of model curvature, making it possible to obtain a predictive regression model and thus draw a map of the values of the procedure's attributes predicted throughout the experimental domain (response surface or contour plot)[17,32]. For the chromatographic separation of verapamil hydrochloride and its impurities, the central composite design was selected to adjust a regression model that explained the chromatographic response as a function of the analytical conditions used as factors (buffer pH, ammonium hydroxide concentration and injection volume). The regression models made it possible to study the response surface, evaluate the interaction between the factors and construct the MODR[40].

Some articles also use the strategy of using DoE for robustness, after the optimization DoE. In this case, a large number of factors are evaluated and, normally, no effect is expected during the tests. However, it should be noted that for robustness evaluation, the robustness DoE step may not be necessary, since a MODR can be defined as part of the optimization in development that provides results that meet the ATP requirements. This approach automatically creates robustness in the procedure by defining the MODR associated with the procedure's parameter ranges[17].

Robustness

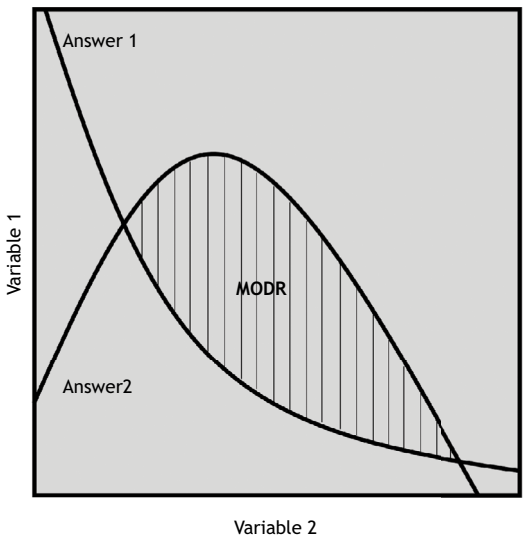
National and international regulatory agencies have recognized that the robustness of the analytical procedure must be demonstrated during the development of the method[26,27]. Thus, by implementing the AQbD approach, when multivariate experiments are conducted using DoE tools that generate mathematical models for predicting the values of the procedure's attributes, the MODR can be constructed. The MODR can be considered as a robustness zone and is the multivariate space of the parameters of the analytical procedure that guarantee that the ATP is met and therefore provides assurance of the quality of the measured value with a specified level of probability[21]. Figure 4 shows an example of a MODR.

When defining the MODR, aspects such as the uncertainty of the model's parameters, as well as the probability of meeting the specifications of the procedure's attributes, must be taken into account. Monte-Carlo simulations are useful tools for this task[41]. Furthermore, since working within the MODR is not considered a change to the procedure[6], a more flexible approach to the method from a regulatory point of view is obtained.

Two options represent examples of approaches to MODR validation, also allowing for differentiated solutions: (1) a single set of MODR operating parameters is selected (usually the intended operating conditions), which for future changes to the parameters within the MODR, an assessment regarding additional validation activities must be carried out or (2) intended operating conditions and the extremes of the MODR are selected, allowing for total operational flexibility without requiring additional validation[21]. This multi-point verification within the MODR, with joint assessment of accuracy and precision, probably represents the highest probability of the procedure's ability to meet ATP requirements.

Control strategy for the analytical procedure and establishment of analytical procedure conditions

At this stage, the initial version of the analytical procedure's control strategy should be drawn up; this should be defined before validation and confirmed after validation has been completed. The preliminary control strategy is defined during the procedure development process and includes the SST and other environmental or procedural controls necessary for it to



Source: Prepared by the authors, 2023.

Figure 4. Illustrative example of Method Operable Design Region (MODE).

meet the ATP. The attributes of the procedure, identified during development as critical, must be controlled and their conditions, materials or acceptable criteria must be explicitly specified in the procedure. Acceptance criteria should be based on performance criteria of the analytical procedure and the components of the SST should be selected using risk assessment as well as knowledge and understanding of the development data. In addition, the experimental scheme for future parameter movements within the MODR can be predefined in the control strategy[20,21].

Throughout the stages of the AQbD, the knowledge obtained from the analytical procedure should be recorded. It is recommended to compile the information on the analytical procedure with the following information: performance characteristics described in the ATP, acceptance criteria for the attributes of the analytical procedure, the parameters of the analytical procedure and their definitions (working point), control strategy for the analytical procedure, and even the configuration of a validation strategy for the analytical procedure for the performance characteristics.

The working points or conditions established for the method are the definitions for each analytical parameter evaluated. They can be chosen according to various criteria, based on convenience or operational facilities, such as less solvents or additives, lower costs, shorter analysis time, as well as based on statistical criteria, such as greater probability of meeting the ATP requirements, and so on. When using the univariate strategy, points within the PARs can be established for the analytical procedure. When using the multivariate strategy, the specific points within the MODR selected can be defined as operating points before carrying out the validation[33].

In the literature, there is no consensus on how the control strategy should be carried out. There is the use of control charts to monitor the analytical procedure[42], regression models to calculate the risks of the method not meeting specifications in the routine[42] and the definition of SST parameters, along with their limits[43].

CONCLUSIONS

The implementation of AQbD should be an important part of the QbD process, as it supports the development and implementation of methods with a focus on the product quality attributes that must be controlled to ensure the safety and efficacy of the drug. Developing analytical procedures with the AQbD approach results in a broader understanding of the method, which means easier improvements and more flexible regulatory approaches. However, several challenges hinder the full implementation of this approach, such as the interpretation of AQbD concepts and the scope of its key elements. This is because the first guidelines for the pharmaceutical industry have only recently been drawn up and the scientific literature presents studies on the subject, but with differences between the stages of AQbD and, above all, with descriptions of only some of the stages and not all of them. This article therefore describes the steps considered necessary for analytical development based on AQbD, combining scientific knowledge with the most recent guidelines for the pharmaceutical industry.

The AQbD stages include establishing the ATP, identifying the critical attributes and parameters of the analytical procedure, developing and optimizing the analytical procedure, assessing robustness, and defining the MODR and control strategy. In addition, the entire method life cycle must be considered, such as development by AQbD (stage 1), the validation stage (stage 2), and continuous monitoring (stage 3), the latter being implemented after the establishment of an analytical method for quality control. Considering that AQbD has been gaining attention from academia, industry, and regulatory agencies, it is believed that the requirement to apply this approach in the pharmaceutical industry will soon become a reality. Thus, this work also makes it possible to publicize and demonstrate the wider applicability of AQbD for future activities and regulatory standardization for the national pharmaceutical industry.

REFERENCES

1. Yu LX. Pharmaceutical quality by design: product and process development, understanding, and control. Pharm Res. 2008;25:781-91. https://doi.org/10.1007/s11095-007-9511-1
2. Handoo S, Arora V, Khera D, Nandi PK, Sahu SK. A comprehensive study on regulatory requirements for development and filing of generic drugs globally. Int J Pharm Invest. 2012;2(3):99 -105. https://doi.org/10.4103/2230-973X.104392
3. Zhang L, Mao S. Application of quality by design in the current drug development. Asian J Pharm Sci. 2017;12(1):1-8. https://doi.org/10.1016/j.ajps.2016.07.006
4. Juran JM. Juran on quality by design: the new steps for planning quality into goods and services. New York: Simon and Schuster; 1992.
5. European Medicines Agency – EMA. ICH guideline Q10 on pharmaceutical quality system. London: International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use; 2008.
6. European Medicines Agency – EMA. ICH guideline Q8 (R2) on pharmaceutical development. London: International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use; 2009.
7. European Medicines Agency – EMA. ICH guideline Q9 (R1) on quality risk management. London: International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use; 2021.
8. Breaux J, Jones K, Boulas P. Understanding and implementing efficient analytical methods development and validation. Pharm Technol Analyt Chem Test. 2003;(Suppl.1):6-13.

9. Chatterjee S. QbD considerations for analytical methods: FDA perspective. In: US IFPAC annual meeting; Baltimore, United States. Silver Spring: US Food and Drugs Administration; 2013[accesso 13 dez 2022]. Disponível em: https://qbdworks.com/wp-content/uploads/2014/06/AnalyticalQbDIFPAC.pdf
10. Borman P, Nethercote P, Chatfield M, Thompson D, Truman K. The application of quality by design to analytical methods. Pharm Technol. 2007;31(10):142-52.
11. Schweitzer M, Pohl M, Hanna-Brown M, Nethercote P, Borman P, Hansen G et al. Implications and opportunities of applying QbD principles to analytical measurements. Pharm Technol. 2010;34(2):52-9.
12. Vogt FG, Kord AS. Development of quality-by-design analytical methods. J Pharma Sci. 2011;100(3):797-812. https://doi.org/10.1002/jps.22325
13. Reid GL, Cheng G, Fortin DT, Harwood JW, Morgado JE, Wang J et al. Reversed-phase liquid chromatographic method development in an analytical quality by design framework. J Liqu Chromatogr Relat Technol. 2013;36(18):2612-38. https://doi.org/10.1080/10826076.2013.765457
14. Hubert C, Houari S, Rozet E, Lebrun P, Hubert P. Towards a full integration of optimization and validation phases: an analytical-quality-by-design approach. J Chromatogr A. 2015;1395:88-98. https://doi.org/10.1016/j.chroma.2015.03.059
15. Peraman R, Bhadraya K, Padmanabha Reddy Y. Analytical quality by design: a tool for regulatory flexibility and robust analytics. Int J Anal Chem. 2015;2015. https://doi.org/10.1155/2015/868727
16. Prajapati PB, Jayswal K, Shah SA. Application of quality risk assessment and DoE-based enhanced analytical quality by design approach to development of chromatography method for estimation of combined pharmaceutical dosage form of five drugs. J Chromatogr Sci. 2021;59(8):714-29. https://doi.org/10.1093/chromsci/bmaa118
17. Verch T, Campa C, Chéry CC, Frenkel R, Graul T, Jaya N et al. Analytical Quality by Design, life cycle management, and method control. AAPS J. 2022;24(1):34. https://doi.org/10.1208/s12248-022-00685-2
18. Jackson P, Borman P, Campa C, Chatfield M, Godfrey M, Hamilton P et al. Using the analytical target profile to drive the analytical method lifecycle. Anal Chem. 2019;91(4):2577-85. https://doi.org/10.1021/acs.analchem.8b04596
19. Orlandini S, Hancu G, Szabó ZI, Modroiu A, Papp LA, Gotti R et al. New trends in the quality control of enantiomeric drugs: quality by design-compliant development of chiral capillary electrophoresis methods. Molecules. 2022;27(20):1-28. https://doi.org/10.3390/molecules27207058
20. United States Pharmacopeia – USP. 45-the National Formulary 40. Rockville: United States Pharmacopeial Convention; 2022.
21. European Medicines Agency – EMA. ICH guideline Q14 on analytical procedure development. London: International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use; 2022.
22. Botelho LL, Cunha CCA, Macedo M. O método da revisão integrativa nos estudos organizacionais. Gest Soc. 2011;5(11):121-36. https://doi.org/10.21171/ges.v5i11.1220
23. Agência Nacional de Vigilância Sanitária – Anvisa. Resolução RDC Nº 658, de 30 de março de 2022. Dispõe sobre as diretrizes gerais de boas práticas de fabricação de medicamentos. Diário Oficial União. 31 mar 2022.
24. Jagan BG, Mahapatra AK, Murthy NP, Patra RK. Quality by design (QbD): principles, underlying concepts and regulatory prospects. Thai J Pharm Sci. 2021;45(1):54-69.
25. Carmo AC, Piras SS, Rocha NF, Gratieri T. Main reasons for registration application refusal of generic and similar pharmaceutical drug products by the Brazilian Health Regulatory Agency (Anvisa). BioMed Res Int. 2017;2017:1-11. https://doi.org/10.1155/2017/7894937
26. European Medicines Agency – EMA. ICH guideline Q2(R2) on validation of analytical procedures. London: International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use; 2022.
27. Agência Nacional de Vigilância Sanitária – Anvisa. Resolução RDC Nº 166, de 24 de julho de 2017. Dispõe sobre a validação de métodos analíticos e dá outras providências, 2017. Diário Oficial União. 25 jul 2017.
28. Tome T, Zigart N, Casar Z, Obreza A. Development and optimization of liquid chromatography analytical methods by using AQbD principles: overview and recent advances. Org Process Res Dev. 2019;23:1784-802. https://doi.org/10.1021/acs.oprd.9b00238
29. Bousses C, Ferey L, Vedrines E, Gaudin K. Using an innovative combination of quality-by-design and green analytical chemistry approaches for the development of a stability indicating UHPLC method in pharmaceutical products. J Pharm Biomed Anal. 2015;115:114-22. https://doi.org/10.1016/j.jpba.2015.07.003
30. Kovac L, Casar Z, Lusin TT, Roskar R. Development of an analytical method for determination of related substances and degradation products of cabotegravir using analytical quality by design principles. ACS Omega. 2022;7(10):8896-905. https://doi.org/10.1021/acsomega.1c07260
31. Parr MK, Schmidt AH. Life cycle management of analytical methods. J Pharm Biomed Anal. 2018;147:506-17. https://doi.org/10.1016/j.jpba.2017.06.020
32. Deidda R, Orlandini S, Hubert P, Hubert C. Risk-based approach for method development in pharmaceutical quality control context: a critical review. J Pharm Biomed Anal. 2018;161:110-21. https://doi.org/10.1016/j.jpba.2018.07.050
33. Orlandini S, Pinzauti S, Furlanetto S. Application of quality by design to the development of analytical separation methods. Anal Bioanal Chem. 2013;405:443-50. https://doi.org/10.1007/s00216-012-6302-2

34. Otašević B, Šljivić J, Protić A, Maljurić N, Malenović A, Zečević M. Comparison of AQbD and grid point search methodology in the development of micellar HPLC method for the analysis of cilazapril and hydrochlorothiazide dosage form stability. Microchem J. 2019;145:655-63. https://doi.org/10.1016/j.microc.2018.11.033

35. Žigart N, Časar Z. Development of a stability-indicating analytical method for determination of venetoclax using AQbD principles. ACS omega. 2020;5(28):17726-42. https://doi.org/10.1021/acsomega.0c02338

36. Bonde S, Bonde CG, Prabhakar B. Quality by design based development and validation of HPLC method for simultaneous estimation of paclitaxel and vinorelbine tartrate in dual drug loaded liposomes. Microchem J. 2019;149. https://doi.org/10.1016/j.microc.2019.103982

37. Zhang H, Wang J, Chen Y, Shen X, Jiang H, Gong X et al. Establishing the chromatographic fingerprint of traditional Chinese medicine standard decoction based on quality by design approach: a case study of *Licorice*. J Sep Sci. 2019;42(6):1144-54. https://doi.org/10.1002/jssc.201800989

38. Montgomery DC. Design and analysis of experiments. 10a. ed: Hoboken: John Wiley & Sons; 2019.

39. Rodrigues MI, Iemma AF. Experimental design and process optimization. Boca Raton: CRC; 2014.

40. Moreira SC, Lourenco FR. Development and optimization of a stability-indicating chromatographic method for verapamil hydrochloride and its impurities in tablets using an analytical quality by design (AQbD) approach. Microchem J. 2020;154. https://doi.org/10.1016/j.microc.2020.104610

41. Rozet E, Lebrun P, Hubert P, Debrus B, Boulanger B. Design spaces for analytical methods. Trends Anal Chem. 2013;42(24):157-67. https://doi.org/10.1016/j.trac.2012.09.00

42. Deidda R, Avohou HT, Baronti R, Davolio PL, Pasquini B, Del Bubba M et al. Analytical quality by design: Development and control strategy for a LC method to evaluate the cannabinoids content in cannabis olive oil extracts. J Pharm Biomed Anal. 2019;166:326-35. https://doi.org/10.1016/j.jpba.2019.01.032

43. Yao H, Vancoillie J, D'Hondt M, Wynendaele E, Bracke N, De Spiegeleer B. An analytical quality by design (aQbD) approach for a L-asparaginase activity method. J Pharm Biomed Anal. 2016;117:232-9. https://doi.org/10.1016/j.jpba.2015.08.042

Authors' Contribution

Moure RB, Sousa FFM, Nascimento DD, Magalhães JL, Oliveira CA, Prado LD - Conception, planning (study design), acquisition, analysis, data interpretation, and writing of the work. All the authors approved the final version of the work.

Conflict of Interest

The authors inform that there is no potential conflict of interest with peers and institutions, political or financial, in this study.