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Biotechnology — Analytical methods — General requirements and considerations for the testing and characterization of cellular therapeutic products

Biotechnologie — Méthodes analytiques — Exigences et considérations générales pour les essais et la caractérisation de produits de thérapie cellulaire

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Foreword

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The procedures used to develop this document and those intended for its further maintenance are described in the ISO/IEC Directives, Part 1. In particular, the different approval criteria needed for the different types of ISO documents should be noted. This document was drafted in accordance with the editorial rules of the ISO/IEC Directives, Part 2 (see www.iso.org/directives).

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This document was prepared by Technical Committee ISO/TC 276, *Biotechnology*.

Any feedback or questions on this document should be directed to the user's national standards body. A complete listing of these bodies can be found at www.iso.org/members.html.

Introduction

The emergence of cellular therapeutic products has increased the need for high quality, robust, and validated measurements for the characterization and testing of products containing cells as the active substance. These products are regulated by regional health authorities who evaluate product quality in terms of their quality attributes (QAs) via appropriate biological, physical and chemical assays (analytical methods).

Analytical methods are performed on cellular starting materials, in in-process testing and as a part of product conformance testing, comparability studies, and stability testing. These analytical methods are used to assess attributes associated with product quality features and manufacturing controls (in-process controls), and are performed to establish identity, purity, cell count, viability, potency, and stability in all phases of clinical study and commercialization. Quality attributes are used to ensure that only product lots that meet defined specifications are released. Quality attributes are also used for stability testing and trending purposes as well as in-process indicators.

Analytical methods also underpin the development of new cellular therapeutic products by providing insight into biological mechanisms of action and facilitating the research and development that advances manufacturing. In addition, analytical methods are used to evaluate and compare cellular therapeutic products from different batches that have, for example, been produced on different days, at different locations, or via a changed manufacturing process.

Quantitative measurement of a cellular therapeutic product is challenging due to the complex and highly dynamic nature of viable cells and cell samples, the varying vulnerability of cell types and processing steps, a lack of understanding of fundamental cell biology, and the large number of parameters associated with bioprocessing and measurement processes. Biological variability further complicates measurements. Additionally, different donor samples can have different susceptibilities to processing steps, making the need for in-process controls during the measurement process even more critical. As such, analytical methods are key to evaluate cellular therapeutic products, as well as the cellular starting material and intermediate, although the specific performance criteria can be different from those of the final cellular therapeutic products.

This document provides a general approach to design fit for purpose analytical methods to measure and assess quality attributes of a cellular therapeutic product. Aspects of this document can also be applicable to the testing and characterization of cells used in viruses, exosome, and antibody production. The general process to select and design fit for purpose analytical methods can be applied to cellular starting material, intermediates, cell end products, control cells, feeder cells, and cells used in assays (e.g. target cells). It also provides general approaches to understand, minimize, and monitor sources of variability. Acceptable levels of accuracy and precision are guided by the biological implications of the measurement result and the practical limitations of the measurement process.

This document also provides general considerations for setting specifications for the testing of a final cellular therapeutic product. General considerations are also provided for establishing analytical methods and analytical strategies (including analytical method matrix approaches) for common categories of critical quality attributes (CQAs) (i.e. attributes used to establish identity, cell count, purity or impurity, potency or relevant biological activity, viability, sterility, stability, and maturation profile).

This document was developed to provide additional technical guidance on cell characterization and specifically outlines approaches for strategic development of analytical methods cellular therapeutic product characterization and testing (see Annex A for schematic outline of concepts presented in this document).

Biotechnology — Analytical methods — General requirements and considerations for the testing and characterization of cellular therapeutic products

1 Scope

This document provides general requirements for the testing of cellular therapeutic products intended for human use.

This document also provides considerations for the characterization of cellular therapeutic products, including approaches to select and design analytical methods that are fit for purpose.

Such considerations can be used to establish critical quality attributes for a cellular therapeutic product.

This document is applicable to cellular starting materials (including those for tissue engineered products) and intermediates of cellular therapeutic products.

This document is not applicable to tissues used in transplantation.

2 Normative references

There are no normative references in this document.

3 Terms and definitions

For the purposes of this document, the following terms and definitions apply.

ISO and IEC maintain terminology databases for use in standardization at the following addresses:

— ISO Online browsing platform: available at https://www.iso.org/obp

— IEC Electropedia: available at http://www.electropedia.org/

3.1
acceptance criteria
numerical limits, ranges, or other suitable measures for acceptance of the results of analytical procedures which the product or materials at other stages of manufacture is intended to meet

3.2
adventitious agent
microorganisms unintentionally introduced into the manufacturing process

Note 1 to entry: Microorganisms can include bacteria, fungi, mycoplasma or spiroplasma, mycobacteria, rickettsia, protozoa, parasites, transmissible spongiform encephalopathy (TSE) agents and viruses.

Note 2 to entry: Adventitious agents are a subset of impurity for *cellular therapeutic product* (3.15).

3.3
analytical method
investigative procedure for qualitatively or quantitatively measuring or assessing the presence, amount, or functional activity of a target entity (the analyte)

3.4
analytical target profile
ATP
predefined objective that stipulates the *performance criteria* (3.34) for a *test method* (3.52)

Note 1 to entry: The ATP states the required quality of the results produced by a *test method* (3.52).

3.5
ancillary material
AM
material that comes into contact with the cell or tissue product during cell-processing, but is not intended to be part of the final product formulation

Note 1 to entry: AMs exclude non-biological consumables (e.g. tissue culture flasks, bags, tubing, pipettes, needles) and other plastic ware that come into contact with the cell or tissue but include consumables which can have a biological component (e.g. coated dishes or beads).

Note 2 to entry: AMs exclude feeder cells (cells that are used in the manufacturing process but are not a part of the cellular starting material).

Note 3 to entry: In some cases, AMs are described as raw materials.

[SOURCE: ISO/TS 20399-1:2018, 3.1, modified — Note 2 was reduced to the exclusion of feeder cells and the example was adjusted accordingly.]

Note 4 to entry: For the purposes of this document, the final product formulation is a *cellular therapeutic product* (3.15).

Note 5 to entry: Nature of both biological and synthetic material as well as their impact on cells can be highly complex. Thus, making assumptions as to that a synthetic material is less variable or complex than biological material would be wrong.

3.6
area density
<cells> *cell count* (3.14) of adherent cells on a surface, typically expressed as number of cells per unit area

[SOURCE: ISO 20391-1:2018, 3.4]

3.7
analytical method matrix
set of two or more complementary *analytical methods* (3.3) to measure different aspects of a *quality attribute* (3.38)

3.8
attribute
physical, chemical, biological, or microbiological property or characteristic

[SOURCE: ISO 20391-1:2018, 3.5]

3.9
attribute component
quantity (3.39) used to derive a *quality attribute* (3.38)

3.10
biological activity
specific ability or capacity of the product to achieve a defined biological effect

Note 1 to entry: The biological activity is potentially modified by stimulations including for example chemical, physical, or mechanical stimuli as well as other changes in environment and with time.

3.11
biological property
biological phenomenon that is evaluated to assess the *quality attribute* (3.38)

3.12
calibration
operation that, under specified conditions in a first step, establishes a relation between the *quantity* (3.39) values with measurement uncertainties provided by measurement standards and corresponding indications with associated measurement uncertainties and, in a second step, uses this information to establish a relation for obtaining a measurement result from an indication

[SOURCE: ISO/IEC Guide 99:2007, 2.39, modified — The notes were deleted.]

3.13
cell concentration
cell count (3.14) per volume

Note 1 to entry: Typically used for cells in suspension.

[SOURCE: ISO 20391-1:2018, 3.6]

3.14
cell count
discrete number of cells

Note 1 to entry: Cell count is typically expressed as *cell concentration* (3.13) or *area density* (3.6).

[SOURCE: ISO 20391-1:2018, 3.7]

3.15
cellular therapeutic product
product containing cells as the active substance

[SOURCE: ISO/TS 20399-1:2018, 3.5, modified — Example deleted.]

Note 1 to entry: The following are examples of cellular therapeutic product:

a) a cell therapy medicinal product;

b) a tissue engineered product.

3.16
cellular starting material
living and functional cellular material present at the beginning of a *cellular therapeutic product* (3.15) manufacturing process

Note 1 to entry: There are other types of starting materials also relevant for *cellular therapeutic products* (3.15).

3.17
certified reference material
reference material characterized by a metrologically valid procedure for one or more specified properties, accompanied by an RM certificate that provides the value of the specified property, its associated uncertainty, and a statement of metrological traceability

[SOURCE: ISO Guide 33:2015, 3.2]

3.18
comparable
conclusion that products have highly similar *quality attributes* (3.38) before and after manufacturing process changes and that no adverse impact on the safety or efficacy, including immunogenicity of the drug product occurred

Note 1 to entry: This conclusion can be based on an analysis of product *quality attribute* (3.38). In some cases, non-clinical or clinical data can contribute to the conclusion.

3.19
contaminant
any adventitiously introduced material not intended to be part of the manufacturing process of the drug substance or drug product

Note 1 to entry: Adventitiously introduced materials can be e.g. chemical, biochemical, or microbial species.

3.20
critical quality attribute
CQA
physical, chemical, biological, or microbiological property or characteristic intended to be within an appropriate limit, range, or distribution to ensure the desired quality and consistency of a product

Note 1 to entry: CQA is generally related to the clinical efficacy and safety of the product.

3.21
final cellular therapeutic product
formulated *cellular therapeutic product* (3.15) intended for administration to human subjects

3.22
fit for purpose
fitness for the intended purpose
in line with prearranged requirements for an *intended use* (3.26)

[SOURCE: ISO 20387:2018, 3.24, modified — Note deleted.]

3.23
impurity
any component present in the product which is not the desired product, a product-related substance, or excipient including buffer components

Note 1 to entry: An impurity can be either process- or product-related.

3.24
in-house reference material
non-certified material or substance, produced by one laboratory, one or more of whose property values are sufficiently homogeneous and well established to be used for the *intended use* (3.26)

Note 1 to entry: The use of in-house reference materials can include, but is not limited to, *validation* (3.54), *calibration* (3.12), monitoring of comparability, and *potency* (3.36) and process evaluations.

[SOURCE: ISO 16140-1:2016, 2.32, modified — “validation” replaced with “the intended purpose”, note added.]

3.25
installation qualification
IQ
establishing by objective evidence that all key aspects of the process equipment and ancillary system installation adhere to the manufacturer’s approved *specification* (3.50) and that the recommendations of the supplier of the equipment are suitably considered

3.26
intended use
intended purpose
use for which a product, process, or service is intended according to the *specifications* (3.50), instructions or information or multiple of them provided by the manufacturer or user

[SOURCE: ISO/IEC Guide 63:2019, 3.4, modified — Added new term, as well as “or multiple of them” and “or user” to the definition; “and” was replaced by “or”.]

3.27
intermediate
material in manufacturing process that is nominally between two unit operations

3.28
intermediate precision
measurement *precision* (3.37) under a set of intermediate precision conditions

[SOURCE: ISO/IEC Guide 99:2007, 2.23, modified — “measurement” was deleted from the term, note deleted.]

Note 1 to entry: The intermediate precision condition refers to a set of conditions that includes the same measurement procedure, same location, and replicate measurements on the same or similar objects over an extended period of time, but can include other conditions involving changes (e.g. analyst or instrument).

3.29
limit of detection
lowest amount of analyte in a *sample* (3.46) which can be detected but not necessarily quantitated as an exact value

3.30
limit of quantitation
lowest amount of analyte in a *sample* (3.46) which can be quantitatively determined with suitable *precision* (3.37) and accuracy

Note 1 to entry: The limit of quantitation is a parameter of quantitative analytical methods for low levels of compounds in sample matrices and is used particularly for the determination of *impurities* (3.23) or degradation products or both.

3.31
measurement target
intended object of measurement

Note 1 to entry: A measurement target can denote a feature or complex features of cells that is informative of cellular status or quality. The term is additional to the term analyte or measurand in situations where the use of those terms is not appropriate or possible.

3.32
nominal property
property of a phenomenon, body, or substance, where the property has no magnitude

[SOURCE: ISO/IEC Guide 99:2007, 1.30, modified — Notes and examples deleted.]

Note 1 to entry: The nominal property of a measurement target is one that can be described but not quantified with a magnitude.

3.33
operational qualification
OQ
establishing by objective evidence process control limits and action levels which result in an *analytical method* (3.3) that meets all predetermined requirements

3.34
performance criteria
required functionality and behaviour of the *test method* (3.52)

3.35
performance qualification
PQ
establishing by objective evidence that the *analytical method* (3.3), under anticipated conditions, consistently meets all predetermined requirements

3.36
potency
measure of the *biological activity* (3.10) using a suitably quantitative *analytical method* (3.3), based on the *attribute* (3.8) of the product which is linked to the relevant *biological properties* (3.11)

3.37
precision
closeness of agreement between indications or measured *quantity* (3.39) values obtained by replicate measurements on the same or similar objects under specified conditions

Note 1 to entry: Precision is usually expressed numerically by measures of imprecision, such as standard deviation, variance, or coefficient of variation under the specified conditions of measurement.

Note 2 to entry: The 'specified conditions' can be, for example, repeatability conditions of measurement, intermediate precision conditions of measurement, or reproducibility conditions of measurement.

[SOURCE: ISO/IEC Guide 99:2007, 2.15, modified — Term "measurement precision" was deleted. Notes 3 and 4 were deleted.]

Note 3 to entry: Measured *quantity* (3.39) value refers to the *quantity* (3.39) value representing a measurement result.

3.38
quality attribute
physical, chemical, biological, or microbiological property or characteristic that is an indicator of the quality

3.39
quantity
property of a substance with a magnitude that can be expressed as a number and a reference

[SOURCE: ISO/IEC Guide 99:2007, 1.1, modified — Notes, example, and "phenomenon, body, or" were deleted.]

3.40
reference material
material, sufficiently homogeneous and stable with reference to specified properties, which has been established to be fit for its intended use in measurement or in examination of *nominal properties* (3.32)

[SOURCE: ISO/IEC Guide 99:2007, 5.13, modified — Notes and examples deleted.]

3.41
repeatability
measurement *precision* (3.37) under a set of repeatability conditions of measurement

[SOURCE: ISO/IEC Guide 99:2007, 2.21, modified — Term "measurement repeatability" was deleted.]

Note 1 to entry: Repeatability conditions of a measurement refers to condition of measurement, out of a set of conditions that includes the same measurement procedure, same operators, same measuring system, same operating conditions and same location, and replicate measurements on the same or similar objects over a short period of time.

3.42
representative sample
sample (3.46) that accurately represents or reflects the *attributes* (3.8) of the system

Note 1 to entry: Generally intended to provide information on the system, often to serve as a basis for decision on the system or its production.

3.43
reproducibility
measurement *precision* (3.37) under reproducibility conditions of measurement

[SOURCE: ISO/IEC Guide 99:2007, 2.25, modified — Note and term "measurement reproducibility" deleted.]

Note 1 to entry: Reproducibility conditions of measurement refer to the condition of measurement, out of a set of conditions that includes different locations, operators, measuring systems, and replicate measurements on the same or similar objects.

3.44
robustness
measure of a *test method* (3.52) capacity to remain unaffected by small, but deliberate variations in method parameters and provides an indication of its reliability during normal usage

3.45
ruggedness
degree of *reproducibility* (3.43) of test results obtained by the analysis of the same *samples* (3.46) under a variety of normal test conditions

Note 1 to entry: Normal test conditions can include for example: different laboratories, different analysts, different instruments, different reagent lots, different analysis days, different elapsed times, different temperatures etc.

3.46
sample
one or more parts taken from a system

3.47
sensitivity
quotient of the change in an indication of a measuring system and the corresponding change in a value of a *quantity* (3.39) being measured

[SOURCE: ISO/IEC Guide 99:2007, 4.12, modified — Notes and term "sensitivity of a measuring system" deleted.]

3.48
specificity
characteristic of a *test method* (3.52) that expresses qualitatively and quantitatively its ability to detect or determine an individual analyte without interferences from accompanying species

Note 1 to entry: Specificity increases with *sensitivity* (3.47) and amount of analyte and decreases with increasing cross-sensitivity and amounts of accompanying species and larger disturbing effects.

3.49
shelf life
specific time period for which a *cellular therapeutic product* (3.15) can maintain suitability for its *intended use* (3.26)

3.50
specification
list of tests, references to analytical procedures, and appropriate *acceptance criteria* (3.1) that would be expected to be met to demonstrate suitability for its *intended use* (3.26)

[SOURCE: ISO/TS 20399-1:2018, 3.9, modified — Replaced "intended use definition" by "intended use".]