Establishing quality specifications for medicines, vaccines and in vitro diagnostics

Week of Quality 2023 training kit
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Week of Quality 2023 training kit
Establishing quality specifications for medicines, vaccines and in vitro diagnostics: Week of Quality 2023 training kit

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Photo credits: © Reuters/Johnson & Johnson, Gloved hand holding a vaccine vial.
# Contents

<table>
<thead>
<tr>
<th>Acknowledgements</th>
<th>iv</th>
</tr>
</thead>
<tbody>
<tr>
<td>Abbreviations</td>
<td>v</td>
</tr>
<tr>
<td>Introduction</td>
<td>1</td>
</tr>
</tbody>
</table>

## Setting the standard: achieving quality specification for vaccine manufacturing

<table>
<thead>
<tr>
<th>Session 1</th>
<th>Establishing patient-centric specifications</th>
<th>3</th>
</tr>
</thead>
<tbody>
<tr>
<td>Session 2</td>
<td>Clinical qualification of vaccines acceptable ranges</td>
<td>6</td>
</tr>
<tr>
<td>Session 3</td>
<td>Establishing harmonized specifications for new vaccines: lessons learned from the COVID-19 vaccines</td>
<td>10</td>
</tr>
<tr>
<td>Session 4</td>
<td>Replacement of in vivo potency testing by in vitro assay</td>
<td>13</td>
</tr>
<tr>
<td>Session 5</td>
<td>Addressing the setting of specification in a Common Technical Document application</td>
<td>16</td>
</tr>
</tbody>
</table>

## Ensuring excellence: getting the specification for medicines right

<table>
<thead>
<tr>
<th>Session 6</th>
<th>Active pharmaceutical ingredient monographs</th>
<th>22</th>
</tr>
</thead>
<tbody>
<tr>
<td>Session 7</td>
<td>Control of active pharmaceutical ingredient characteristics</td>
<td>27</td>
</tr>
<tr>
<td>Session 8</td>
<td>Nitrosamines: risk factors and control strategies</td>
<td>31</td>
</tr>
<tr>
<td>Session 9</td>
<td>Dosage forms specific requirements</td>
<td>33</td>
</tr>
<tr>
<td>Session 10</td>
<td>Setting requirements for dissolution: the importance of the biobatch</td>
<td>36</td>
</tr>
</tbody>
</table>

## Bridging the gap between design validation and quality specifications for in vitro diagnostics

<table>
<thead>
<tr>
<th>Session 11</th>
<th>Designing, implementing, and applying design controls</th>
<th>42</th>
</tr>
</thead>
<tbody>
<tr>
<td>Session 12</td>
<td>Design control challenges</td>
<td>43</td>
</tr>
<tr>
<td>Session 13</td>
<td>Design verification and validation</td>
<td>45</td>
</tr>
<tr>
<td>Session 14</td>
<td>Design transfer and changes</td>
<td>50</td>
</tr>
<tr>
<td>Session 15</td>
<td>Understanding performance evaluation of in vitro diagnostics</td>
<td>51</td>
</tr>
</tbody>
</table>

References | 55 |
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### Abbreviations

<table>
<thead>
<tr>
<th>Abbreviation</th>
<th>Description</th>
</tr>
</thead>
<tbody>
<tr>
<td>3Rs</td>
<td>Replacement, Reduction and Refinement</td>
</tr>
<tr>
<td>Ab</td>
<td>Antibody</td>
</tr>
<tr>
<td>ADE</td>
<td>Antibody-dependent enhancement</td>
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<tr>
<td>AI</td>
<td>Acceptable intake</td>
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<tr>
<td>AI</td>
<td>Artificial intelligence</td>
</tr>
<tr>
<td>API</td>
<td>Active pharmaceutical ingredient</td>
</tr>
<tr>
<td>AQPd</td>
<td>Analytical quality by design</td>
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<tr>
<td>BCS</td>
<td>Biopharmaceutics Classification System</td>
</tr>
<tr>
<td>BE</td>
<td>Bioequivalence</td>
</tr>
<tr>
<td>BP</td>
<td>British Pharmacopoeia</td>
</tr>
<tr>
<td>CFR</td>
<td>Code of Federal Regulations</td>
</tr>
<tr>
<td>cGMP</td>
<td>current Good Manufacturing Practices</td>
</tr>
<tr>
<td>CHMP</td>
<td>Committee for Medicinal Products for Human Use</td>
</tr>
<tr>
<td>CMC</td>
<td>Chemistry, Manufacturing, and Controls</td>
</tr>
<tr>
<td>CMI</td>
<td>cell-mediated immunity</td>
</tr>
<tr>
<td>COA</td>
<td>Certificate of Analysis</td>
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<tr>
<td>COVID-19</td>
<td>Coronavirus Disease 2019</td>
</tr>
<tr>
<td>CPCA</td>
<td>Carcinogenic potency categorisation approach</td>
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<tr>
<td>CQAs</td>
<td>Critical quality attributes</td>
</tr>
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<td>CQPs</td>
<td>Critical quality parameters</td>
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<tr>
<td>CT</td>
<td>Clinical trial</td>
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<tr>
<td>CTD</td>
<td>Common Technical Document</td>
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<tr>
<td>DHF</td>
<td>Device History File</td>
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<tr>
<td>DMF</td>
<td>Drug Master File</td>
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<tr>
<td>DNA</td>
<td>Deoxyribonucleic Acid</td>
</tr>
<tr>
<td>DP</td>
<td>Drug product</td>
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<tr>
<td>DQ</td>
<td>Design qualification</td>
</tr>
<tr>
<td>DS</td>
<td>Drug substance</td>
</tr>
<tr>
<td>DSC</td>
<td>Differential scanning calorimetry</td>
</tr>
<tr>
<td>DSV</td>
<td>Dose-solubility volume</td>
</tr>
<tr>
<td>EDQM</td>
<td>European Directorate for the Quality of Medicines and Healthcare</td>
</tr>
<tr>
<td>EEA</td>
<td>European Economic Area</td>
</tr>
<tr>
<td>ELISA</td>
<td>Enzyme-linked immunosorbent assay</td>
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<td>EMA</td>
<td>European Medicines Agency</td>
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<tr>
<td>EPAR</td>
<td>European Public Assessment Report</td>
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<tr>
<td>EQAS</td>
<td>External quality assessment schemes</td>
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<tr>
<td>EU</td>
<td>European Union</td>
</tr>
<tr>
<td>EUL</td>
<td>Emergency Use Listing</td>
</tr>
<tr>
<td>FAVN</td>
<td>Fluorescent antibody virus neutralization</td>
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<tr>
<td>FDA</td>
<td>Food and Drug Administration</td>
</tr>
<tr>
<td>FPP</td>
<td>Finished pharmaceutical product</td>
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<tr>
<td>GMP</td>
<td>Good Manufacturing Practice</td>
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<tr>
<td>GP</td>
<td>Glycoprotein</td>
</tr>
<tr>
<td>HPAEC</td>
<td>High performance anion-exchange</td>
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<tr>
<td>HPLC</td>
<td>High Pressure Liquid Chromatography</td>
</tr>
<tr>
<td>HPV</td>
<td>Human papillomavirus</td>
</tr>
<tr>
<td>ICH</td>
<td>International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use</td>
</tr>
<tr>
<td>IFU</td>
<td>Instructions for use</td>
</tr>
<tr>
<td>IMDRF</td>
<td>International Medical Device Regulators Forum</td>
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<tr>
<td>IOVV</td>
<td>Input Output Verification Validation</td>
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<td>IPC</td>
<td>In-process control</td>
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<tr>
<td>IPV</td>
<td>Inactivated polio vaccine</td>
</tr>
<tr>
<td>IQ</td>
<td>Installation qualification</td>
</tr>
<tr>
<td>IR</td>
<td>Infrared</td>
</tr>
<tr>
<td>ISO</td>
<td>International Organization for Standardization</td>
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<tr>
<td>IVD</td>
<td>In vitro diagnostic</td>
</tr>
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<td>IVDR</td>
<td>In vitro diagnostic regulation</td>
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<td>JP</td>
<td>Japanese Pharmacopoeia</td>
</tr>
<tr>
<td>LL</td>
<td>Lower clinical limit</td>
</tr>
<tr>
<td>LMICs</td>
<td>Low- and middle-income countries</td>
</tr>
<tr>
<td>LOD</td>
<td>Loss on drying</td>
</tr>
<tr>
<td>LPA</td>
<td>Local Production and Assistance Unit in WHO</td>
</tr>
<tr>
<td>LTL</td>
<td>Less-than-lifetime</td>
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<tr>
<td>MA</td>
<td>Marketing authorisation</td>
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<tr>
<td>mAb</td>
<td>Monoclonal antibody</td>
</tr>
<tr>
<td>MAH</td>
<td>Marketing authorisation holder</td>
</tr>
<tr>
<td>MHP</td>
<td>Medicines and Health Products Division in WHO</td>
</tr>
<tr>
<td>mRNA</td>
<td>Messenger ribonucleic acid</td>
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<tr>
<td>MS</td>
<td>Member states</td>
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<tr>
<td>NAb</td>
<td>Neutralizing antibody</td>
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<tr>
<td>NOAEL</td>
<td>No-observed-adverse-effect level</td>
</tr>
<tr>
<td>NOEL</td>
<td>No-observed-effect level</td>
</tr>
<tr>
<td>NRA</td>
<td>National regulatory authority</td>
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<tr>
<td>OOS</td>
<td>Out-of-specification</td>
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<tr>
<td>PAT</td>
<td>Process analytical technology</td>
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<tr>
<td>PBV</td>
<td>Protein-based vaccine</td>
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<tr>
<td>PC</td>
<td>Patient-centric</td>
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<tr>
<td>Abbreviation</td>
<td>Full Form</td>
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<tr>
<td>PCS</td>
<td>Patient-centric specification</td>
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<tr>
<td>PCV</td>
<td>Polysaccharide-conjugate vaccines</td>
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<tr>
<td>PDE</td>
<td>Permitted daily exposure</td>
</tr>
<tr>
<td>PDG</td>
<td>Pharmacopoeial Discussion Group</td>
</tr>
<tr>
<td>Ph Eur</td>
<td>European Pharmacopoeia</td>
</tr>
<tr>
<td>pH</td>
<td>Potential of hydrogen</td>
</tr>
<tr>
<td>PMPF</td>
<td>Post-market performance follow-up</td>
</tr>
<tr>
<td>PMS</td>
<td>Post-market surveillance</td>
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<tr>
<td>ppm</td>
<td>Parts per million</td>
</tr>
<tr>
<td>PQ</td>
<td>Performance qualification</td>
</tr>
<tr>
<td>PQDx</td>
<td>Prequalification of Diagnostics</td>
</tr>
<tr>
<td>PSD</td>
<td>Particle size distribution</td>
</tr>
<tr>
<td>PSV</td>
<td>Polysaccharide-based vaccine</td>
</tr>
<tr>
<td>Q&amp;A</td>
<td>Questions and answers</td>
</tr>
<tr>
<td>QbD</td>
<td>Quality by design</td>
</tr>
<tr>
<td>QC</td>
<td>Quality control</td>
</tr>
<tr>
<td>QMS</td>
<td>Quality management system</td>
</tr>
<tr>
<td>RA</td>
<td>Regulatory authority</td>
</tr>
<tr>
<td>RCT</td>
<td>Randomized clinical trial</td>
</tr>
<tr>
<td>RLD</td>
<td>Reference listed drug</td>
</tr>
<tr>
<td>rpm</td>
<td>Revolutions per minute (rotation speed)</td>
</tr>
<tr>
<td>RPQ</td>
<td>Regulation and Prequalification</td>
</tr>
<tr>
<td>SAMD</td>
<td>Software as a medical device</td>
</tr>
<tr>
<td>SAR</td>
<td>Structure-activity relationship</td>
</tr>
<tr>
<td>SD</td>
<td>Standard deviation</td>
</tr>
<tr>
<td>SLS</td>
<td>Sodium lauryl sulphate</td>
</tr>
<tr>
<td>SOP</td>
<td>Standard operating procedure</td>
</tr>
<tr>
<td>TD50</td>
<td>Median toxic dose</td>
</tr>
<tr>
<td>TGS</td>
<td>Technical Guidance Series</td>
</tr>
<tr>
<td>ToC</td>
<td>Table of Contents</td>
</tr>
<tr>
<td>TRS</td>
<td>Technical Report Series</td>
</tr>
<tr>
<td>TTC</td>
<td>Threshold of theoretical concern</td>
</tr>
<tr>
<td>UL</td>
<td>Upper clinical limit</td>
</tr>
<tr>
<td>UoM</td>
<td>Uniformity of Mass</td>
</tr>
<tr>
<td>US</td>
<td>United States</td>
</tr>
<tr>
<td>USP</td>
<td>United States Pharmacopoeia</td>
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<tr>
<td>UV</td>
<td>Ultraviolet</td>
</tr>
<tr>
<td>VoC</td>
<td>Variants of concern</td>
</tr>
<tr>
<td>XRPD</td>
<td>X-ray powder diffraction</td>
</tr>
</tbody>
</table>
Introduction

Ensuring quality of the health product ensures its safety and efficacy. Manufacturers in low- and middle-income countries (LMICs) face challenges to achieve quality in local production, such as the lack of an available manufacturing workforce trained in quality and understanding regulatory quality standards and difficulties in implementing a quality culture in the manufacturing facility.

The Local Production and Assistance (LPA) Unit in the Regulation and Prequalification Department (RPQ), Access to Medicines and Health Products Division (MHP), WHO, supports Member States (MS), particularly low- and middle-income countries (LMICs), to strengthen sustainable local production and technology transfer to improve timely, equitable access to quality, safe and effective essential medical products. The LPA Unit provides assistance and support to MS with an ecosystem-wide and holistic approach, such as conducting ecosystem assessments for sustainable, quality local production, developing & implementing strategies/roadmaps and tools, providing comprehensive capacity building and technical assistance, including for WHO Prequalification (PQ)/Emergency Use Listing (EUL), and facilitating technology transfer (TT).

To support MS to overcome challenges to locally produce quality-assured vaccines, medicines and in vitro diagnostics (IVDs), the LPA Unit organized a new global training event – Week of Quality – to complement the annual Virtual cGMP Training Marathons organized by the LPA Unit. The first Week of Quality was organized from 16 to 20 June 2023 and focused on establishing quality specifications of vaccines, medicines and IVDs based on WHO and other international guidelines, one of the stepping stones to achieve quality. More than 1700 vaccine and biopharmaceutical manufacturers and regulators attended the sessions on vaccine quality specifications. More than 1400 pharmaceutical manufacturers and regulators strengthened their foundations in establishing quality specifications for medicines. And for the first time, over 1000 IVD manufacturers and regulators, built capacity in narrowing the gap between quality specifications of IVDs and design validation.

Questions raised by manufacturers and regulators in the Week of Quality have been assembled in this training material with questions-and-answers (Q&A) with answers prepared and peer-reviewed by experts with long and rich experience in the vaccine, medicine or IVD industry, national regulatory authority (NRA), and other organizations. This document has been developed to provide manufacturers and other relevant stakeholders with a continuous learning resource and reference document to acquire new and fortify existing, knowledge and capacities to strengthen the local production of quality vaccines, medicines and IVDs.
Setting the standard: achieving quality specification for vaccine manufacturing

Day 1 - 2

Photo credits: © Reuters/Johnson & Johnson, Gloved hand holding a vaccine vial.
SESSION 1 - Establishing patient-centric specifications

1. **To what extent can thorough control of critical process parameters and/or the adoption of continuous process verification alleviate product release testing?**

   If there is a robust manufacturing control strategy, test results on critical quality attributes (CQAs) that do not meet their acceptance criteria should be very rare. Where the process is well-controlled, not all CQAs need to be tested. However, according to current regulations, certain CQAs, such as potency, need to be tested even if the process provides assurance that they will be in an acceptable range.

2. **It is assumed that patient-centric specifications (PCS) are based on data collected from groups of patients, not isolated patients. How can these groups be harmonised, considering individual differences (environment, origin, etc.)?**

   PCS can be supported by a range of data, such as clinical data from trials of the product and, at times, preclinical data, or studies with related products. The key is that regulators and developers, in the context of the manufacturing process and its control strategy, need to have confidence that products meeting their specifications will be safe and effective.

3. **What are the challenges involved in establishing PCS?**

   All specifications need to be patient-centric (PC) in the sense that they must provide assurance of safety and efficacy. However, in a Quality-by-design paradigm, we seek to define the relationship more explicitly between specifications and patient outcomes, consistent with the purpose of specifications. One major challenge is the insistence of some stakeholders those specifications, in addition to providing a connection to patient outcomes, should also provide assurance of manufacturing consistency. However, these are separate concepts and tying specifications to manufacturing consistency risks losing the connection to patient outcomes. This function is much better left to the manufacturing control strategy, which should be thought of as separate from the specifications.

4. **Should the national regulatory authorities (NRA) make PCS a mandatory requirement of product registration?**

   According to current regulations, all specifications must be PC, in the sense that they must assure safety and efficacy. Specifications are not required by regulations in most jurisdictions to assure product consistency - though this is an overlay that some regulators have added. To the extent that broader specifications are justified on clinical (PC) grounds, this provides major advantages to patients, regulators, and developers.

5. **Can a manufacturer use historical data from potency tests on an existing product to set potency limits for another product?**

   Such historical data can contribute to the assessment, though the data are unlikely to be the only factor. Historical data from closely related products may play an even larger role in setting specifications for non-potency CQAs.

6. **80% to 125% around the stated potency is customarily accepted for biotherapeutics. Is such an approach acceptable for establishing the acceptance criteria for vaccine potency?**

   Traditionally, 80–120% has been accepted for certain therapeutics based on assumptions about manufacturing and assay variability. For products (including vaccines) where those assumptions may not hold, it is not possible to pre-define an acceptable range.

7. **If the assay variability is high, how can we be ensured that the actual potency of a lot that passes the potency test is well within the lower and upper clinical limits (UL) boundaries?**

   This is one of the big challenges in production of biological products, including vaccines. Often, both the manufacturing process and the assay
can be significant sources of variability that can increase the difficulty of knowing whether the true potency of a batch will be within a CQA range that assures safety and efficacy. For example, if the assay provides a result at the low end of the range for the CQA where it is believed the product will be effective, it may be 50% likely that the true assay result is above versus below that clinical boundary. This should be addressed by building room for this variability into the specification for the test result. Thus, the acceptance criteria for a test of a CQA need to fall within the clinically acceptable range for the CQA, by a margin that accounts for assay variability.

8. The result of a potency test provides an estimate of the mean potency of the lot, but due to the variability of the test, the actual potency may be more or less than this estimate. In addition, the vial-to-vial variability adds further uncertainty on the actual individual vial potency. How can this risk be mitigated? How reliable is the mean potency for ensuring the efficacy of the whole lot?

The product that was tested in clinical trials (CT) used to establish product safety and efficacy also would have been characterized based on its mean potency, and if the manufacturing process is the same, the variability of product delivered from batches pre- vs. post-commercialization will be similar. Therefore, in the context of the same manufacturing process, the mean potency provides the best indicator of the expected outcome from a given lot. More replicates in determining the mean potency can improve the precision of the mean potency estimate. Please also refer to the answer to the above question.

9. Are statistical justifications mandatory for submitting a Common Technical Document (CTD) application?

It is not possible to reliably assess specifications for products with variable assays and variable manufacturing processes without using statistics.

10. Is it controversial to establish product specifications in the form of a mean ±3 standard deviation (SD)?

Mean ±3 SD might yield specifications for some attributes that are either narrower than would be justified by the actual clinical situation, or that are much broader than are justified by predictions of safety and efficacy. A developer can propose such specifications, but it will still be up to the regulator to determine whether those specifications provide adequate assurance of safety and efficacy.

11. In statistical analyses, is it recommended to use an interval of ±2.58 SD around the mean, as is frequently used in inference statistics, or rather the commonly used ±3 SD limits?

Please refer to the above question. For other statistical analyses, the critical range around the mean depends on what is being analysed and the degree of confidence that is desired in the result.

12. It is assumed that a dose ranging study is an important element in setting the upper and lower clinical limits (LL). Is there another approach to establishing these limits for different products? If not, and if no dose ranging study has been carried out during clinical development, could specification acceptance criteria be established considering manufacturing variability alone?

If the developer provides a justification supporting that product which meets specifications will be safe and effective, and the regulator agrees with that justification, this approach can be used. However, setting specifications solely based on manufacturing variability, at least for some attributes, will reduce flexibility to implement Quality by Design (QbD) and may lead to spurious and unnecessary test failures. In the absence of any alternative, it may be possible to set narrow specifications based on a combination of manufacturing variability data and clinical inference.

13. How is production variability assessed? Given that it encompasses both assay variability and manufacturing variability, how can we distinguish between the two? What parameters should be
Is it based on the results of the process validation?

As noted, production variability (the variability in the assessed attribute when measured on different batches manufactured using the same production process) encompasses assay variability and manufacturing variability. A solid comprehension of assay variability allows deduction of manufacturing variability based on information about the variability in CQA results across the produced batches. Assay variability can be inferred from the assay validation (which is often a best-case scenario) from the stability evaluation (which typically comprises repeat measures of an attribute on the same batch), and/or data from assay performance (for example, when multiple replicates are performed on the same batch to arrive at a mean). Knowing the variability of results on produced batches and understanding assay variability enables the calculation of manufacturing variability in the test results on produced batches by isolating or deducting the assay variability.

**14. Should the batches used to estimate the assay variability be different from those used to estimate production variability?**

There is no requirement for these batches to be different. In fact, manufacturing variability may be more reliably assessed when assay variability is known for the same batches.
SESSION 2 - Clinical qualification of vaccines acceptable ranges

15. From a practical point of view, how can we establish the required link between vaccine analytical potency and clinical efficacy?

Since the potency assay is not a direct measure of efficacy or the potential thereof, the “link” of the potency assay to clinical efficacy must be viewed with caution. Clinical efficacy was established through the clinical development studies while the potency assay and other analytical measurements are designed to demonstrate process consistency of the respective current lot to the original lots used during clinical development.

The so-called link of the potency assay to the clinic rather measures an aspect of the vaccine that is a critical characteristic to result in the desired immune response. For example, in the case of pneumococcal vaccines, antibodies are critical for protection against disease. In turn, the potency assay measures the presence of polysaccharide epitopes. In the case of human papillomavirus (HPV) vaccines, neutralizing antibodies are viewed as critical to protection. In turn, the potency assay measures the presence of neutralizing antibodies.

From a practical perspective, the clinical response and mechanism of action of a vaccine must first be understood. Since potency assay development usually precedes clinical development, thorough scientific understanding in the discovery space can help shaping the understanding of critical characteristics of the immune response and the vaccine. Based on this understanding of the (potential) mechanism of action, one can evaluate which vaccine characteristics are critical to shape and evoke the desired response. These characteristics could then become the target of the potency assay.

For recombinant vaccines, the potency assay often can be the flip side of the clinical immunogenicity assay. That is, the (antigenic) targets of a protective immune response also can become the attributes measured by the potency assay. For complex live virus vaccines, the potency assay often simply measures replication, for example by plaque assay, since this is a critical characteristic for live virus vaccines while the presence of immune targets may be limited to assessment by immunodiffusion assays.

As always, there is no single approach to potency assay design due to the diversity of vaccines’ mechanisms of actions.

16. Given the variability of response in animal models, how reliable is the predictive value of these models for vaccine safety and efficacy in humans? Can data from animal studies be used to predict results in humans prior to clinical studies to limit the size of clinical samples and increase the speed and accuracy of clinical studies outcome?

No, animals are not predictive of the human immune response. However, animals provide value to assess the safety profile to identify high risk parameters. Animal models must be carefully chosen and assessed to be representative for the disease and the vaccine’s mechanism of action.

17. Clinical development cannot consider the great heterogeneity of human physiology and genetic background. How can results obtained from a limited number of subjects/groups be extrapolated to the entire target population? Is the variability of subjects enrolled in CTs considered an advantage to be sought or a disadvantage to be avoided? How can we assess the impact of population variability on vaccine performance?

Yes, clinical development will not be able to capture the full diversity of the human population and the associated diversity of the immune responses. Therefore, often bridging or follow-up studies are carried out beyond the initial pivotal trials to broaden assessments to additional populations.

Particularly late-stage clinical development can benefit from capturing greater diversity to allow conclusions applicable to the entire population of interest. Ideally, sufficiently large numbers of
participants from diverse sub-populations would be included to potentially allow statistical differentiation in case needed. As always, thorough scientific understanding of the vaccine’s mechanism of action, the associated immune response, and its association with specific diversity-related factors is critical and can guide the clinical design and assessments.

The benefit of diversity in early-stage clinical development may be risk-dependent. Since earlier trials contain fewer participants, it is difficult to capture diversity at statistically significant levels. Without statistical power to detect potential signals, i.e. differences associated with specific patient backgrounds, the benefit of added diversity may be limited to potentially detecting worst-case reactions or warning signs that would warrant detailed follow-up. There are also risks of population diversity potentially masking efficacy when there are differences across sub-populations.

Post-approval monitoring and studies can also help to capture the impact of greater diversity beyond initial pre-approval clinical development.

18. How can the risk of antibody-dependent enhancement (ADE) of infection induced by vaccines be effectively addressed? Furthermore, where can one find reliable information on the regulatory requirements to mitigate and manage this potential adverse reaction?

There are no specific regulatory requirements regarding the risk of ADE. It is to be addressed as part of the assessment of the safety profile of the vaccine. This issue is briefly addressed in WHO Technical Report Series (TRS) 979, Annex 2, Guidelines on the quality, safety and efficacy of dengue tetravalent vaccines (live, attenuated), where it is mentioned that the phenomenon of ADE can be observed in a mouse model, but the relevance of this model may be limited with regard to vaccine evaluation. (1,2,3)

19. What might be the potency assay characteristics and readouts of pneumococcal protein-based vaccines (PBV)?

As with other potency assays, the assay characteristics and readouts should be based on the mechanism of action or a closely related aspect thereof.

For polysaccharide-based vaccines (PSV), the protective immune response is linked to antibody (Ab) development. In turn, the potency assays need to confirm presence of the polysaccharide epitopes. For polysaccharide-conjugate vaccines (PCV), the immune-stimulating conjugated protein or the complex of conjugated polysaccharide need to be measured.

For a purely PBV, its properties to elicit a protective immune response should be assessed. For example, if a neutralizing antibody (NAb) response is critical for vaccine efficacy, corresponding neutralizing epitopes might be measured in the vaccine. If a more complex cell-mediated immune response is critical for efficacy, vaccine aspects that lead to the desired immune response should be assessed.

A blanket response is not possible because the potency assay is linked closely to the often-unique mechanism of action. Thorough scientific understanding what aspects of the vaccine lead to protection can guide potency assay development. With that in hand, a discussion with applicable regulatory agencies is encouraged as well.

20. How can conjugate vaccines be tested for conjugated and unbound (free) polysaccharide?

As explained in TRS 977, Annex 3 Recommendations to assure the quality, safety and efficacy of pneumococcal conjugate vaccines, “Methods that have been used to separate unbound polysaccharide before assay, and that are potentially applicable to pneumococcal conjugates, include hydrophobic chromatography, acid precipitation, precipitation with carrier protein-specific antibodies, gel filtration and ultrafiltration. The amount of unbound polysaccharide can be determined by specific chemical or immunological tests, or by high performance anion-exchange chromatography (HPAEC) after hydrolysis.”

21. What are the main differences between analytical quality by design (AQbD) and
the canonical validation of analytical procedures?

AQbD is a holistic approach combining target expectations that are rooted in product performance requirements, thorough scientific understanding, formal risk management, and continuous improvement. In contrast, the canonical approach often is based on process or analytical capability. However, the best cases of the canonical approach often closely resemble to AQbD approach with the latter being more systematic.\(^{(4,5)}\)

22. In the long term, can we envisage a situation where the application of QbD to analytical methods and real time release testing will render final product release testing partially or wholly unnecessary?

QbD, AQbD, and Process Analytical Technology (PAT) have multiple advantages. Foremost, they offer the potential to improve overall product quality through a targeted development process, thorough product understanding, and design approaches to build quality inherently into the product rather than confirming by downstream testing.

In addition, thorough process and analytical design combined with PAT can potentially reduce the testing burden throughout the manufacturing process. However, reduced testing needs to be balanced with the risk of unplanned issues and their mitigation.

PAT tends not to be applicable to the filled, labelled, and packaged product. While one might envision PAT potentially being able to confirm product quality through many of the necessary manufacturing steps, issues such as mislabelling, mispackaging, or damages during labelling or packaging, will likely still require release testing of final packaged product.

The ultimate decision whether such final release testing will still be required should be a conversation with the applicable regulatory agencies and consider quality risks as well as their respective mitigation.

23. Regarding the chimeric vesicular stomatitis virus containing the Ebola glycoprotein (GP), is the test for GP a routine release test required for each drug product lot or, on the contrary, a test to be carried out on the bulk or occasionally, for example as part of the validation or revalidation processes?

Testing requirements should be discussed on a case-by-case basis with the respective RAs. For two approved EBOLA vaccines, the potency release assays are linked to an assessment of the Ebola virus transgene expression.

In the case of Ervebo\(^{\circledR}\), a plaque assay is used for potency assessment together with an ID assay to confirm presence of the Ebola virus transgene. Both were required for release, stability, and process validation; refer to the European Public Assessment Report – EPAR.\(^{(6)}\)

In the case of Zabdeno\(^{\circledR}\), a PCV test is used for potency assessment together with a Western Blot to confirm transgene expression. Both were required here as well for release, stability, and process validation; refer to the respective EPAR.\(^{(7)}\)

24. Down-dosing studies may reveal that fractional doses show efficacy comparable to the target. In this case, should the target dose not be reduced such as to improve the safety profile of the vaccine?

The choice of a vaccine dose needs to balance several risk factors including the safety profile, the effectiveness range, the variability associated with the process/assay, and the potential impact from unknown factors. Depending on the safety profile of the vaccine, lowering the dose might not offer significant benefits but could increase the risk associated with variability and other factors by moving the target potency closer the edge of lower efficacy. The further the target and the product distribution around the target are from the specification limits, the lower is the risk of product failures and/or patients receiving a sub-potent dose.

In a vaccine with a higher safety risk profile, lowering the dose based on down-dosing studies can indeed improve the risk-benefit ratio and could be considered.
As shown in the Ebola vaccine case study, in some cases the risk from disease may also be too great to explore the efficacy range limits safely and ethically.

25. As exemplified in case study 2, all partial-dose formulations, including the 20%, were non-inferior to the full-dose lot. So, strictly speaking, we cannot talk about a subpotent lots. However, data from subpotent lots would be needed to establish the correlation between vaccine efficacy and potency. How should subpotent lots be obtained?

Correct, the lots were not sub-potent. However, they were formulated below the target potency. As such, they could be used to support the usable range around the target. The study did not identify potency limits which would have required including sub-potent lots. On the other hand, that was not necessarily clear from the beginning before generating actual clinical data associated with lower potency lots.

In the example of the case study, the chosen potencies could have been formulated at additional lower targets to reach the limits of acceptable ranges around the target potency. However, the associated patient risk should be balanced with the need for the information. If ranges can be established around the target that are sufficient to support shelf-life specifications, it may not be ethical to expose patients to the risk of lower protection during the time of the trial only to explore the maximum limits of the potency range. After the trial, patients would of course still receive the standard of care, that is the full protective dose.

Formulation at lower potencies than the target allows to define specific potency targets. Using partially degraded lots from probed stability studies may be another route to get to lots with lower potency. However, it would be more difficult to characterize the degree of change in the vaccine so that conclusions on the acceptable potency range may be confounded by other factors related to stability-related changes in the product.
SESSION 3 - Establishing harmonized specifications for new vaccines: lessons learned from the COVID-19 vaccines

26. If all the specification results for the submitted batches exceed the upper limits, as a regulator, should I request the applicant to tighten the specifications?

The following assumes that the situation described is that the test results for all submitted batches exceed the specification. If this is case, and the specification upper limit is the level demonstrated to be safe and no additional safety information is available, then the batches should not be marketed. Additionally, the manufacture should be required to adjust the manufacturing controls related to formulation of the product, such that it complies with the specification.

However, if there is data to support the batches described with an appropriate safety and efficacy profile, then the specification should be adjusted to allow the release of the batches.

Specifications should be clinically justified to ensure appropriate safety and efficacy profiles for a product.

27. If the lower and UL are established by immunogenicity studies, how can we be sure that this immune response will conclusively correlate with the efficacy of the vaccine?

There are some key points to mention before the question posed is addressed. For simplicity, vaccine examples to establish a PC / clinically justified potency specification will be used.

- Even when a potency specification is based on the lots use in a Phase 3 randomized clinical trial (RCT) with an efficacy endpoint, that specification is still clinically justified, and PC.
- Therefore, except when new clinical data either necessitates or supports a change, there is no justification for a regulator to tighten that specification based on improvements in manufacturing capability (for example, process or assay improvements, greater manufacturing experience etc.).

As presented, the importance of maintaining (rather than tightening) PCS is manyfold and includes:

- This rewards manufacturers that invest in continuous improvements regarding process or assays, which is therefore more likely to occur, which benefits patients and industry.
- This provides a scientific and rational basis for a harmonized specification, which, when in place, means that real world data from any jurisdiction is representative of the product and therefore is transferable to all jurisdictions, and not just the product in a specific region.
- A harmonized PCS insures equity of product quality in all jurisdictions.

Regarding the use of immunogenicity to support a broader speciation that would be justified based on the potencies of lots used in a clinical end point Phase 3 RCT, this is an option that a developer may (or should be able to) choose to invest in. However, it is up to the developer to justify that approach to the regulator. The specifics of such an approach are critical and complex.

While the specific details regarding the potency specifications for each product could not be disclosed during the presentation due to the proprietary nature of that information, principles involved were described.

It is also important to note that in each case the actual specification for each product was not set based on the minimum potency tested in the dose ranging Phase 2 studies. The minimum acceptable limit of potency was set for each vaccine at a level consistent with the totality of nonclinical and clinical data, that was supportive of a relationship between the broad characterization of immune responses for both cell-mediated immunity (CMI) and NAb seen in the efficacy studies (i.e., judged to be consistent with the range of efficacy observed in the RCTs over a narrower potency range).

The success of this approach is supported by the clinical effectiveness studies with the
messenger ribonucleic acid (mRNA) COVID-19 vaccines post-authorization using the approved specification, and by the confirmation of robust relationship between NAb and efficacy for the COVID-19 across platforms. Also, please note that even with such a robust correlation, this does not mean that NAb is a mechanistic correlate of protection.

It also important to remember that in the case of Shingrix®, the relationship between (NAb and efficacy was not as robust. This is consistent with the understanding that CMI is more critical to protection against Zoster than NAab, which was known prior to conducting the CTs. This was carefully considered while setting the potency specification of Shingrix®.

In summary, using broad immunological characterization to justify a specification beyond that supported by the Phase 3 lot data is not a simple task. Nor is it a requirement. However, if a manufacture wishes to invest in such an approach, it is incumbent on stringent regulators to have the internal scientific regulatory capacity to appropriately engage with a manufacture to evaluate the robustness of such a proposal, given that it has be demonstrated to be feasible.

28. Should product specifications be based entirely on the PC principle or, on the contrary, a mixed approach could be envisaged, in which the specifications for certain parameters would be clinically based, while for others they would be established on the process capability?

For the reasons summarized in the reply to question 27, in general, all specifications should be PC. However, that does not mean that all the data in support of a PCS needs to be generated from a product specific CT. A PCS may be supported by a defined pharmacopeial limit or another relevant standard, that has been established to be appropriate. Additionally, a PCS may and be established thorough prior knowledge, as justified, and such considerations are for a competent regulator to evaluate.

The central point is that a specification should be understood (i.e. demonstrated directly or indirectly) to be supportive of a defined acceptable level of safety and efficacy and therefore would be PC. The corollary is that just because a product process capability of a manufacturer is established over a specified range, that does not make it suitable or acceptable.

29. In the Shingrix® example, it was observed that the rate of Ab response varied according to the dose administered. Has a minimum response threshold been established and, if so, what is it?

The reply to question 28 is relevant to this question as well. As suggested in the reply to the question 28, for Shingrix® the Ab response at the lowest dose in the phase 2 trial (25 µg gE in AS01®) was not considered to overlap with upper two doses. Hence, the lower limit was interpolated using those results, as well as consideration of the cell-mediated response to the three doses. Also as noted in the reply to above question, the specific lower limit is proprietary information.

30. It is understood that regulatory agencies collaborate to harmonize the drug product specifications. Does this effort extend to specifications of raw and starting materials, or excipients (e.g. adjuvants, stabilizers, and/or preservative)?

All materials should be appropriately controlled to ensure product consistency in compliance with the specification. That would include adjuvants, stabilizes and/or preservatives. However, what controls are in place, and which of the CQAs are defined on the specification is for an agency (or agencies) to determine with the manufacturer.

31. Is the PCS approach also applicable to products other than vaccines, such as biotherapeutics or medicines, including, for example, oral solid dosage forms?

The need of a clinical justification for product specifications would apply to all products. However, the manufacturing and control strategies for vaccines, therapeutics and pharmacologic drugs are quite different, as is the level of control and product consistency achievable with each.
As is well understood, manufacturing of biologic drugs is inherently more variable than with pharmacologic drugs. Additionally, the complexity of control with a vaccine that is required to be immunogenic is essentially the opposite of that required for a biotherapeutic, that must be biologically functional but nonimmunogenic.

This also has implications for what can realistically be demonstrated directly in the clinic for biotherapeutics versus vaccines, but clinically relevant information must still be provided to justify a specification.

32. How can preclinical data be used to establish specifications, given that the scale of immune responses is likely to be different?

This is another good question in which the specifics can be complex, but it is possible to provide some general guidance.

It is generally correct to assume that responses or outcomes in animal models are not necessarily predictive of those in humans, which is why RCTs are undertaken. However, in specific situations it is possible to demonstrate relationships between responses in animal models and humans that are useful. This can also be the case with in vitro data involving both human and non-human cell systems for both vaccines and biotherapeutics.

One current example with the COVID-19 vaccines involves the use non-clinical immunogenicity studies in mice to confirm the functionality of updated COVID-19 vaccines against variants of concern (VoC) prior to authorization in the absence of clinical data. This is not because mice are a suitable replacement for human CTs. It is because numerous studies have been undertaken in mice, hamsters, and non-human primates, in addition to the extensive and robust RCTs in humans, to establish that for the authorised vaccine platforms, there is an acceptably high degree of confidence that robust immunogenicity in mice translates to robust immunogenicity in humans. Additionally, this has been confirmed by post-authorization vaccine effectiveness studies in humans.

33. What are the methods used to estimate the T cell and cellular immune responses?

The methods used for T cell and other cell mediated responses vary widely and numerous approaches are published. While these data can be very useful within the context of a single study, or potentially a family of related studies, it is not generally possible to compare results between groups. Even data variability between studies within the same group can be a challenge, and considerable work is ongoing to address this important topic.

34. How can you predict whether a vaccine will elicit the production of immunoglobulin G or M, which are effective NAb?

While certain adjuvants are associated with different classes or subtypes of antibodies (Ab), this often varies with the proteins and the vaccine platform involved. As a result, the type and extent of NAb production is predictable to some extent but needs to be verified empirically.

35. Could we have more details on the predictive models of immune protection to be used to establish correlates of protection?

a general lecture on correlates of protection can be found in the used references. (8,9)
SESSION 4 - Replacement of in vivo potency testing by in vitro assay

36. Would it be possible to share a standard operating procedure (SOP) for an in vitro enzyme-linked immunosorbent assay (ELISA) potency assay for rabies vaccines?

A collaborative study to standardize an ELISA method is ongoing. The SOP could be shared after completion of the study, its integration in the European Pharmacopoeia (Ph Eur) and publication in a scientific review. Unfortunately, this will take time. (10)

37. What are the different approaches to establish the correlation between in vivo and in vitro potency assays? Is there any guidance document that explains how to establish such a correlation?

There is no specific guidance on how to establish a correlation. To assess properly the correlation between in vivo and in vitro methods it is important to have lots of product that differ in the magnitude of the response: compliant lots with the in vivo assay from the regular production (commercialized lots), non-compliant lots with the in vivo assay – sub-potent lots - obtained by alteration/degradation or sub formulation and highly-concentrated lots with the in vivo assay – sus-potent lots - obtained for example by over formulation. With this panel of lots, both tests should be carried out. A regression analysis of the results including an analysis of variance should be done and the correlation coefficient R^2 should be determined. (11,12,13)

38. For adjuvanted combined vaccines, how can we be sure that the in vitro trend result is representative of the in vivo trend result?

The trend of in vivo result is representative of the specific antigen of the vaccine even if the adjuvant enhances the immune response. Sometimes one component of the combined vaccines may act also as an adjuvant of the other components. The in vivo trend represents the protection or the immune response against the specific antigen (active substance) and not against the adjuvant which plays as a booster.

And the in vitro trend measures the quantity and quality of the active substance (the specific antigen) which induces the specific protection or the specific immune response. Both trends appraise the specific immunogenicity that is given by the active substance although the adjuvant induces a non-specific immune response.

The adjuvants serve 1) to better present the antigen to the immune system because without adjuvant the protein would be less recognized by the immune system as an antigen and 2) to slow the spread and destruction of the antigen in the body allowing a better and longer reactivity of the immune system against the antigen.

The adjuvants allow also to reduce the quantity of antigen in the vaccine and to decrease the number of injections.

39. Are the 3Rs (Replacement, Reduction and Refinement) principles also applicable to the non-clinical development, leading to the possible discontinuation of studies such as the determination of no-observed-effect level (NOEL) or no-observed-adverse-effect level (NOAEL)?

The 3Rs principles are implemented at an international level in the International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) guidelines, and more specifically the ICH M3 (R2) guideline which provides guidance on how to perform non-clinical studies required for the initiation of human CTs. The 3Rs principles are also reflected in other non-clinical ICH guidance documents, such as ICH S9. In the European Union (EU) regulatory framework, “the protection of animals used for scientific purposes” and the 3Rs principles applies, with a few exceptions, to all scientific or educational studies where animals are used, including non-clinical studies for the development of medicinal products. Therefore, the 3Rs principles are applicable to non-clinical studies performed in relation to development of human and veterinary medicinal products.
Currently, a complete discontinuation of repeat-dose toxicity studies which are performed to determine NOEL and NOAEL is not possible. However, as for example seen with marketing authorisation (MA) applications for mRNA and viral vector vaccines, the acceptance of non-clinical toxicity studies conducted with the same technology platform is an approach that will significantly reduce the number of animal studies.

40. **May an in vitro potency assay be implemented early, from the very beginning of the development of a new vaccine, without having to go through preliminary animal potency test?**

Yes of course. This is the best strategy to develop an in vitro potency assay in parallel to the development of a new vaccine. The *in vitro* assay should be demonstrated relevant to appraise the protection or the induction of immunity. This is why depending on the drug substance (DS) the potency may be measured by two in vitro assays.

It is crucial to link the in vitro potency results with the lots used in the CTs to demonstrate that the in vitro potency assay(s) reflect(s) the seroconversion and immunogenicity on human induced by the immunization of the different formulations of the vaccine.

41. **How to validate an in vitro virus neutralization test? Would it be possible to share a validation protocol addressing the validation of a fluorescent antibody virus neutralization (FAVN) test?**

There is no specific validation protocol to validate an in vitro virus neutralisation assay such as the FAVN test. The principles of an assay validation are common for all the assays. For international guidance addressing the validation of analytical procedures, including potency assays, please refer to ICH Q2(R1): Validation of analytical procedures: text and methodology.

42. **When developing an in vitro assay, is it advisable to validate for robustness in the event of minor changes in the method parameters that might occur when the method is conducted in a different laboratory?**

It is not necessary to perform a full validation for minor changes in the method parameters in the same laboratory. But a comparison of the results obtained with the new version of the assay with the results obtained with the current version of the assay is required.

For a transfer of the method from one lab to another lab, a validation is required except the linearity and specificity.

43. **In general, in vivo results are collected over time, whereas the achievement of in vitro test is more immediate, hence the difference in lifespan between in vivo and in vitro tests. Does this pose a problem and, if so, how can it be resolved?**

This is not an issue. An in vivo potency assay takes about two months. The in vitro test can be carried out at any time during this period.

44. **What are the recommended acceptable regulatory acceptance criteria for the comparison of in vivo and in vitro test methods as part of the method comparability and suitability study for a vaccine?**

There are no regulatory acceptance criteria for the comparison of in vivo and in vitro tests methods. But it should be demonstrated that:

- a compliant lot with the in vivo test is also compliant with the in vitro test;
- a failing lot with the in vivo test, also fails the in vitro assay.

However, as the in vitro assay is more sensitive and discriminant than the in vivo assay, a compliant lot (a borderline lot) with the in vivo assay may fail the in vitro assay: (14)

45. **How can the linearity of an in vivo test method be checked?**

The linearity of a method is checked in the same way whether the method is in vivo or in vitro. Please refer to the chapter on linearity in ICH Q2(R1): Validation of analytical procedures: text and methodology.

46. **Assessment of in vivo activity considers factors such as absorption, distribution, metabolism, and excretion of the drug in an organism. These processes can**
significantly affect the overall efficacy of a compound. In vitro assays generally focus on the specific target or mechanism of action, neglecting the wider pharmacokinetic and metabolic aspects. Wouldn't that be a limitation of in vitro potency assays?

This is correct. In fact, the optimal goal of the in vivo assay is to demonstrate the protective ability of the vaccine or its immunogenicity by the induction of antibodies in a certain species of animals. However, most of the time the animal species selected for in vivo potency assay does not reflect what happens in humans regarding absorption, distribution, metabolism, and excretion of the product as the human metabolism is different. For example, the in vivo potency assay for inactivated polio vaccine (IPV) could be carried out on chicken, guinea pigs or rats. But only the protective activity of the IPV vaccine performed on rats correlates with the protective activity of the IPV vaccine in humans.

The objective of the in vitro potency assay is to quantify the quality attributes of a product and its functionality which was shown to induce protection and immunogenicity in humans. This why it is crucial to carefully characterize the in vitro assay (or its critical reagents) in its ability to appraise the functionality of the relevant protein/epitopes of the product involved in the induction of neutralizing antibodies and the protection.
SESSION 5 - Addressing the setting of specification in a CTD application

47. What is meant by “potency does not mean efficacy”?
A potency assay is an analytical procedure that does not measure clinical efficacy; instead, it measures biomarkers such as antibodies. Potency, thus, refers to the ability of a vaccine to elicit an immune response. Vaccine efficacy is assessed separately, by means of clinical studies and refers to the ability of a vaccine to prevent disease.

48. Who should determine the specifications of a product, the manufacturer, or the regulator?
Where compendial documents or regulatory recommendations exist, these are authoritative. If no such provisions exist, the specifications should be proposed and justified by the manufacturer and approved by regulatory authorities. Because of its knowledge of the product and the process, the manufacturer plays a prominent role in establishing a range of tests and acceptance criteria.

49. Should the drug product be tested for formaldehyde when this material is used in the manufacturing process? Alternatively, could an intermediate product be tested instead?
The WHO TRS stipulate that formaldehyde residues must be tested in the final bulk. For instance, consider the following:

- **TRS 993, Annex 3 Recommendations to assure the quality, safety, and efficacy of poliomyelitis vaccines (inactivated)**
  A.4.6.3 Residual formaldehyde: the content of free residual formaldehyde in the trivalent bulk should be determined by a method approved by the NRA. The limits should be approved by the NRA.
  However, there are RAs that allow some flexibility. For instance, the Ph Eur allows the free formaldehyde to be tested on the bulk purified antigen/toxoid.

- **Ph Eur 0443 Diphtheria vaccine (adsorbed)**
  Provided the free formaldehyde content has been determined on the bulk purified antigens or on the final bulk and it has been shown that the content in the final lot will not exceed 0.2 g/L, the test for free formaldehyde may be omitted on the final lot.

- **Ph Eur 0452 Tetanus vaccine (adsorbed)**
  Provided the free formaldehyde content has been determined on the bulk purified toxoid or on the final bulk and it has been shown that the content in the final lot will not exceed 0.2 g/L, the test for free formaldehyde may be omitted on the final lot.

Therefore, it is not excluded that a regulatory authority (RA) accepts that the test for free formaldehyde is performed earlier in the process than being performed as a release test on the finished product, if adequately justified. This is to be discussed with the NRA.

50. Is potency referring to a quantity, a specific activity, or both?
Potency (expressed in units) is the quantitative measure of biological activity. Potency of a biologic is the equivalent to the "strength" of a medicine. However, whereas the strength of medicines is based on mass of active ingredient, the strength of biological products cannot be measured in mass units as mass is a reflection of the physical quantity of material and does not reflect its biological activity. Potency can be reported in absolute terms or expressed per unit of mass or, most often, volume.
51. What is the ideal and acceptable process for managing seasonal strain changes and their impact on final product specifications, allowing for a more rapid response to the market?

For seasonal influenza vaccines, annual changes in vaccine strain composition are considered to be moderate quality changes. Specifications are not expected to be impacted by these changes. Guidance can be found in section 8.2 of WHO TRS 993, Annex 4 Guidelines on procedures and data requirements for changes to approved vaccines.

52. When applying for emergency use listing (EUL), to what extent can test and trial parameters be rationally reduced?

As explained in the scope of the guidance document EUL procedure, Version 9, August 2022, EUL is a special procedure for unlicensed vaccines, medicines and in vitro diagnostics in the event of a public health emergency when the community/public health authorities may be willing to tolerate less certainty about the efficacy and safety of products, given the morbidity and/or mortality of the disease and the lack or paucity of treatment, diagnosis/detection or prevention options. It is therefore acknowledged that at the moment of submission, data will not be complete. A Point to consider has been published, providing a guidance on the level of expected completeness of information for COVID-19 vaccines EUL applications (15,16)

53. Is it possible to reduce the frequency of tests or omit certain batches while maintaining quality?

Using the enhanced approach, QbD, could support real time release testing. As such, it has the potential to reduce the extent of drug product release testing (refer to ICH Q8(R2) Pharmaceutical development). Moreover, some guidelines make clear reference to the fact that testing can be reduced when experience is gained (e.g. TRS 1039, Annex 3, Evaluation of the quality, safety, and efficacy of mRNA vaccines for the preventions of infectious diseases: regulatory considerations).

54. If the storage temperature of a COVID-19 vaccine is 2 to 8 °C, at what temperature can it be said to have lost its efficacy?

As COVID-19 vaccines display different stability profiles, there is not direct answer to this question. Moreover, the inactivation is not only a matter of temperature reached but also of duration of exposure to temperatures above the traditional 2–8 °C cold chain. For instance, the Pfizer-BioNTech COVID-19 vaccine is thought to lose its efficacy after being stored at room temperature (20-25 °C) for more than 12 hours.(17)

55. Does potency always decrease over time?

Some vaccines have a very high stability profile and as such, suffer little or no loss of potency under normal storage conditions (e.g. heat stable, live attenuated rotavirus vaccine).

56. What are the clean room grade recommendations for vaccine production activities, including storage of raw materials and finished products?

The production of vaccine is in the scope of the WHO document on good manufacturing practices (GMP) for sterile pharmaceutical product (TRS 1044, Annex 2), where the different operations are assigned to their respective cleanroom grade. Storage of raw materials is not reported in the operations to be conducted in classified areas (refer to table 4 of TRS 1044, Annex 2), indicating that unless otherwise justified, that there is no need to classify warehouses. However, temperature and humidity should be controlled. As for the vaccine finished product, the classification of storage areas may range from non-classified to grade C, depending on the product to be stored and the risk of contamination.

57. If no international standard is available, how can the value of internal standards be calibrated?

Where no international reference standard exists, a characterized in-house reference material should be established. In-house units should be used to report assay results of production lots if bridging to an international reference standard is not possible.
58. How can the potency assay reflect the mechanism of action when we don’t know exactly how the body’s immune system responds?

As explained in the principles of vaccine potency assays, publication referenced below, a live virus vaccine, for example, causes a mild ‘infection’. The attenuated virus must replicate to elicit the desired immune response. Thus, the potency is determined by the number of infectious particles present in the vaccine. As such, infectivity reflects to some extent the mechanism of action. But it is recognised that in some cases, the vaccine’s mechanism of action is unknown. A potency test that reflects the mechanism of action is an ideal that cannot always be achieved. (18)

59. Is the reprocessing of vaccine drug products allowed?

Whilst reprocessing of certain pharmaceutical products may be permitted under certain conditions, one of which is that the original drug application includes a proposal for reprocessing procedures, this is difficult to apply to sterile final products for which, therefore, reprocessing should not be permitted. In contrast, reprocessing procedures can be considered for intermediates. In such cases, these procedures should be validated and pre-approved as part of the MA. Refiltration of the final bulk is often allowed if the filter fails the post-filtration integrity test. (see TRS 986; Annex 2)

60. Where can I find guidance documents or guidelines on the regulation of vaccines?

The WHO’s regulatory guidance documents on vaccines can be consulted on the web page dedicated to vaccine standardisation. (19)

61. Do all vaccines, including protein and peptide vaccines, fall within the scope of ICH guideline Q6B on the specification of biological and biotechnological products?

The principles adopted in ICH Q6B apply to highly purified and characterized proteins and polypeptides, their derivatives, and products of which they are components (e.g. conjugates). This document does not cover conventional vaccines, synthetic peptides, and RNA or deoxyribonucleic acid (DNA) products.

62. What is the stance of regulatory authorities regarding in-process testing during the intermediate stages of vaccine manufacturing? It is known that in-process testing is recommended, but there are concerns that regulatory authorities discourage it due to the perception that it indicates a lack of validation for critical quality parameters (CQPs) and may compromise the safety of the product. Please provide guidance on this matter.

The question assumes that in-process control (IPC) is a sign of poor process validation and discouraged by regulators, but it is believed that these statements are not correct. IPC is expected to be part of the process control strategy, and it is expected to be well justified based on the development and technology transfer work, and the process validation itself. Moreover, there is a trend towards real-time release based on IPC, which contradicts the concepts mentioned in the question. However, IPC are not intended to fully replace quality control (QC) testing. For instance, an intermediate product should undergo a formal QC test, rather than just an IPC.

63. How can putative nitrosamine contaminations be considered in the specifications of vaccine or monoclonal antibody (mAb) drug products?

Apart from those containing chemically synthesised fragments, those using processes with nitrosating reagents, or those packed in certain primary packaging material such as blister packs containing nitrocellulose, biologicals including vaccines and non-conjugated mAb are considered at low risk of nitrosamine contamination. Consequently, it is unlikely that a nitrosamine risk assessment would lead to a change in the specifications.

64. Is it possible to carry out an independent batch release based on a review of the documentation alone?

Yes, this is possible. Each vaccine batch should be released by the national regulatory authority
(NRA) from the country that produced the vaccines. This official batch release can consist of a review of the summary protocol only, or review of the summary protocol combined with independent testing.

65. What is the process for the NRA to releasing batches of vaccines produced by the manufacturer?

WHO guidance on independent batch release is provided in TRS 978, Annex 2 Guidelines for independent lot release of vaccines by regulatory authorities.

66. How can the proposed acceptance criteria for impurities in vaccines be justified?

The justification of specification for impurities can be based on:

- compendial documents (e.g. endotoxin);
- official recommendations (e.g. WHO recommended limit of 10 ng host cell DNA/parenteral dose);
- Threshold of toxicological concern, or accepted daily intake for chemical impurities;
- Non-clinical and clinical studies for those impurities where no regulatory or scientifically recognised threshold exist.

67. Could you briefly explain any product-related impurities that need to be considered in the DS, final bulk or Drug Product (DP) specifications for various vaccines?

In the case of therapeutic proteins (e.g. recombinant proteins, mAb), structural heterogeneity should be avoided as much as possible, as it may modulate the mechanism of action. The development of an immune response against therapeutic proteins, enhanced by the presence of impurities, may impact their efficacy and safety, especially when administered as multiple doses over prolonged periods. In contrast to this, the development of an immune response is precisely the desired effect for vaccines, and if it is raised against a wider spectrum of variants, even better. As a result, variants of the desired product that would have been considered as product-related impurities in therapeutic proteins turn out to be product-related substances in the case of vaccines. However, product-related impurities have been identified in some vaccines such as, for instance, non-infective virus particles in live attenuated vaccines. Sometimes, they may be critical to the vaccine’s safety profile, as in the case of residual pertussis toxin, toxoids reverting to toxicity, or neurovirulent mutants in oral polio vaccines.

68. Could you briefly explain how to approach the validation of the elimination of impurities (such as DNA or host cell protein)?

Validation of the elimination of impurities can be carried out in the same way as validation of the manufacturing process, e.g. by following the traditional process validation approach and showing on an appropriate number of batches that the impurities are below the threshold limit.

69. Are distinct release and shelf-life specifications applicable to vaccines?

Yes, these two sets of specifications are applicable to vaccines. As explained in TRS 962, Annex 3 Guidelines on stability evaluation of vaccines, shelf-life specifications are those specifications that should be met throughout the shelf-life of the vaccine, whereas release specifications are specifications that a lot of a product should meet at the time of release in order to assure that the lot will maintain adequate quality throughout its shelf-life. Where no distinction is made between release and shelf-life specifications, it is considered that the drug product specifications reported in the dossier are those that should be met throughout the shelf-life of the vaccine, i.e. the shelf-life specifications.

70. Should the shelf-life specifications be based on the LL?

Yes, the shelf-life specifications should be based on the LL as determined during the clinical development.

71. If a CT is to be conducted over 24 months, how much stability data need to be generated to support the stability profile of the clinical material?
As explained in TRS 962, Annex 3 Guidelines on stability evaluation of vaccines, states that sufficient stability data should be generated to characterize the stability of the lots during CTs. Mathematical models may be used to estimate the potency of vaccines given in CTs. This guideline also mentions that modelling of the minimum release specification with less than 12 months of data is highly unreliable.

Other regulatory documents such as the European Medicines Agency’s Guideline on the requirements for quality documentation concerning biological investigational medicinal products in CTs, specify that the maximum shelf-life after the extension should not be more than double than the period covered by real time stability data obtained with representative batch(es). While it should be emphasized that the needed amount of stability data is to be assessed on a case-by-case basis, we can cautiously assume that Ras could require at least 12-month real-time stability data in this case. Supportive accelerated stability studies could provide further guarantee of the stability profile of the vaccine. (20)

72. Can the stability data also be used to derive the specification acceptance criteria?

Indeed, as specified in ICH Q6B Specifications: test procedures and acceptance criteria for biotechnological/biological products, specifications, and especially shelf-life specifications, should account for the stability of the drug product.

73. What is the rationale for discontinuing the abnormal toxicity test?

During the 2018 WHO Expert Committee on Biological Standardization, was recommended the immediate discontinuation of the inclusion of the innocuity test. As specified in section 3.1.3 of TRS 1016, "current manufacturing processes, which include the implementation of GMP and comprehensive QC measures (including IPC), were considered to be more appropriate than the innocuity test in assuring the quality and safety of vaccines and other biological products. The Committee then reviewed the historical inclusion of the innocuity test in documents published in the WHO TRS and concluded that its complete omission would not compromise the quality and safety of vaccines and other biological products."

74. Can we expect an impact of artificial intelligence (AI) on the establishment of PCS, or on the replacement of in vivo tests by in vitro methods?

Currently, it is difficult to anticipate what will be the actual impact of AI on product testing, but it is likely that this innovation will provide a panel of applications for the vaccine industry including, but not limited to, monitoring and controlling advanced manufacturing processes. In this regard, it should be noted that the United States (US) Food and Drug Administration (FDA) issued a discussion paper on AI in drug manufacturing, “meant to facilitate early input from stakeholders outside the Agency. The Agency intends to consider such input in developing a future regulatory framework”. (21)

75. If it is demonstrated that a potency higher than the target can be used safely, is it necessary to optimise the process as to reach the target limit without exceeding it?

If it has been demonstrated that a potency higher than the target is safe, the actual potency can exceed the target potency provided it does not exceed the upper clinical limit (UL). In this regard, it is emphasised that many vaccines have been approved without an upper limit of potency.

76. What is the difference between up- and down-dosing studies?

In the down-dosing studies, sub-potent vaccine batches were used, while in the up-dosing studies, vaccines with a potency higher than the efficacious dose w
Ensuring excellence: getting the specification for medicines right

Day 3 – 4

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SESSION 6 – Active pharmaceutical ingredient monographs

1. What are the major factors to be considered when setting specifications for APIs?

When establishing specifications for APIs the regionally applicable pharmacopoeial monograph, if available, and the general notices and monographs of that Pharmacopoeia must be taken into consideration, as well as the recommendations for universal and specific tests and criteria described in the ICH guidelines Q6A, Q3A, Q3C. Please refer to WHO Technical Report Series (TRS) 970, Annex 4.

When establishing specifications for APIs, reference should be made to the recommended approach and considerations; as well as the universal and specific tests and criteria, described in ICH guidelines Q6A, Q3A, Q3C. Please refer to WHO TRS 970, Annex 4. In addition, the applicable pharmacopoeial monographs, and the general notices and monographs of that Pharmacopoeia may need to be taken into consideration, as well.

2. To what extent are the different internationally recognized pharmacopoeia interchangeable, i.e. International, European, British, US and Japanese Pharmacopoeia (JP)?

The Pharmacopoeial Discussion Group (PDG) of the European Pharmacopoeia (Ph Eur), the JP, and the United States Pharmacopeia (USP) have expressed a commitment to achieving harmonization of the procedures in a timely fashion. Where harmonization has been achieved, an appropriate reference to the harmonized procedure and acceptance criteria is considered acceptable for a specification in all three regions. For example, after harmonization sterility data generated using the JP procedure, as well as the JP procedure itself and its acceptance criteria, are considered acceptable for registration in all three regions. To signify the harmonized status of these procedures, the pharmacopoeias have agreed to include a statement in their respective texts which indicates that the procedures and acceptance criteria from all three pharmacopoeias are considered equivalent and are, therefore, interchangeable. Please also refer to ICH guideline Q6A.

Information on pharmacopoeial harmonization and the status for the general texts on the PDG work plan is publicly available, for example on the website of the European Directorate for the Quality of Medicines and Healthcare (EDQM).

3. Can description and solubility of an API differ from the information stated in the respective pharmacopoeial monograph?

Appearance and solubility are inherent to the API and can therefore not be different from the information in the respective API monograph. Note that the solubility stated is the approximate solubility of the API.

4. Should solubility be tested for every API batch if it is under ‘Characters’ instead of ‘Tests’ in a monograph?

Some sections of an API monograph provide statements that are for information only, whereas other sections include pharmacopoeial requirements. The lay-out and terminology, including section headings of monographs, may not be identical among the different pharmacopoeias. Information on how to interpret a monograph can generally be found in the respective pharmacopoeia, for example under General Notices of the Ph Eur.

Generally, regulators do not require solubility to be tested routinely in the presence of acceptable identity and other relevant tests.

Please refer to ICH guideline Q6A for information on the frequency of testing of individual parameters. Note that batches that are not tested for a specific parameter still must meet the requirements for that parameter.

5. Should the test for identification and impurities be performed on every API batch for release testing?

Please refer to ICH guideline Q6A for information on the frequency of testing of individual parameters. Note that batches that are not tested for a specific parameter still must meet the requirements for that parameter.
Any decision to skip-test must be justified and documented by the manufacturer and this justification available for review. This justification should not simply be limited to a statistical analysis of historical batch data but should consider the scientific nature of the parameter being skipped, the risks this creates, and the circumstances surrounding the preparation of the batch being tested.

Generally, it is expected that every batch of API is tested for identification. Since impurities are an important quality characteristic of an API, skip-testing of one or more impurity tests needs to be considered carefully.

6. Can test parameters in the specification be taken from different pharmacopoeia for a single API? For example, Impurities from USP and Assay from the British Pharmacopoeia (BP)?

The specifications of an API should first be determined based on the specific preparation of that API, not the availability of an API pharmacopeial monograph. Regarding impurities, the outcomes of investigations undertaken will determine the presence and levels of impurities present. However, available monographs are an excellent reference when trying to understand the possible impurities that may be present.

Typically, a quality standard for the active substance must be stated, i.e. a specific pharmacopoeial standard or in-house standard. If compliance with the regional pharmacopoeia is mandatory or a manufacturer voluntarily claims their API complies with a pharmacopoeial standard, then compliance with the standards and tests outlined in the pharmacopeia are mandatory.

If the proposed test parameters, analytical procedures, and criteria are different from the standard claimed, this must be justified in the dossier. In general, it should be demonstrated that the proposed control is at least equivalent or more stringent than that of the claimed pharmacopoeial standard. If alternative analytical procedures are used, it should be demonstrated that compliance with the monograph is still achieved (cross-validation).

7. What is the difference between an API specification and an API's Certificate of Analysis (COA)?

Per ICH Q6A guideline, a specification is defined as a list of tests, references to analytical procedures, and appropriate acceptance criteria, which are numerical limits, ranges, or other criteria for the tests described. The specification establishes the set of criteria to which an API should conform to be considered acceptable for its intended use. The COA includes the results for a specific batch that is tested for compliance with the specification.

8. Some companies store all APIs at refrigerated conditions, even those that can be stored at or above 25 °C according to the label. Is this acceptable?

The storage conditions for an API should be appropriate for that API and should follow the labelling, which is based on the results of stability testing. Please refer also to WHO TRS 908.

9. Can the storage conditions for an API from a certain manufacturer be different from what is stated in the pharmacopoeia the manufacturer claims?

Manufacturers should develop stability data as part of API development and continue to supplement this with on-going stability studies.

In general, a deviation from the storage condition of the claimed pharmacopoeial monograph should be supported by evidence that the proposed storage condition is suitable. This may include evidence that, for instance, protection from light, is not required for the API. This evidence should be generated in line with relevant guidance; see ICH Q1A, Q1B, Q1D and Q1E.

Note that the storage conditions should be clearly defined, e.g. store below 25 °C, rather than reference to subjective terms such as room temperature.

10. How often should you revise your API specification when utilizing a monograph that undergoes yearly revisions?
The API specifications should be reviewed on a periodic basis and if there are changes to the API preparation or any other factor that might impact API quality.

When compliance to a pharmacopoeial standard is claimed, the API should comply with the current version of that monograph and therefore the specification should be updated when a revised monograph is issued. For implementation timelines regional regulation must be consulted.

11. Is it possible to source the API from two different manufacturers for the manufacturing of a finished product if both sources are approved and qualified?

Regulators accept the use of APIs sourced from multiple suppliers if the API from each source is demonstrated to be of acceptable quality and of comparable characteristics in terms of user parameters such as polymorphic form and particle size distribution (PSD) when these are relevant.

In a case where there is more than one supplier of the API, the product dossier should include one complete “3.2.S” section for the API from one manufacturer, followed by another complete “3.2.S” section for the API from each of the other manufacturers. The applicant’s/finished pharmaceutical product (FPP) manufacturer’s API specification should be one single compiled specification. The test parameters, analytical methods and acceptance criteria do not need to be identical for each supplier. When justified, it is acceptable, for example, to include a statement “for API from manufacturer A” (e.g., in the case of residual solvents). Please refer to WHO TRS 970, Annex 4.

12. If we claim two pharmacopoeial standards for our API specification, do we need to include impurities from both references?

When two specific pharmacopoeial standards are claimed, the API must comply with the requirements of both monographs and the general chapters and monographs of each pharmacopoeia. In many cases, however, there is overlap in the tests, limits, and equivalency of test methods, so this may not always mean the need to conduct two separate sets of tests.

13. For APIs that are currently not available in any monograph, what type of quality information will be required by WHO for Prequalification (PQ)?

The required API information for submission to WHO PQ is described in WHO TRS 970, Annex 4 and the guidance documents published on the WHO PQ of Medicines webpage.

Note that with few exceptions, there is little difference between the information required to be submitted for a pharmacopoeial API and a non-pharmacopoeial API. This is true for PQ related submissions and submissions to other medicine regulators.

14. Is it permissible for the FPP manufacturer not to perform all the tests described in the monograph and to use the results of the API manufacturer’s COA? And if so, which tests are mandatory?

The applicant/FPP manufacturer is responsible for the quality of the API and should include his own specification for the control of the API in the product dossier. The applicant/FPP manufacturer is also responsible for testing of the API as described in the product dossier. The process of API qualification and the prerequisites for implementing a system for reduced sampling and testing upon receipt of the API should meet the regionally applicable GMP requirements and be based on trend data on several batches tested by the applicant/FPP manufacturer and the principles of vendor qualification. The frequency of testing should not normally be stated in the specification by the applicant/FPP manufacturer.

15. What is the acceptable limit for the control of residual solvents that belong to ICH guideline Q3C class 3 by loss on drying (LOD)?

For solvents in class 3 it is considered that amounts of these residual solvents of 50 mg per day or less (corresponding to 5 000 ppm (parts per million)) or 0.5% under Option 1 of ICH guideline Q3C would be acceptable without justification. Residual solvents are typically
determined using chromatographic techniques such as gas chromatography. If only class 3 solvents are present, a non-specific method such as LOD may be used. A limit for LOD of 0.5% is generally acceptable. When this level is exceeded, residual solvent content should be determined by a specific method.

16. How are the limits for class 4 solvents established?

For solvents in Table 4 of ICH guideline Q3C no adequate toxicological data on which to base a Permitted Daily Exposure (PDE) is available. Manufacturers should therefore provide a toxicological justification for residual levels of these solvents in pharmaceutical products.

17. During the synthesis of the API, if a manufacturer uses a Class I solvent (for example, benzene) in the final stage of the synthesis, can this be accepted if the API manufacturer can demonstrate that the levels of this residual solvent in 3 representative pilot scale batches are within ICH guideline Q3C limits?

No solvents that are known to cause unacceptable toxicities, such as Class 1 solvents, should be used in the synthesis of the API unless their use is unavoidable. One of the few acceptable justifications is if the material itself forms part of the final API structure.

18. If solvents used in the last step of synthesis or purification are those known to be potentially contaminated with benzene, like methanol, and the manufacturer demonstrates that the manufacturing process is robust enough to always reduce benzene to an acceptable level as stated in ICH guideline Q3C, is there a need to request him to include a skip test in the specifications?

Because class 1 solvents cause unacceptable toxicities it is generally not acceptable to omit a control in the final API when the contaminated solvent is introduced in the final step of the synthesis or purification. A routine test may not be required depending on the scientific justification and data provided.

19. When justifying impurity limits, how can innovator impurity profiles be obtained to compare with?

To justify impurity limits above the ICH qualification thresholds, a comparison with levels present in comparator products may be applied. Comparator products should be purchased from a well-regulated market overseen by a regulator recognized as competent by the authority to which the application is being made. Some authorities may require that the innovator reference product be used for the comparison. It is recommended to use batches near the end of the shelf-life, and the identical validated analytical procedure should be used to generate the impurity levels for the comparison.

20. Can Lab Grade of any excipient be used after testing and complying with the respective Pharmacopoeial monograph, despite it not having any monograph stated on the COA issued by the manufacturer?

As excipients are often present in relatively large quantities in medicines and may contain impurities, they can affect the quality of a FPP and must therefore be adequately controlled. The product dossier should include the specifications from the finished product manufacturer/applicant for all excipients used, and it is the responsibility of the finished product manufacturer/applicant to ensure that the finished product is manufactured using excipients meeting this quality standard. Manufacturers of excipients are recommended to consider relevant pharmacopoeial monographs, when available, as a basis for the development of their internal manufacturer’s specifications.

21. If an API monograph includes a limit for any other impurity which is considered higher than ICH guideline proposed identification limit, is there a need to tighten this acceptance limit for any other impurity to be in line with the ICH guideline identification limit?

In general, the limit for “any other impurity” must be set in accordance with the ICH guideline Q3A identification limit of not more than 0.10%,
regardless of the requirements of the pharmacopeial monograph. In some exceptional cases, the requirements of ICH guideline Q3A do not apply, and different thresholds can be prescribed. In this case, a specific statement can be found in the related substances section of the monograph.

22. **Re-test date is established by the API manufacturer.** Is it possible that for the same API one manufacturer describes a re-test period of 2 years and a second manufacturer describes a re-test period of one year? How should this be addressed in registration dossier?

The stability of an API is dependent on the manufacturing process and storage conditions. The re-test period of an active substance is therefore established by each manufacturer individually. In the case of a product dossier with an API sourced from multiple manufacturers, one complete “3.2.S” section should be provided for the API from one manufacturer, followed by another complete “3.2.S” section for the API from each of the other API manufacturers.

23. **What information should be present in the open part of a Drug Master File (DMF)?**

Please refer to WHO TRS 948, Annex 4; Guidelines on API master file procedure and/or local regulatory guidance.

24. **What are the requirements if the generic manufacturer claims “Full Details” as option for submission of API information?**

Please refer to WHO TRS 970, Annex 4; Guidelines on submission of documentation for a multisource (generic) FPP for the WHO PQ of Medicines Programme: quality part.

25. **Why is the control for Water Content or LOD test not included in the universal tests for active substances described in ICH guideline Q6A?**

Possibly because a test for water content or LOD might not be important for all APIs. It is mainly important in cases where the API is known to be hygroscopic or degraded by moisture or when the API is known to be a stoichiometric hydrate, but generally a control is required for all solid APIs. In some cases, a LOD procedure may be considered adequate; however, a detection procedure that is specific for water (e.g. Karl Fischer titration) is preferred.
SESSION 7 – Control of active pharmaceutical ingredient characteristics

26. Is an identification test based solely on retention time by High Pressure Liquid Chromatography (HPLC) analysis sufficient for an API?

Retention time is the time it takes for a compound to elute from an HPLC column. It is a characteristic of a compound, but it can be affected by factors, such as the mobile phase, the column temperature, and the particle size of the packing material. Thus, retention time alone is not considered sufficient for identification. For the API at least two identification tests based on different principles (physical versus chemical) should be used. In most cases a single near infra-red test would be preferred as it is very specific (fingerprint test) and an HPLC test combined with ultraviolet (UV) diode-array detection.

27. Explain what is meant by a test for polymorphism of an API.

Polymorphism is the ability of a substance to exist in two or more crystal forms due to a different three-dimensional arrangement of molecules. The crystal form of an API can have a significant impact on its properties, such as its dissolution rate, and its stability. It may be important, therefore, to control the polymorphic form of an API to ensure that the API is in the desired crystal form. Some techniques that are used to study polymorphism of an API include the following:

- X-ray powder diffraction (XRPD): XRPD is a technique that uses X-rays to determine the crystal structure of a substance.
- Differential scanning calorimetry (DSC): DSC is a technique that uses heat to measure the thermal properties of a substance.
- Optical microscopy: Optical microscopy is a technique that uses light to magnify and view the crystal structure of a substance.

If an API exists in multiple crystal forms, it is important to determine which crystal form is the most desirable from a dissolution perspective. For low solubility drugs, i.e., drugs that belong to the Biopharmaceutics Classification System (BCS) Class II or IV, the polymorphic form can be of critical importance.

28. Which test should be conducted in a FPP manufacturing unit to verify the polymeric form of an externally sourced API?

The method that is accepted by most regulatory agencies (and WHO) for identification of the polymorphic form of an API is XRPD. Other methods such as DSC or even infrared (IR) for some APIs may be accepted if demonstrated to be sensitive and selective. It is best to consult the regulatory agencies to which an applicant wants to make a submission for product authorization.

29. If the API used in the manufacture of a solid oral dosage form is known to be the most stable polymorph, is it necessary to include a test for polymorphism as part of the stability protocol of the FPP?

According to the US Food and Drug Administration (FDA) Guidance: ANDAs: Pharmaceutical Solid Polymorphism, specifications (release and stability) for polymorphs in drug products are generally not necessary if the most thermodynamically stable polymorphic form is used or if the same form is used in an approved product of the same dosage form (reference listed drug - RLD). This is especially true if the API has a high solubility, i.e., is a BCS class I or III substance. Please refer also to the ICH guideline Q6A and its decision tree.

If the desired form is achieved via conversion that occurs as part of the FPP manufacturing process, control of polymorph identity/content in the FPP specification may be required.

30. Is the most thermodynamically stable polymorph the most active polymorph in all cases?

In most cases, the most thermodynamically stable polymorph of a compound may not necessarily be the most active polymorph (soluble). While thermodynamic stability refers to the lowest energy state of a crystal form at a
given environmental temperature and pressure, pharmacological activity of a polymorph is determined by its solubility and so the extent of the API available as a solubilized form at the site of action.

31. Is it accurate to state that when a metastable form of an API undergoes a transformation to a more stable form, the solubility of the more stable form is always lower than that of the previous metastable form?

Yes, this is true. More stable polymorphs have a more ordered crystal lattice structure, have higher melting points and consequently, are thermodynamically more stable than metastable forms. These factors make it difficult for solvent molecules to enter the crystal lattice resulting in their lower solubility compared to metastable polymorphs of the API, which exist in a higher energy state.

However, to ensure a consistent safety and efficacy profile of the FPP, API lots intended for use in production batches should be consistent and of the same form as the API lot(s) used in clinical/bioequivalence (BE) studies.

32. If an API has a low aqueous solubility (according to the pharmacopoeial definition), but the calculated dose-solubility volume (DSV) indicates that it is soluble across all three physiological pH levels (pH 1.2, 4.5, and 6.8) due to the quantity of API used in the FPP, is it still necessary for the FPP manufacturer to incorporate controls for PSD in the API specifications for the FPP?

An API is considered highly soluble according to WHO guidelines (and BCS solubility criteria) when the highest therapeutic dose is completely soluble in 250 ml or less of aqueous medium over the pH range of 1.2 to 6.8. Thus, if the calculated DSV is less than or equal to 250 ml then the API is highly soluble irrespective of the pharmaceutical or pharmacopoeial definition of solubility. In terms of dissolution, setting criteria for API PSD may, therefore, not be necessary. However, if the PSD is critical to product processibility, product stability, content uniformity (e.g. low dose drug product) or maintaining product appearance, then PSD must be controlled. Please refer to ICH guideline Q6A, decision tree #3.

33. When are polymorphism and PSD of the API considered as stability-indicating parameters?

Polymorphism and PSD are critical attributes of an API that must be controlled at release and during stability when the API has a low solubility according to BCS solubility criteria (i.e., when the DSV > 250 ml at any pH across the physiological pH range) and when the API can either be affected by (change in polymorph) or impact (e.g. powder flow properties and content uniformity by PSD) the manufacturing process. Please refer to ICH guideline Q6A, decision trees #3 (for PSD) and # 4 (for polymorphism) to investigate the need for setting acceptance criteria.

34. Are polymorphism and PSD controls necessary for APIs used in the formulation of solutions and intravenously administered formulations?

If the API is formulated as an aqueous solution for oral or parenteral administration, then neither polymorphism nor PSD are important as the API is not present in its solid-state form because it is dissolved in the solvent. Solid-state properties of the API, such as polymorphism and particle size, therefore, do not need to be controlled in aqueous solutions.

35. Is it mandatory to include d10, d50, and d90 particle size measurements for APIs that belong to Class I of the BCS?

If an API belongs to BCS Class I, it is defined as highly soluble, and hence, will have a high dissolution rate. If it is formulated as a solution in an aqueous dosage form, then PSD does not have to be controlled. If the drug product is a solid dosage form or liquid containing undissolved API particles, but PSD is not critical to product processibility, product stability, content uniformity or maintaining product appearance, then PSD also does not need to be controlled. If any one of these criteria is not satisfied, then acceptance criteria for PSD at d10, d50 and d90 must be set. Please refer to ICH guideline Q6A, decision tree #3.
36. Is there a correlation between particle size determination using different methods, such as laser diffraction versus microscopy?

Particle size determination using different methods, such as laser diffraction and microscopy, can yield different results due to the inherent differences in the principles and limitations of each method. Laser diffraction measures particle size based on the scattering of light by particles, whereas microscopy involves direct imaging and measurement of particles. These different measurement principles can result in variations in the particle size data. Sampling methods and sample preparation techniques are also different. Laser diffraction analyses a disperse sample, while microscopy examines individual particles. These and other factors (instrument capabilities, particle morphology, measuring range, etc.), make it difficult to directly compare results from these two methods.

Therefore, the method for testing future API lots intended for use in production batches of the FPP should be the same as the one used to characterize /test the API lots used in the manufacture of the clinical/BE batches.

37. In terms of PSD, is there always a specification for d90, which indicates that it should not exceed a certain value?

Whether or not to include a one-side d90 value as part of the specification for PSD depends on the regulatory agency to which an application for product registration is submitted. However, for comparison of the particle size of API batches, such as comparing the API from a prospective API supplier to that of the biobatch API and for which PSD is a critical quality attribute, the determination of PSD at all three intervals (d10, d50, and d90) would be important to ensure that the two API batches have similar PSDs.

38. Is it permissible to use multiple batches to establish PSD specifications, or is it strictly determined based on the biobatch?

PSD specifications for the API of a FPP must be based on the API that was used in the manufacture of the biobatch as the biobatch is the one that was shown to be bioequivalent to the reference drug product. This will help ensure that all commercial scale batches of the generic product will exhibit a similar pharmacokinetic and pharmacodynamic profile as the reference product.

39. Which is the best method for determining PSD?

The two methods which are generally accepted by regulatory authorities are sieve analysis and laser diffraction; however, different PSDs are obtained with each unless the particles are spherical. In general, laser diffraction is preferred, but according to USP <786> sieve analysis may be used if at least 80% of particles are shown to have sizes greater than 75 µm.

40. Explain how acceptance criteria for PSD of an insoluble API are determined.

When critical for the performance or manufacturability of the API of the FPP, the PSD is often controlled by establishing one-side limits of d10, d50, and d90 or by both upper and lower limits for each of d10, d50, and d90. Setting the values of the upper and lower limits will largely depend on how significant the effect of the PSD is, which is typically based upon development studies. Acceptance criteria should be set based on the observed range of variation of the biobatch.

It is important to confirm that the used instrument operates correctly. According to the Ph Eur (2.9.31 Particle size analysis by laser light diffraction), unless otherwise specified in the individual API monograph, the response of a laser diffraction instrument is considered to meet the system qualification requirements if the d50 value does not deviate by more than 10 per cent from the range of values of the reference material, i.e., the mean value together with its standard deviation (SD). If optionally the values at the sides of the distribution are evaluated (e.g. d10 and d90), then these values must not deviate by more than 15% from the certified range of values. This is for particle sizes ≥ 10 µm. When the particle sizes are below 10 µm, these values must be doubled.

Thus, if the PSD is to be controlled as a range (with upper and lower limits) for each of d10,
d50, and d90 and the sizes of the reference material were 75 µm for d10, 200 µm for d50 and 410 µm for d90 (i.e., the particle size diameters are ≥ 10 µm), the requirements will be as follows:

\[
\begin{align*}
d_{10} &= 75 \pm 15\% \text{ or } 64–86 \mu m \\
d_{50} &= 200 \pm 10\% \text{ or } 180–220 \mu m \\
d_{90} &= 410 \pm 15\% \text{ or } 349–472 \mu m
\end{align*}
\]

If the maximum particle size diameter is < 10 µm (for example, \(d_{10} = 3 \mu m\); \(d_{50} = 6 \mu m\) and \(d_{90} = 9 \mu m\)), then the ranges change as follows:

\[
\begin{align*}
d_{10} &= 3 \pm 30\% \text{ or } 2.1–3.9 \mu m \\
d_{50} &= 6 \pm 20\% \text{ or } 4.8–7.2 \mu m \\
d_{90} &= 9 \pm 30\% \text{ or } 6.3–11.7 \mu m
\end{align*}
\]

It should be noted that these tolerance limits do not consider the variability of the manufacturing process (including the method of particle size reduction), which can be significant. The calculated ranges may, thus, be too tight. It is best, therefore, to include the manufacturing variability when calculating tolerances. Consult your regulatory agency for guidance, ICH guideline Q6A, WHO TRS No. 970, Annex 4 as well as the cited references.(22, 23)
SESSION 8 – Nitrosamines: risk factors and control strategies

41. Regarding limits for nitrosamines, are the limits calculated based on medicines that are expected to be taken throughout lifetime or are there different limits for medicines that are taken over a short period, i.e., less-than-lifetime (LTL)?

The calculation of the acceptable intake (AI) assumes a lifelong daily administration of the maximum daily dose of the medicinal product and is based on the approach outlined in the International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH) guideline M7(R1). Besides, other principles such as those described in relation to the toxicological evaluation in the assessment report of the Committee for Medicinal Products for Human Use (CHMP) Article 5(3) opinion on nitrosamine impurities in human medicinal products are also considered.

The LTL approach should not be applied to calculate the limits but can only be considered after consultation with competent authorities or WHO PQ as a temporary measure until further measures can be implemented to reduce the contaminant at or below the established limits. This will be decided on a case-by-case basis and only in exceptional circumstances (for example, to avoid a drug shortage of a drug product that is considered medically necessary or medically important).

42. How can a structure-activity relationship (SAR) approach be used to support limits for nitrosamines?

If N-nitrosamines are identified with sufficient substance specific animal carcinogenicity data, the median toxic dose (TD50) should be calculated and used to derive a substance-specific limit for lifetime exposure as recommended in ICH guideline M7(R1).

International regulatory authorities (RA) recently announced a new method to determine the AI of a nitrosamine, the carcinogenic potency categorisation approach (CPCA). Based on an assessment of activating and deactivating structural features present in the molecule, a nitrosamine is placed in one of five categories with each category having its own limit ranging from the class-specific Threshold of theoretical concern (TTC) for N-nitrosamine impurities of 18 ng/day for category 1 and 1500 ng/day for category 4 and 5 (unless other robust data are available that would override this AI).

In addition, the Ames test protocol for nitrosamines has been enhanced to make it more sensitive. A negative result obtained using the enhanced Ames test allows for control of the N-nitrosamine at 1.5 µg/day. For substances testing positive, the AI should be established using the CPCA-approach or, if a surrogate nitrosamine is available with sufficiently robust carcinogenicity data, the TD50 from the surrogate substance can serve as a point of departure for derivation of AI by SAR and read across. Lastly, a negative result in a relevant well-conducted in vivo mutagenicity study can allow control of the N-nitrosamine as a non-mutagenic impurity, i.e., according to ICH guideline Q3A/B limits.

It is recommended to liaise with the relevant competent authorities to verify the acceptability of an approach taken.

43. When a product is at risk for nitrosamine contamination, what will be the best mitigation method. Do we need to remove the product from the market or monitor the levels during dossier evaluation, based on the AI limit?

This is to be decided on a case-by-case basis and should be discussed with the relevant competent authorities.

44. How are nitrosamines detected or quantified?

Testing methods for determination of various nitrosamines in sartans with a tetrazole ring, metformin and ranitidine have already been developed and are available for reference, for example on the websites from of the EDQM, Health Canada and US FDA. These methods may serve as a starting point for the development and validation of analytical methods for testing other active substances and finished products.
45. What is the recommended course of action for FPP manufacturers whose active API suppliers did not consider the risk of nitrosamines? Should the risk assessment and mitigation strategy start from the API processes?

Although there are several risk factors linked to the manufacture and storage of the FPP, and even to GMP, most of the risk factors are linked to the manufacture and storage of the API. None of the risk factors, raw materials, or manufacturing steps can be omitted in the evaluation. Manufacturers of active substances and finished products and their raw material suppliers should provide marketing authorisation holders (MAH) with all information necessary for a comprehensive risk evaluation.

46. Can the shelf life of the FPP impact in the probability of formation of nitrosamine impurities in the FPP? Or is this rather related to nitrosamines in blister packages?

There are different root causes for nitrosamines in the FPP, and the impact of shelf-life on the probability of formation of nitrosamines or the levels of nitrosamines being formed may vary depending on the root cause. Independent of the root cause, it should always be ensured that the AI of any nitrosamine impurity is not exceeded until the end of shelf life of the finished product.

47. Does the nitrosamine risk evaluation need to be included in the MA dossier?

Most authorities require the risk evaluation to be submitted as an attachment to Module 1 with a corresponding reference in Module 3.2 of the MA dossier.

The documentation on the risk evaluation and/or risk assessment of the formation and occurrence of potential nitrosamines in the API is commonly required to be incorporated in the common technical document (CTD) sections 3.2.S.2.6 Manufacturing process development or 3.2.S.3.2 Impurities and/or 3.2.S.4.5 Justification of specification. Other CTD sections may be involved depending on the outcome (e.g. 3.2.S.2.3 Control of Materials, 3.2.S.2.4 Control of Critical Steps and Intermediates).

The documentation on the risk evaluation and/or the risk assessment of the overall formation and occurrence of potential nitrosamines in the FPP should preferably be presented in the CTD section 3.2.P.5.5 Characterisation of impurities or 3.2.P.5.6 Justification of specification. Other CTD sections may be involved depending on the outcome (e.g. 3.2.P.3.4 Control of Critical Steps and Intermediates).

48. Is it acceptable to use lidding foil containing nitrocellulose when the printing ink does not contain amines?

During the blister heat-sealing process, nitrogen oxides can be generated thermally from lidding foil containing nitrocellulose primer. Under these conditions, nitrosamines have been shown to form from low molecular weight amines present either in printing ink or in the finished product and to transfer to the product and/or to the cavity via evaporation and condensation. Thus, not only the printing ink must be considered when evaluating the risk. Whether or not the use of lidding foil containing nitrocellulose is acceptable depends on the specific situation and is the final responsibility of the MAH. They should implement a control strategy regarding N-nitrosamines, which should include current and prospective measures to minimise the risk of generation of/contamination with nitrosamines (e.g. change of manufacturing process, change of raw material quality - including printing ink, introduction of appropriate specifications and development of appropriate methods, and measures on the premises and equipment such as cleaning procedures and environmental monitoring).
49. Is it acceptable to widen the specification of assay test from 95-110% to 115-130% to ensure that the results remain within specification?

Unless there is appropriate justification, the acceptable limit for the API content in the release specifications of the FPP is ± 5% of the label claim (i.e. 95.0–105.0%). Wider release limits may be acceptable for specific APIs and during shelf-life, but this should be adequately justified from a quality, safety, and efficacy point of view. Generally, a shelf-life limit of 90.0-110.0% may be acceptable.

50. There are no acceptance criteria mentioned for fine particle dose in dry powder inhaler capsules. How do we interpret with monograph in this situation?

For generic products, fine particle mass is generally required to be comparable to that of the comparator product.

51. What are the acceptable criteria for visual inspection, 100%, a sample or online, for oral preparations?

The International Pharmacopoeia prescribes that for visual inspection a sample of at least 20 tablets or capsules must be inspected. Other approaches could be justified.

52. For liquid, how do you conduct the test for Uniformity of Mass (UoM).

The procedures described in the International Pharmacopoeia or other officially recognised Pharmacopoeia can be used for determination of UoM of liquid preparations for oral use that are presented as single-dose preparations, or for determination of UoM of doses delivered by the measuring device.

53. When UoM is performed, is it mandatory to perform uniformity of dosage units?

The uniformity of dosage units can be demonstrated by either of 2 methods: content uniformity or mass variation. The content uniformity method may be applied in all cases. The test for mass variations may only be applied when the conditions as described in the applicable pharmacopeial monograph are met. If the test for uniformity of content is prescribed or justified and authorised for all the active substances, the test for UoM is not required.

54. What measures should be taken when a target dosage form (e.g. soft gels) does not have a specific monograph in any of the recognized pharmacopoeias?

When establishing specifications for a new dosage form the general texts of the regionally applicable pharmacopoeia must be taken into consideration, as well as the recommendations for universal and specific tests and criteria described in the ICH guideline Q6A. Please refer to WHO TRS 970, Annex 4.

55. How can you justify the variance between the specifications for mass variation between the various pharmacopoeia and will there be any harmonization in the foreseeable future?

The PDG of the Ph Eur, the JP, and the USP has expressed a commitment to achieving harmonization of the procedures in a timely fashion. Where harmonization has been achieved, an appropriate reference to the harmonized procedure and acceptance criteria is considered acceptable for a specification in all three regions. For example, after harmonization, sterility data generated using the JP procedure, as well as the JP procedure itself and its acceptance criteria, are considered acceptable for registration in all three regions. To signify the harmonized status of these procedures, the pharmacopoeias have agreed to include a statement in their respective texts which indicates that the procedures and acceptance criteria from all three pharmacopoeias are considered equivalent and are, therefore, interchangeable. Please refer to ICH guideline Q6A. Information on pharmacopoeial harmonization and the status for the general texts on the PDG work plan is publicly available, for example on the website of the EDQM.

56. Is it necessary that a product must always have a measuring device in its pack to assure correct dosing?
WHO TRS 970, Annex 4 describes that a device is required to be included with the container-closure system for administration of oral liquids or solids (e.g. solutions, emulsions, suspensions and powders or granules), whenever the package provides for multiple doses.

57. How do we check for extractables?

There is currently no globally standardized guidance available for the assessment and control of extractables and leachables. Local and regional guidance may be consulted, as applicable. For example, the US FDA Guidance to Industry - Container Closure Systems for Packaging Human Drugs and Biologics. A new ICH guideline (Q3E) on the assessment and control of extractables and leachables is under development. Information can be found on the ICH website.

58. Is it mandatory to establish release limits for impurities in the FPPs and how is this calculated?

The concept of different acceptance criteria for release vs. shelf-life for FPP specifications pertains to the establishment of more restrictive criteria for the release of a FPP than are applied to the shelf-life. Examples where this may be applicable include assay and impurity (degradation product) levels. In Japan and the US, this concept may only be applicable to inhouse criteria, and not to the regulatory release criteria. Thus, in these regions, the regulatory acceptance criteria are the same from release throughout shelf-life; however, an applicant may choose to have tighter in-house limits at the time of release to provide increased assurance to the applicant that the product will remain within the regulatory acceptance criterion throughout its shelf-life. In the European Union (EU) there is a regulatory requirement for distinct specifications for release and for shelf-life, where a tighter release criterion is required for assay. Please refer to ICH guideline Q6A.

59. Must the pH of parenteral FPPs be in the range of the pH where the product will be active?

Parenteral formulations should have a target pH as close as possible to physiological pH to prevent potential local reactions and irritation. However, an optimal pH range may be set considering, among other factors, the route of administration, volume of injection, solubility, and stability of the active ingredient in the formulation. Normally a generic product should be of very similar pH range and osmolality as the comparator product.

60. Is maximum volume the same as deliverable volume?

The minimum fill volume for a liquid, semi-solid FPP or a solid FPP (following reconstitution and/or dilution, where applicable) must be chosen in such a way that it is ensured that the required volume can be withdrawn (test for extractable volume) and delivered (test for deliverable mass or volume) to the patient. This implicates the use of an overfill, an excess volume. At the same time the overfill (maximum fill volume) should not introduce a risk of unsafe handling and injection techniques, e.g. use of left-over product for a second dose or pooling of several left-over doses to produce a second dose. The minimum and maximum fill volume of single use containers are generally based on a risk assessment approach.

61. Does in-use stability need to be tested at the end of stability study? Accelerated and long term?

The in-use storage condition and period should ensure the acceptable quality, safety, and efficacy of the FPP when administered to a patient up to the end of its shelf-life. Therefore, the in-use stability studies, when applicable, must be repeated with batches at the beginning and end of shelf life.

62. What is the maximum acceptable in use shelf life for sterile FPPs?

In-use stability studies demonstrate the duration and conditions that physical and chemical stability of the FPP is maintained. From a microbiological point of view, it is preferred that sterile products are used immediately. It is recommended that the in-use storage period and conditions meet, but not exceed, the clinical needs.

63. Excessively short in-use storage periods should be avoided. What is the reference
or best practice for a minimum acceptable period?

In-use stability studies demonstrate the duration and conditions that physical and chemical stability is maintained. From a microbiological point of view, it is preferred that sterile products are used immediately. It is recommended that the in-use storage period and conditions meet but not exceed the clinical needs. To avoid any potential posology errors, it is recommended to prevent differences in shelf-life after opening and following reconstitution and/or dilution, and differences in recommended solvents for reconstitution and/or dilution between products containing the same active substance on the market.

64. What is the general acceptable in-use stability period of oral liquids in multidose containers?

To avoid any potential posology errors, it is generally recommended to apply a shelf-life and storage conditions after opening that are comparable to that of the comparator product.

65. Is it possible that products on the market with the same active substance and dosage form but from different manufacturers have different in-use storage conditions, for example in the refrigerator and at room temperature?

To avoid any potential posology errors, it is generally recommended to apply a shelf-life and storage conditions after opening that are comparable to that of the comparator product. Storage conditions are based on conditions employed during stability studies.

66. In the manufacturing process of sterile FPPs, how is the shelf life of the product calculated when it involves solely the filling of a sterile API into vials?

For products that consist of a single API filled into a container, the initial filling in vials is considered the start of production. It is common practice that the shelf-life is calculated from the date of release of a batch, which should not exceed 30 days from the production date otherwise the date of production should be used for calculation of the shelf-life.

67. The requirements in the International Pharmacopoeia seem generally more relaxed in comparison to the USP or BP. Is there a preference to any specific pharmacopoeia?

WHO TRS 970, Annex 4 refers to ICH guidelines Q6A, Q3A, Q3B, Q3C and officially recognized pharmacopoeias for PQ to establish the specification for an API or FPP. Often, compliance with the regional pharmacopoeia is mandatory.
### SESSION 10 - Setting requirements for dissolution: the importance of the biobatch

**68. When is it necessary to determine the dissolution profile of a drug product?**

A dissolution profile study should be done on the biobatch of each product and each strength.

**69. Is it necessary to conduct comparative dissolution studies for each strength of a solid oral dosage form of a medicine?**

Additional strength biowaivers should be supported by comparative dissolution profiles at three different buffers (normally pH 1.2, 4.5 and 6.8) and, if different, the media and conditions intended for drug product release (quality control - QC media) between the biobatch or biowaiver batch of the reference strength and each additional strength. These profiles may be required also for amendments (variations). Please refer to local regulations and the specific Annotations for WHO Guidelines for Additional Strength Biwaiver Applications for PQ of medicines. (24)

**70. Is it possible to forgo conducting a comparative dissolution study for a lower strength dosage form of the FPP by relying on matrixing and bracketing? The comparative dissolution study on the higher strength dosage form has already been performed and has yielded satisfactory results. The only difference in the formulations of the two dosage strengths is the mass of the API.**

No, not even if the compositions are proportional, which is a requirement for a biwaiver, comparative dissolution testing remains required. If a BE study has been performed between one strength of a generic (biobatch) and the same strength of a comparator or reference product, then to qualify for a biwaiver for any additional strengths of the generic, comparative dissolution studies must be performed for all of them.

**71. Can comparative dissolution be demonstrated by utilizing registered data from a product with an old API source in comparison to a product with a new API source, given the unavailability of the old API source?**

Yes, as long as the profiles were generated under acceptable conditions, including time points.

**72. What is a reference drug product? Is it the branded product used as a reference for conducting a BE study with a biobatch? If not, which product serves as the reference in such studies?**

A RLD, as per US FDA or an acceptable compactor product per WHO, is an approved drug product that will be used as comparator in a BE study with a new generic version to show that they are bioequivalent. Comparator products should be purchased from a well-regulated market with stringent regulatory authority (RA) (WHO requirement). If the recommended comparator cannot be located for purchase from the market of one of the identified countries, the applicant should consult with WHO regarding the sourcing of an acceptable comparator product. A stringent RA is a RA that is: a) a member of the ICH prior to 23 October 2015, namely: the US FDA, the European Commission (EC) and the Ministry of Health, Labour and Welfare of Japan, also represented by the Pharmaceuticals and Medical Devices Agency; or b) an ICH observer prior to 23 October 2015, namely: the European Free Trade Association, as represented by Swissmedic and Health Canada; or c) a RA associated with an ICH member through a legally-binding, mutual recognition agreement prior to 23 October 2015, namely: Australia, Iceland, Liechtenstein and Norway. It is recommended to consult local regulations for selection of a reference product.

**73. Should the dissolution test be included in the specification for a suspension product, even if it is not mentioned in any of the monographs of the WHO recognised pharmacopoeias?**

Yes, it may be appropriate (e.g. insoluble active pharmaceutical ingredient – API) to include
dissolution testing and acceptance criteria for oral suspensions and dry powder products for resuspension. Please refer to ICH guideline Q6A. Examples on how to perform the dissolution study are described in the USP. The acceptance criterion (time point and Q-value) is derived from the dissolution profile in the QC medium (if not available in the pharmacopoeia).

74. If the product monograph does not specify a dissolution test, is it mandatory to conduct a dissolution test for that formulation?

Pharmacopoeial requirements represent the minimum quality requirements for APIs and FPPs. Dissolution tests are generally not required for FPPs formulated as aqueous solutions from which the API is immediately available after administration (oral solutions and injections). However, for solid oral and other complex dosage forms such as tablets, capsules, suspensions, etc., where the API must first go into solution before it can be absorbed after administration, a test for dissolution would be a requirement. Please refer to ICH guideline Q6A.

75. Is 'Delayed release' defined based on the site of release, while 'Extended release' is defined based on the rate of release; or can these terms be used interchangeably?

Delayed release and extended release are distinct terms that are not synonymous, as they refer to different aspects of drug release in pharmaceutical formulations.

Delayed release refers to a specific type of drug delivery system where the release of the API is intentionally delayed until it reaches a specific site in the gastrointestinal tract. The purpose of delayed release is to protect the API from degradation or to target a specific region of the gastrointestinal tract for optimal absorption. Delayed release formulations typically use enteric coatings that resist dissolution in the acidic environment of the stomach and release the API in the higher pH environment of the intestines.

Extended release, on the other hand, refers to a drug delivery system that is designed to provide a prolonged or sustained release of the API over an extended period. The primary goal of extended-release formulations is to maintain a consistent and controlled drug release rate, thereby reducing the frequency of dosing and providing a sustained therapeutic effect. Extended-release formulations employ various mechanisms, such as matrix systems, reservoir systems, or osmotic systems, to control the release of the API over an extended period.

In summary, delayed release focuses on the targeted release of the API at a specific site in the gastrointestinal tract, while extended release refers to the controlled and prolonged release of the API over an extended period.

76. Does the size of a molecule factor into the Biopharmaceutics Classification System (BCS) Class 1 classification, given that high solubility can pose challenges for APIs to cross the phospholipid bilayer?

The BCS is based solely on a) aqueous solubility and b) intestinal permeability of an API. The molecular mass or size of the drug molecules does not directly factor into its BCS classification, although these factors may impact the permeability of the API.

77. Does the definition of solubility include the concept of time?

Equilibrium solubility, as opposed to solubility rate/dissolution, is a time independent attribute.

The definition of dissolution according to the BCS does factor in time. For example, the BCS differentiates between rapidly dissolving and very rapidly dissolving APIs. For a rapidly dissolving API: ≥ 85% dissolves in ≤ 30 minutes. For a very rapidly dissolving API: ≥ 85% dissolves in ≤ 15 minutes. This applies to all three BCS media (normally pH 1.2, 4.5 and 6.8). For more information see the WHO solubility study guideline (TRS 1003, Annex 6, Appendix 2).

78. When conducting QC batch release, the single-point dissolution or multiple-point tests pass, but the dissolution profiles of the current batch are not comparable to the dissolution profiles of the biobatch, reference batch, or previous batches.
**What are the next steps that should be taken to investigate and resolve this issue?**

Firstly, the dissolution acceptance criteria are likely not strict enough. So, that is the reason why the acceptance criteria (time point, Q-value) should be strictly in accordance with the biobatch profile. Then, profile comparisons against the biobatch profile are only required in certain circumstances, such as for process validation and for certain amendments (variations). Otherwise only the QC-test is required (which is based on the biobatch).

The root cause for any findings of non-similar dissolution profiles (when compared to the biobatch profile) should be investigated and notified to the regulator.

**79. What are the reasons for the differences in the definitions of high solubility between WHO, the US FDA, and other regulatory agencies?**

Since the ICH Harmonized Guideline M9 has been adopted by regulatory authorities such as the US FDA and the European Medicines Agency (EMA), the definition of high solubility has also been harmonized. The definition employed in M9 is consistent with the definition employed in WHO guidelines.

Previous differences may have existed because of differences in scientific perspectives, regulatory frameworks, and specific agency objectives. What is important to appreciate is that regulatory agencies are independent of each other, and hence, may have their own specific requirements. The purpose of the ICH process is to harmonise between these agencies.

**80. Do we conduct bioequivalence (BE) tests for injectable products?**

If the proposed formulation is a simple aqueous solution for injection, then a BE study is not required as the API is immediately available to exert its therapeutic effect unless the product contains other excipients not present in the comparator product that could affect the availability of the API. A BE study will in principle be required if the product is a complex formulation such as a suspension for injection. In some situations, a biowaiver could be applied, however in addition to in vitro release data this often requires additional in vitro comparisons (for example, physicochemical characteristics, PSD, and particle morphology) or in vivo data.

**81. Why are decimals not typically used in reporting dissolution results?**

The precision of the results is not typically high enough to warrant the use of decimals. This is because there are several variables that can affect the rate of dissolution, and which restrict the accuracy of results to whole numbers. Examples of the variables in question include:

- De-aeration (heat) + 1% (reduction in volume)
- Volume measurement ± 1% (as per pharmacopoeias)
- Evaporation during test + 1% (as per pharmacopoeias)
- Excipient interference + 2% (UV detection, allowed by validation)
- Analytical variance ± 2%

**82. How are dissolution results typically expressed, as a range of values or the average of six values?**

As the average value AND range (or individual values instead of range). The average value provides a representative measure of the dissolution behaviour, considering the variability inherent in the experimental process. Reporting the average value helps to summarize the overall dissolution performance of a product under specific test conditions. The range is to get the lowest value, necessary to see if Q+5 is met in the acceptance criteria.

**83. At which time-point(s) are optimal for single-point and multi-point dissolution testing, respectively?**

For single point dissolution testing, e.g. for QC, there can only be the one, and that time point should be derived from the biobatch profile – and it should be tight. For multipoint dissolution testing, see for instance the PQT/MED-specific Annotations for WHO Guidelines for Additional Strength Biowaiver Applications published on
the PQ of Medicines webpage for selection of the timepoints. (24)

84. If the comparator product shows a slower release (e.g. 75% in 15 minutes) compared to the release of the BE batch of the test product (e.g. 85% at 15 minutes), but the products are still deemed bioequivalent, would it be acceptable to set the dissolution specification aligned with the release of the comparator product (e.g. Q=75% in 15 minutes)?

No. It should always be set according to the profile of the test product, i.e., the BE batch to be product specific.

85. Will a change in agitation speed (e.g., from 75 rpm to 50 rpm) or a change in apparatus (from paddle to basket) have any impact on the stability of a FPP? Assume that the stability study has already been completed.

It will not influence the FPP stability, but the dissolution data will not be comparable to the previously generated stability data.

86. If a tablet contains two different APIs with different rates of solubility, can the dissolution specification be adjusted to only one time point?

No. Each API should have its own specific acceptance criteria. If there is a difference in dissolution rates, different time points may be necessary.

87. If we conducted a comparative dissolution profile study with the RLD using a dissolution method available in the pharmacopoeia, can we utilize the same dissolution method for the comparative dissolution in all dissolution media, such as 0.1N HCl, buffer 4.5, and buffer 6.8?

Yes, if the pharmacopoeial method has acceptable paddle speed (rpm) and volume and the suitability of the method has been confirmed.

88. Is the use of a 5% sodium lauryl sulphate (SLS) solution acceptable as a dissolution medium for QC?

Yes. The quantity of any solubility-enhancing substance must be justified. A concentration of 5% SLS is very high and will have to be based on solubility or dissolution profile data of the API. That is, evidence must be provided that a lower percentage concentration will not be usable. For QC methods the lowest concentration that provide sink conditions may be selected although achieving sink conditions is not always necessary.

89. Is a biobatch the same as a pilot or trial batch, or are there distinct differences between them? If they are different, please provide an explanation.

There are differences between a biobatch, clinical trial (CT) batch and pilot batch, which are based on the purpose of the batch. A biobatch is the batch used to establish BE or similarity to the comparator product as determined in BE or biowaiver studies, respectively.

A pilot scale batch is a batch of an API or FPP manufactured by a procedure fully representative of and simulating that to be applied to a full production-scale batch. For example, for solid oral dosage forms, a pilot scale is generally, at a minimum, one-tenth that of a full production scale or 100,000 tablets or capsules, whichever is the larger, unless otherwise adequately justified.

A CT batch is a batch of a FPP that is used in a controlled setting, i.e., a CT that is conducted in humans to determine the safety and efficacy of an FPP. Please refer to WHO TRS 981 - Annex 3: WHO guidelines on variations to a prequalified product.

90. If a monograph for a product already specifies limits for dissolution, is it necessary to establish your own QC dissolution limits?

Yes. The test methods and limits should be product specific and based on the dissolution of the biobatch.

91. If a monograph publishes a new method for dissolution, should we test the dissolution profile of our new batch using the method we used for the
biobatch, or can we use the new method in the monograph?

No. Continue with the old method, since that has been used for the biobatch data generation (also refer to the previous questions).

92. What are the critical areas of concern for the proposed dissolution criteria?

It is important to ensure that the dissolution method is acceptable and that the QC acceptance criteria are derived from the biobatch profile and are sufficiently tight to discriminate any production batch that may be exhibiting a different dissolution behaviour from the biobatch and other production batches.

93. For very rapidly/fast dissolving FPPs, the acceptance criteria typically requires that the drug be released equal to or greater than 85% within 15 minutes. However, some regulatory agencies also allow for the use of a V-value when setting the acceptance criteria. The V-value is calculated by subtracting 10% from the release concentration of the lowest time point. However, considering that the example value of 85% already meets the acceptance criteria for a rapidly dissolving drug product, does this approach contradict the requirement of 85% release within 15 minutes as defined by regulatory guidelines?

There is no contradiction with this approach. If the dissolution rate is not 85% in 15 minutes, then the V-value is determined.

94. If a product fails the disintegration test but passes the dissolution test, is it possible to release the product for distribution and sale?

This is not expected, unless the dissolution limits are far too loose, in which case it should be investigated if this happens. The dissolution test is the principal test unless the product is a dispersible tablet in which case the disintegration test is also a required test. Regardless, the failure to meet the set disintegration criteria should be investigated including review of data collected during in process testing. For dispersible tablets, the batch should not be released. In other cases, the relevant regulatory agency should be contacted before batch release.

95. If a commercial batch is released at stage S3, while the reference biobatch has a dissolution profile evaluated at stage S1, what are the implications for the release of this commercial batch?

The batch can be released in principle; however, investigation of the observation as an out-of-trend value should be considered.

96. Do we need to conduct an out-of-specification (OOS) investigation if results do not meet acceptance criteria for S1 or S2 stage?

No. You first do the testing at S3 stage and then an OOS investigation is conducted. An S3 pass should be considered for investigation as an out-of-trend value.
Bridging the gap between design validation and quality specifications for in vitro diagnostics

Day 5

Photo credits: © WHO/Andy Craggs, Rapid diagnostic test.
SESSION 11 - Designing, implementing, and applying design controls

97. What is the meaning of IOVV?

IOVV stands for Input Output Verification Validation. This term IOVV goes along with the traceability matrix required throughout the design and development cycle of the medical device. The IOVV matrix is a valuable tool that provides a high-level view of the connections that exist between User Needs & Specifications and how these will be validated and verified, respectively. This very common tool used by medical device companies demonstrates compliance to US Food and Drug Administration (FDA) 21 Code of Federal Regulations (CFR) Part 820.30, Design Controls, as well as International Organization for Standardization (ISO) 13485:2016, Section 7.3, Design and Development.

98. What is IVD?

IVD stands for in vitro diagnostic. IVD is a medical device intended by the manufacturer for the in-vitro examination of specimens derived from the human body solely or principally to provide information for diagnostic, monitoring, or compatibility purposes. IVD medical devices can be used alone or in combination with instruments or apparatus or other articles such as software. It usually includes reagents, calibrators, control materials, specimen receptacles. In vitro tests may be done in laboratories, health care facilities or even in the home. The tests themselves can be performed either manually or on a variety of instruments ranging from small, handheld tests to complex laboratory instruments. They allow doctors to diagnose patients effectively and work to provide appropriate treatments.

99. How to differentiate diagnostic use and screening use?

Screenings and diagnostic tests initially differ based on their intended users and whether the intended population for testing is symptomatic or not.

A screening test is often used to detect potential health disorders or diseases in people who do not have symptoms of disease. Screening tests may enable early detection, and more effective and successful treatments. Screening tests also allow for monitoring of potential risks for certain diseases.

A diagnostic test is often used to investigate a specific concern to determine the presence or absence of disease. It is often used to diagnose conditions in patients that display symptoms or asymptomatic patients with a positive screening test.

100. What is the objective of asking the questions "Who - On Whom - What - Where - When" during design control?

The objective of asking the questions "Who - On Whom - What - Where - When" is to help define the intended use of the device. The intended use consists of the objective intent of the manufacturer regarding the use of a product, process or service as reflected in the specifications, instructions and information provided by the manufacturer. The intended use should be defined at the early stage of the device design and development process because Design Controls will be developed and applied in a way that the medical device has been proven to meet the user needs and product’s intended use.

101. Why regulatory expectation is not considered in design control?

The objective of design controls (DC) is to require that manufacturers follow a methodologically-sound process to develop a medical device, with the intent of improving the probability that the device will reach an acceptable level of efficacy and safety. DC should be integrated into the Quality Management System (QMS). ISO 13485 is the main QMS standard for medical devices. While compliance of the manufacturer’s QMS and manufacturing practices with applicable international standards may be required in certain regions, it may not be in other regions. There are various quality regulations and standards for different regulatory agencies.
SESSION 12 - Design control challenges

102. What is benchtop testing?

Bench testing, or benchtop testing, is a type of testing designed to rule out performance related, mechanical or design flaws in a medical device, including in vitro diagnostic (IVD) devices. Performance bench testing of medical devices also evaluates their endurance and capability to perform with the same efficiency under different forms of load. It is a crucial step in the early device design process.

103. Are instructions for use (IFU) equivalent to a standard operating procedure (SOP)?

IFU and SOP are not the same. The IFU is a critical part of an IVD medical device (IVD); it is expected to effectively communicate the product information to the intended user and ensure the safe and proper use of the IVD. The IFU communicates the purpose for which the IVD should be used, who should use it, how the IVD works, what types of specimens it should be used with, what materials and reagents are needed to perform the IVD, how to perform the test, how to interpret the test result, the limitations of the test, warnings and precautions that need to be considered when using the IVD, and evidence to support IVD performance claims. It therefore communicates all that the user needs to know to make good clinical use of the IVD.

SOPs are the documented practices and processes that a company has in place to ensure services and/or products are delivered consistently every time while meeting minimum quality standards. In practical terms, most SOPs are written as a step-by-step series of operating instructions that can show employees what they need to do to accomplish a given task. SOPs are designed to ensure an efficient, quality output on a consistent basis, regardless of who follows them.

104. Is ISO 13485 certification required for WHO prequalification (PQ) assessment of IVDs?

ISO 13485 certification is not a requirement of the WHO PQ assessment of IVDs. However, compliance with ISO 13485 requirements is still expected during both product dossier review and inspection of manufacturing site(s). The manufacturer shall provide procedures that document monitoring, measurement, analysis, and improvement of the product to ensure the conformity of the product and QMS, and to maintain the effectiveness of the QMS, including those relevant to control of nonconforming goods/processes and complaint handling and vigilance. A WHO inspection of the manufacturing site(s) is part of a PQ assessment, is normally product specific, and is based on the principles outlined in ISO 19011:2018 “Guidelines for auditing management systems.” The aim of the inspection of manufacturing site(s) is to assess compliance of the manufacturer’s QMS and manufacturing practices with applicable international standards, such as ISO 13485:2016 “Medical devices - Quality management systems - Requirements for regulatory purposes” and others.

105. Is the device history file (DHF) different than the product dossier (PD)?

DHF and product dossier are not the same. DHF consists of the documentation that an organization shall maintain for each medical device type or medical device family. The DHF shall include, or reference, records generated to demonstrate conformity to the requirements for design and development and records for design and development changes. The DHF is a requirement of ISO 13485:2016 - section 7.3 - subsection 7.3.10 as well as of the US FDA 21 CFR 820.30§(j). The PD is assembled with the purpose of seeking assessment of the performance and safety of the IVD for regulatory approval or for WHO PQ. There are many terms used internationally to describe a product dossier. These terms include standard technical documentation, technical file, summary technical documentation, product summary file, product master file and others. For the purposes of WHO PQ of IVDs, WHO uses the term product dossier. The product dossier is a selection of records and documents from the entire collection of technical records and documents that a manufacturer holds for a product.
Manufacturers compile a product dossier from their existing technical documentation to provide evidence that an IVD conforms to the internationally recognized set of quality, safety and performance principles as described in the applicable regulations and standards. Evidence will take the form for example, of results of testing, certifications, SOPs, systems, and any other documentation necessary to support quality, safety, and performance.
SESSION 13 - Design verification and validation

106. What is the applicability of design review and design verification in design controls?

To understand when design reviews and design verification processes are applied, it is important to understand and differentiate the two terms.

Design and development review is described under clause 7.3.5 of ISO 13485:2015. Design and development review is a review process which involves a systematic evaluation of an IVD design's ability of the results to meet expected requirements, to identify any problem and propose necessary actions. Design reviews are checkpoints during an IVD development to ensure the product design is safe, effective, and progressing. Design reviews are moments in time during design and development that ensure the device being developed is on track and in alignment with expectations.

Design and development review is an effective tool to find issues on the medical device design and help to fix it before the product reaches the intended users. Design review is not only manufacturer’s process, but also a regulatory requirement. During a design review, it is likely to find issues or even nonconformities that need to be addressed. Records of the necessary actions that were identified must also be maintained.

Design and development verification is described under clause 7.3.6 of ISO 13485:2015. Design and development verification: means a confirmation through the provision of objective evidence that specified requirements have been fulfilled. During design verification, verification of an IVD design outputs to meet design inputs are performed. In other words, it is a confirmation that a manufacturer has designed an IVD right.

- At what point design verification is applied: during design and development process, user needs are first defined followed by developing design inputs, i.e. manufacturer define what is to be designed. Going through the design process to bring about design results. The result of the design process is the latest iteration of an IVD product which is now termed as design output. Verification happens at this stage to ensure the design output matches the design input. Design verification performed by testing and conducting visual inspections and analysis.

- The importance of design verification is to demonstrate a design output (i.e. the designed IVD product) meets the design inputs (which is derived from the process of defining user needs).

107. Do the notified bodies keep manufacturer’s sensitive information confidential?

The role of a notified body like any other regulatory authority (RA) is to assess the conformity of medical devices and IVDs before being placed on the market. It is normally a strict requirement that notified bodies and regulators make adequate arrangements to ensure the confidentiality of the information obtained during conformity assessment and inspection of the manufacturing site is attained.

108. What are the methods of inspection, and which are the inspection tools for the different validation procedures?

Inspection processes for medical devices and IVDs manufacturers are performed to assess the facility’s compliance with the appropriate regulatory requirements and relevant regulations. WHO guidance document of the WHO PQ of Diagnostics (PQDx) programme PQDx_014 v4 8 September 2017 Information for Manufacturers on the Inspection of manufacturing Sites, provides to medical devices manufacturers with information on the inspection of manufacturing site(s) of product(s) undergoing WHO PQ assessment, including the assessment of their QMS.

Three types of inspections applicable to medical devices inspections are; an initial inspection which is conducted to all new products manufacturer before approval to enter the market, re-inspection which is conducted to an approved IVD manufacturer to ensure ongoing
compliance with regulatory requirements and standards and special inspection which is conducted to verify effectiveness of the manufacturer to implementation of corrective actions to prevent the recurrence of nonconformities, prior to PQ or after a company has made changes to address any compliance issues found during a previous inspection, or if there is serious concerns raised about the ongoing quality of the IVD, suspended production activities and then recommended.

The standards applied for PQ assessment include the quality management standards ISO 13485:2003: Medical devices — Quality management systems and ISO 13485:2016: Requirements for regulatory purposes, and relevant international standards and guidelines produced by the former Global Harmonization Task Force and the International Medical Device Regulators Forum (IMDRF). As a general rule, the criteria for inspection of manufacturing site(s) are product specific.

According to ISO 13485:2016 clause 7.5.6, the organization shall validate any processes for production and service provision where the resulting output cannot be or is not verified by subsequent monitoring or measurement and, therefore, deficiencies become apparent only after the product is in use or the service has been delivered. Validation shall demonstrate the ability of these processes to achieve planned results consistently. The organization shall document procedures for validation of processes, including a) defined criteria for review and approval of the processes; b) equipment qualification and qualification of personnel; c) use of specific methods, procedures, and acceptance criteria; d) as appropriate, statistical techniques with rationale for sample sizes; e) requirements for records f) revalidation, including criteria for revalidation; g) approval of changes to the processes.

Process validation is the collection and evaluation of data throughout a process, from the process design stage through production, with the intent of establishing proof that a process will consistently deliver quality products. It is a requirement to perform process validation for IVDs before it is introduced on the market to ensure that the products are reliably and repeatably produced to satisfy requirements of safety and user needs, technical, and regulatory requirements.

Process validations for IVDs manufacturing is important since it reveals processing deficiencies. Post-production inspection or testing are usually applied to provide verification that a produced IVD meets the requirement, but this is not practical since it requires testing of every IVD device produced. In addition, testing each product never reveal all the possible variations within individual products and the testing itself could potentially destroy the product.

Important process validation methods which IVD manufacturer should perform includes.

- prospective validation which is performed prior to process implementation, retrospective validation which involves the use of historical data to create a retrospective validation for facilities, processes, and process controls that have not already undergone a formally documented validation process, concurrent validation which employs information generated during the actual process to establish evidence that a facility and process do what they claim to do and revalidation which involves repeating the original validation in its entirety or in part, and includes a thorough review of existing performance data.

109. What are the differences between testing, inspection and analysis used in design validation?

Design validation testing means performing specific tests on an IVD to demonstrate it works for end user according to its specific intended use. For example, an IVD which is constituted by software (software as a medical device - SAMD), design validation will include testing methods to demonstration that the software requirements (that include user needs) are fulfilled. Other testing activities related to the safety of an IVD according to its intended use, for example, biocompatibility evaluation, electrical safety and electromagnetic compatibility, in case of active IVDs, packaging validation, usability, clinical investigation must be considered.
Note: Design validation testing activities must be carried out on the finished IVD product. ISO 13485:2016 states that design validation activities should be conducted on representative products, which may include initial production units, batches, or equivalent samples. Therefore, the rationale for selection of the specific IVD for validation testing need to be documented.

The design validation inspection employs the examination of the validation activities, process, component, or subsystem of an IVD or a finished product. It is generally necessary to perform inspection at every stage of design validation process to ensure that all the design specifications, user needs and regulatory requirements are met.

The design validation analysis provides a complete overview of validation outcomes, including intended user test results for each type of testing activity, and any gap discovered throughout the validation process from the expected requirements. Analysis of activities are an important tool to compare the actual IVD performance to an expected performance which in turn helps to determine whether validation requirements and expectations are being met and discover unacceptable risks associated with the designed IVD product. The importance of validation analysis is to provide a documented output in the analysis report which will include, where necessary, recommendations for further action such as repeating a validation, amending a validation protocol and performance of a new validation, or may make recommendations relating to staff training, record keeping, equipment stability, etc.

For example, some of the IVD designs may be validated by comparing with similar IVDs performing similar functions. Validation process will be relevant for validating configuration changes for the proposed design, or standard designs that are to be incorporated in a new product or application. Tests will be performed on the final design that validates the ability of an IVD to operate as per the specified design. Therefore, test plan, execution, and results should be documented and maintained as a part of design records.

110. Is it required to use 3 consecutive batches in validation process, or this can be adjusted based on risk assessment?

The actual number of batches for process validation depends on the risk involved and therefore IVD manufacturers must decide the number of batches required for manufacturing to support the statistical data. For confirmation of reproducibility, we should have enough batches to compare batch to batch variations. Usage of three consecutive batches during validation: The basic concept regarding validation studies has been always to include 3 batches and if the results for all the 3 batches meet the requirements, then conclusion is made that the method is able to reliably and repeatably produce the product and therefore commercial scale production can be initiated. Traditional reason for selecting three batches may be explained as during manufacturing of 1st batch quality may be accidently built in a product and during manufacturing of 2nd batch quality may be regular. If the same quality is achieved in the 3rd batch, it means that the results are consistent and are reproducible. Product compliance to three batches gives assurance that a validated process is valid. This is more linked to specific role of design validation activities which intends to provide documented evidence that ensures that designed product or process or procedure consistently produces results with the same quality standards.

Note: In other words, if the desired quality standards in the first batch is obtained it may be incident and the same quality in the second batch is maintained it may be coincident and if the same quality in the third batch is maintained it is therefore called consistent.

Additionally, ISO 13485:2016 states that design validation should be conducted on representative products, which may include initial production units, batches, or equivalent samples. The rationale behind choosing the specific product and IVD batch/lot for validation purposes must be duly documented.

111. What are the clean room requirements for different classes of medical devices?
Maintenance of cleanrooms in medical devices manufacturing is important to ensure that medical devices and IVDs are safe to the end user. ISO 13485:2016 clause 6.4 Work environment and contamination control requires medical devices manufacturer to maintain environmental cleanliness and control contamination in the development of medical devices to protect patient safety. Similarly, the ISO 14644, outlines the required standards for air quality in manufacturing environments.

Depending on the risk class of a medical device, manufacturing takes place in cleanrooms ranging from Class 100 to Class 100,000. Medical device packaging takes place in Class 10,000 or Class 100,000 cleanrooms with a Class 10,000 gowning room.

**112. How is the validation and verification of IVDs related to in quality control (QC) kits used in biopharma?**

Validation of QC kits used in biopharmaceuticals and other equipment normally involves design qualification (DQ), installation qualification (IQ), operational qualification (OQ), and performance qualification (PQ). The purpose of validation for these kits is to ensure that the process and the products manufactured can perform reliably and precisely for its intended purpose.

On the other hand, IVDs are tests done on samples such as blood or tissue that have been taken from the human body. In vitro diagnostics are used to detect diseases or other conditions and can be used to monitor a person’s overall health to help cure, treat, or prevent diseases. Some tests are used in laboratory or other health professional settings and other tests are for consumers to use at home.

Design validation for in vitro diagnostics is a testing of a designed IVD and the process to prove that an IVD meet end user and regulatory requirements. Design validation activities for an IVD must involve clinical evaluation. This means that the end-user(s) should be involved, and the device should be tested either under simulated use or actual use.

In conclusion, test kits used for QC activities in biopharmaceutical industry are not in vitro diagnostics based on the above definition and therefore, design control requirements are not applicable to these products.

**113. Is design validation required to be carried out both before and after design transfer?**

According to ISO 13485:2016 clause 7.3.7, “Design validation shall be conducted on representative product. Representative product includes initial production units, batches or their equivalents. The rationale for the choice of product used for validation shall be recorded. A medical device used for clinical evaluation or performance evaluation is not considered to be released for use to the customer”.

Design validation focuses on the market specification to confirm it has been met. This includes user needs and regulatory requirements. To do so, the product is put to test in the expected or stimulated environment to understand its performance. This means the medical devices used for validation must be built in the production environment, using drawings and specifications (i.e., design outputs) by production personnel. Validation activities includes the design validation and process validation.

During design validation clinical evaluation of an IVD involving end-user(s) and the device should be tested either under simulated use or actual use under the specific, intended environmental conditions. This includes any changing conditions, such as devices that must remain operable as patients move from room to room. Additionally, design validation activities involve IVD packaging and labelling and IFU. Once validation is completed and the product becomes ready for production, the manufacturer must create a plan to execute proper mass manufacturing. This next step is the design transfer.

Therefore, Design validation activities are performed before design transfer. If there is a change to an IVD design after release and the change affect its performance, function, usability, and safety, then design validation may be required.
114. What are analytical and clinical performance characteristics of IVDs considered during performance evaluation?

Analytical and clinical studies must be based on a detailed and comprehensive study protocol. The specific content of a study protocol for these investigations depends on the characteristic expected to be validated, which in turn depends on the risk management and planning that has been undertaken during IVD design. However, most analytical and clinical performance studies share several common features. The verification and validation performance characteristics are based on the intended use of the investigated IVD and whether an IVD is a qualitative or quantitative. The scope of the performance characteristic and its verification and validation criteria must be defined and documented early in the design process.

Clear understanding of the specific characteristic for the method in use requires well-planned method verification and validation studies. Within this context, carefully designed test methods will provide information to satisfy more than one of the expected parameters. Apart from IVD performance characteristics, it is also important to consider other protocol requirements for verification and validation studies such as study rationale, ethical considerations, study objectives, study methods etc. Some examples of analytical and clinical performance characteristic that should be considered in validation and verification investigations includes: 1. Analytical performance characteristics: such as accuracy, trueness and bias, precision (repeatability and reproducibility), analytical sensitivity (limit of detection, detection of variants), analytical specificity (interference and cross-reactivity), measuring range of the assay, validation of assay reading time, traceability of calibrators and control materials, validation of assay procedure, IVD and reagent stability. 2. Clinical performance characteristics: such as clinical sensitivity, clinical specificity, positive predictive value, negative predictive value, and end-user verification of labelling and IFU (self-testing)
Session 14 - Design transfer and changes

115. What are the requirements for an authorized representative in the European Union (EU) or the European Economic Area (EEA)?

It is a mandatory requirement that every medical device and IVD manufacturer in EU/EEA to have a named person responsible for regulatory compliance according to medical device regulation (MDR)/in vitro diagnostic regulation (IVDR) Article 15.

All manufacturers outside the EU/EEA need to have an authorized representative whose business place is in one of the EU/EEA member states (MS). EU regulations stipulate list of roles for the authorized representative at minimum, verifying that the EU declaration of conformity and the technical documentation have been drawn up and, where applicable, that an appropriate conformity assessment procedure has been carried out by the manufacturer. Also keeping copies of required documents and cooperate with authorities on request. Additionally, the authorized representative is legally liable for defective devices jointly and severally with the manufacturer. If a manufacturer is outside the EU/EEA and not planning to place an IVD medical device on the EU market this requirement is not applicable.
Session 15 - Understanding performance evaluation of in vitro diagnostics

116. For personal use IVDs, there is uncertainty that the end users use these IVDs appropriately and some of them may even not know that the device is no longer providing accurate results. Is there any approach that manufacturers can use to monitor the provided IVDs as part of may be continuous post marketing surveillance (PMS)?

Personal use IVDs or home-use tests are also known as self-testing IVDs. These are IVDs intended for use by a lay user who is responsible for collecting the data or specimen, by themselves and on themselves, relying solely on the instructions provided by the manufacturer. This can also include performing the test and interpreting the results by themselves and on themselves outside the laboratory setting. Self-testing for infectious diseases has been well established and increasing in both well-resourced and low- and middle-income countries (LMICs), supporting equitable access to testing for all. Self-testing IVDs are required to follow requirements of quality, safety, and performance. In addition, most mature regulatory systems require manufacturers to apply the principles of risk management as described in ISO 14971. Manufacturers should apply these principles by assessing potential risks to the device’s safety and performance when it is used as intended and where possible, eradicate these risks via adaptions to design. The effectiveness of these measures should be verified. For IVDs, this would usually include the requirement to have features that indicate that the test is performing properly. For instance, such design features in a rapid test includes the incorporation of a control strip, that reacts in a manner to inform the user that the test has performed as intended. The IFU will provide information to the user of what to do if the control does not work as expected.

It is also a manufacturer’s responsibility to monitor IVDs safety and performance once market authorization has been obtained. For self-tests, this is not simple, but can include actions by the manufacturer such as encouraging users to report potential problems to their distributor, to have the distributor obliged to report back information from users.

WHO has issued updated guidance on post-market and market surveillance of IVDs that encourage users of IVDs to monitor IVDs and report any concerns related to quality, safety, or performance to the manufacturer via their local economic operator (supplier, agent, and authorized representative). Also, manufacturers of IVDs for self-test eligible for WHO PQ assessment are obliged to report adverse events (AE) and subsequent investigations to WHO for review, including an annual report of all incidents to WHO for risk assessment purposes.

117. If environmental conditions can impact IVD significantly, should an IVD imported from other country (e.g. from Western to Asian/Tropical countries) need to go for another performance study factoring these environmental factors?

IVD kits may contain reagents such as proteins, enzymes, fluorescent dyes, buffers, positive and negative controls, and other sensitive materials that react differently to storage conditions. If any one of these reagents change at any time, may affects the performance of the IVD product which may produce a false non-reactive or reactive result which in turn would affect treatment plans that could compromise patient health. For some regulatory regimes, e.g. the EU IVD Regulation, it is clearly the responsibility of the manufacturer to ensure the transport conditions of their IVD through to the end user will ensure the test works as intended. Through their legal relationships with importers and distributors, they must have plans in place to ensure the ongoing stability of IVD kits and its reagents during transportation to ensure that performance characteristics of the product are maintained during shipping from the manufacturer to the end user. However, this regulatory requirement is not universal. In the case of the EU IVDR, this obligation can only be legally enforced when the product is distributed in the EU. As such, in other jurisdictions, the ongoing stability cannot necessarily be assured to the same level.

Good laboratory practice (GLP) as described in standards such as ISO 15189 requires the
laboratory to ensure that the tests that they use are fit for purpose. Laboratories are required to undertake quality assurance activities commensurate to the risk of the tests failing. If transport and storage stability cannot be assured (which is almost always the case) it is the responsibility of the laboratory to undertake quality assurance (QA) activities to ensure performance of a kit or lot before testing patient samples. This may include testing a well selected panel of specimens (large or small, depending on multiple factors). However, this infers that the responsibility lies very much on the laboratories. Manufacturers, under Section 8 of ISO 13485 are required to monitor the ongoing safety and performance of their tests. Thus, the maintenance of test performance will always remain a shared responsibility, but until regulatory systems are similar to that in the EU where distribution within the Union is the clear responsibility of the manufacturer, then it is important for laboratories to be alert to the possibility of poor performance due to suboptimal transport conditions. However, at the end of the day, the labs should monitor the quality and cease use if it is consistently poor or if the manufacturer is showing no intention to respond to their concerns. The laboratory should also inform their local regulatory agency of the issues.

Manufacturers are highly recommended to follow WHO Technical Guidance Series (TGS) - 2 Establishing stability of IVDs, which describes the requirements for assigning shelf life, transport conditions, and storage conditions for IVDs and the reagents in the kit intended to undergo PQ assessment. It also provides much practical advice on how to achieve this. The document also addresses the process for understanding and defining stability once the reagents are opened.

Regardless of the legal requirements, good distribution practice requires importers and distributors to ensure that, while a device is under their responsibility, storage or transport conditions do not jeopardize its compliance with the general safety and performance requirements set out in the applicable regulations and comply with the conditions set by the manufacturer, where available. If importers have reason to believe that the environmental conditions are significantly impacting the performance of the IVD that they have placed on the market and potentially presenting risks to the users, they shall immediately inform the manufacturer and its authorized representative. The same applies to any situation where the IVD is not in conformity with the regulations. Where the device presents a serious risk, they shall also immediately inform the competent authorities of the MS in which they made the device available. Importers shall co-operate with the manufacturer, the manufacturer's authorized representative and the competent authorities to ensure that the necessary corrective action to bring that device into conformity is taken.

118. To what extent is post-market surveillance (PMS) required for IVDs?

ISO 13485 is the standard that describes a quality system to be employed for the manufacturing of medical devices and IVDs. It is generally acknowledged as the standard for good manufacturing practice (GMP) in all regulatory agencies (RA). Section 8 “Measurement, analysis and improvement” of ISO 13485 describes the obligations of the manufacturer to monitor the conformity of the device during its manufacturing processes as well as post-production. The actions of the manufacturer should be planned, analysed and monitored. The outcome of the analysis may be as severe as requiring a manufacturer to recall product from the market. Given that IVDs usually only cause indirect harm (i.e., they rarely do not in themselves harm patients, but wrong or delayed results usually are the cause of harm) it is even more important for an IVD manufacturer to consider implementing a strong PMS plan, that incorporates not only reactive elements but also pro-active elements.

Several different approaches to PMS are possible. The PMS requirements may vary depending on the regulations applicable to different jurisdictions or national regulatory authorities (NRA). Receiving and acting upon user or other feedback is the most basic form of PMS that must always be performed by the manufacturer, irrespective of their resources. Administrative feedback is not typically linked to safety, quality, or performance issues. However, the investigation of administrative feedback might reveal potential issues with quality, safety and/or performance of the product, and should be considered as
technical in nature. As such, timely periodic investigation and analysis of administrative feedback is strongly encouraged. Technical feedback is in nature affecting the safety, quality, or performance of a medical device. The inputs of the PMS activities should feed directly into the risk management assessment and if there is a decline in the benefit risk ratio for use of the IVD, then appropriate action must be taken.

The methods of obtaining post-market data shall be part of established QMS procedures. Manufacturers should establish procedures to collect information from various sources such as users, service personnel, training personnel, incident reports and customer feedback. While a reference to the QMS procedures can suffice in most cases, any product-specific requirements should be directly added to the risk management plan.

**Reactive PMS:** Information on quality, safety, or performance of an IVD on the market is collected reactively through notification by users and evaluation by manufacturers of complaints, including adverse events. The reactive nature of this statement refers to the fact that the problem has already occurred and may have affected a clinical decision.

**Proactive PMS:** Additional information on quality, safety or performance may also be collected proactively through lot verification testing. This relates to proactively trying to identify a problem before it affects a clinical decision. Lot verification testing is conducted after shipment to the buyer (countries) and can be performed both pre-distribution and post-distribution to end users. Manufacturers should also collect PMS through actively gathering evidence from the literature on their product or similar products, through seeking feedback from customers, and post-market clinical follow up.

An example of an effective proactive PMS activity is through enrolment by the manufacturer in independent external quality assessment schemes (EQAS), also known as proficiency testing. These programs collect testing results from testing sites that receive the same blinded specimens to test. Lot numbers used to test these specimens should be recorded to make these useful data sets. In resource-limited settings, national reference laboratories usually coordinate this data collection.

Each manufacturer is obliged to have undertaken risk management and risk assessment with respect to their IVD before placing it on the market, implementing the most stringent controls to those aspects of design, and manufacturing and control steps where the risk of poor safety and performance is greatest. When a complaint is received, the manufacturer should review and update the risk management file for the IVD accordingly. Manufacturers, regulators, and procurers will use the information that comes from complaints and any other information and experience with a given IVD to determine the scope and stringency of post-market actions in the future.

Post-market information on IVDs empowers manufacturers, national regulatory authorities (NRAs) and WHO to detect, investigate, communicate, and contain events that threaten public health security and to take appropriate action.

**119. Is post-market performance follow-up (PMPF) a mandatory requirement for IVD registration?**

Different regulatory requirements apply to different international markets for IVDs. PMPF plans and reports are essential elements of PMS activities. PMS plans are designed to capture elements impacting the safety quality and performance of an IVD, and as such a PMPF is a subset of these broad plans that focus on monitoring the ongoing performance of the IVD. As most jurisdictions recognise ISO 13485 as the quality system for IVDs, there is an expectation that the manufacturer has PMS plans in place. An audit of the manufacturers QMS will usually include an overview of these plans. Some jurisdictions, e.g. the EU, have regulations that are more explicit when it comes to a manufacturer’s PMS plans, and have a requirement for a PMPF, unless justified. In talking with other experts, most are of the opinion that there are virtually no circumstances where a plan is not required.

In the context of the WHO PQ of IVDs Programme, the type of information and necessary documents required to be submitted in a PD for the purposes of WHO PQ assessment of IVDs are well defined in the WHO PQDx guidance documents such as PQDx_018 Instructions for compilation of a product dossier – IMDRF Table of Contents (IMDRF ToC) (November 2022, version 5) and PQDx_049 Product Dossier Checklist (November 2022, version 4), as well as the
corresponding Technical Specification Series (TSS) document(s) relevant to the product for which an application to PQ is made. Once WHO prequalified IVDs are placed on the market, manufacturers are expected to comply with WHO post-qualification obligations and requirements. Manufacturers are advised of such PMS obligations at the time of submission of a product for PQ assessment, to which they must agree. WHO PQ of IVDs guidance documents outline all applicable PMS obligations. As soon as a product is accepted into the PQ assessment process, and if that product is included in WHO’s list of prequalified IVDs, the manufacturers should follow the guidance document entitled “Guidance for PMS and market surveillance of medical devices, including in vitro diagnostics, in particular, comply with the manufacturer’s obligations set forth in that document.”
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