SLOVENSKI STANDARD
SIST-TS CEN/TS 17390-2:2020

01-marec-2020

Molekularne diagnostične preiskave in vitro - Specifikacije za predpreiskovalne procese za cirkulirajoče tumorske celice (CTC) v venski polni krvi - 2. del: Izolirana DNK

Molecular in vitro diagnostic examinations - Specifications for pre-examination processes for circulating tumor cells (CTCs) in venous whole blood - Part 2: Isolated DNA

Molekularanalytische in vitro-diagnostische Verfahren - Spezifikationen für präanalytische Prozesse für zirkulierende Tumorzellen (CTC) in venösen Vollblutproben - Teil 2: Isolierte DNA

Analyses de diagnostic moléculaire in vitro - Spécifications relatives aux processus préanalytiques pour les cellules tumorales circulantes (CTCs) dans le sang total veineux - Partie 2: ADN extrait

Ta slovenski standard je istoveten z: **CEN/TS 17390-2:2020**

ICS:

11.100.10	Diagnostični preskusni sistemi in vitro	In vitro diagnostic test systems

SIST-TS CEN/TS 17390-2:2020 **en,fr,de**

TECHNICAL SPECIFICATION

SPÉCIFICATION TECHNIQUE

TECHNISCHE SPEZIFIKATION

CEN/TS 17390-2

January 2020

ICS 11.100.10

English Version

Molecular in vitro diagnostic examinations - Specifications for pre-examination processes for circulating tumor cells (CTCs) in venous whole blood - Part 2: Isolated DNA

Analyses de diagnostic moléculaire in vitro - Spécifications relatives aux processus préanalytiques pour les cellules tumorales circulantes (CTCs) dans le sang total veineux - Partie 2: ADN extrait

Molekularanalytische in vitro-diagnostische Verfahren - Spezifikationen für präanalytische Prozesse für zirkulierende Tumorzellen (CTC) in venösen Vollblutproben - Teil 2: Isolierte DNA

This Technical Specification (CEN/TS) was approved by CEN on 27 October 2019 for provisional application.

The period of validity of this CEN/TS is limited initially to three years. After two years the members of CEN will be requested to submit their comments, particularly on the question whether the CEN/TS can be converted into a European Standard.

CEN members are required to announce the existence of this CEN/TS in the same way as for an EN and to make the CEN/TS available promptly at national level in an appropriate form. It is permissible to keep conflicting national standards in force (in parallel to the CEN/TS) until the final decision about the possible conversion of the CEN/TS into an EN is reached.

CEN members are the national standards bodies of Austria, Belgium, Bulgaria, Croatia, Cyprus, Czech Republic, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Iceland, Ireland, Italy, Latvia, Lithuania, Luxembourg, Malta, Netherlands, Norway, Poland, Portugal, Republic of North Macedonia, Romania, Serbia, Slovakia, Slovenia, Spain, Sweden, Switzerland, Turkey and United Kingdom.

EUROPEAN COMMITTEE FOR STANDARDIZATION
COMITÉ EUROPÉEN DE NORMALISATION
EUROPÄISCHES KOMITEE FÜR NORMUNG

CEN-CENELEC Management Centre: Rue de la Science 23, B-1040 Brussels

Ref. No. CEN/TS 17390-2:2020 E

Contents

Page

European foreword

This document (CEN/TS 17390-2:2020) has been prepared by Technical Committee CEN/TC 140 “In vitro diagnostic medical devices”, the secretariat of which is held by DIN.

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. CEN shall not be held responsible for identifying any or all such patent rights.

CEN/TS 17390 consists of the following parts, under the general title *Molecular in vitro diagnostic examinations — Specifications for pre-examination processes for Circulating Tumor Cells (CTCs) in venous whole blood*:

— Part 1: Isolated RNA

— Part 2: Isolated DNA

— Part 3: Preparations for analytical CTC staining

According to the CEN/CENELEC Internal Regulations, the national standards organisations of the following countries are bound to announce this Technical Specification: Austria, Belgium, Bulgaria, Croatia, Cyprus, Czech Republic, Denmark, Estonia, Finland, France, Germany, Greece, Hungary, Iceland, Ireland, Italy, Latvia, Lithuania, Luxembourg, Malta, Netherlands, Norway, Poland, Portugal, Republic of North Macedonia, Romania, Serbia, Slovakia, Slovenia, Spain, Sweden, Switzerland, Turkey and the United Kingdom.

Introduction

Solid tumours release cells and bioanalytes into blood and other body fluids. This has opened the option of minimally-invasive tumour detection, diagnosis and characterization from venous whole blood (liquid biopsies). Liquid biopsies are expected to enable earlier detection and diagnosis of cancers and advance personalized patient treatment. These applications have become one of the fastest growing segments of the entire diagnostic market.

Circulating tumour cells (CTCs) in venous whole blood reflect the disease complexity that evolves during tumour progression, with distinct genetic, epigenetic and expression features. Beside the prognostic role of CTC identification and/or enumeration in cancer progression, CTC molecular characterization can improve e.g. disease outcome prediction, therapeutic guidance and post-treatment monitoring of the patient.

CTCs are now considered as a surrogate of tumour tissue in cancer early development, progression and metastatic phase.

Molecular characterization of CTCs can provide for example a strategy for monitoring cancer genotype during systemic therapies [1], identification of mechanisms of disease progression, identification of novel targets for biological treatment [2] and to select targeted therapies. Moreover, CTC single-cell sequencing is as an important tool for tumour genomic heterogeneity analysis [3] [4] [5]. Molecular examination techniques such as qPCR, dPCR and sequencing methods including next generation sequencing (NGS) enable to characterize CTC specific DNA features.

CTCs are fragile and tend to degrade within a few hours when collected in conventional blood collection tubes, e.g. EDTA containing tubes, without dedicated CTC stabilizers. CTCs are extremely rare, especially in early disease, e.g. less than 10 cells per 10 ml of blood, representing a ratio of approx. 1:10^7 CTCs to white blood cells (WBCs). This low ratio represents a significant challenge to CTC enrichment required for examination. Furthermore, co-enrichment of normal blood cells causes a dilution of CTCs. The challenge is to minimize the amount of co-enriched WBCs for subsequent accurate and sensitive detection of CTC specific genetic and epigenetic alterations, especially when dealing with minor tumour cell clones.

Special measures need to be taken to get rid of the WBCs in order to obtain good quality DNA samples characterized by high purity and thus representative of the mutational pattern within the tumour.

Standardization of all steps of the pre-examination process is required. This includes blood collection and stabilization, transport, storage, CTC enrichment, CTC isolation (if required), and DNA isolation. An exemplary complete workflow for the molecular characterization of single CTCs is provided in Annex A. A decision guideline for the critical steps of the CTC pre-analytical workflow for DNA isolation is provided in Annex B.

This document describes special measures that need to be taken to obtain appropriate quality and quantity of DNA from CTC containing blood specimens for subsequent examination.

In this document, the following verbal forms are used:

— “shall” indicates a requirement;

— “should” indicates a recommendation;

— “may” indicates a permission;

— “can” indicates a possibility or a capability.

1 Scope

This document gives guidelines on the handling, storage, processing and documentation of venous whole blood specimens intended for the examination of human DNA isolated from circulating tumour cells (CTCs) during the pre-examination phase before a molecular examination is performed.

This document is applicable to molecular *in vitro* diagnostic examinations including laboratory developed tests performed by medical laboratories. It is also intended to be used by laboratory customers, *in vitro* diagnostics developers and manufacturers, biobanks, institutions and commercial organizations performing biomedical research, and regulatory authorities.

This document does not cover the isolation of genomic DNA directly from venous whole blood containing CTCs. This is covered in EN ISO 20186-2, *Molecular in vitro diagnostic examinations - Specifications for pre-examination processes for venous whole blood - Part 2: Isolated genomic DNA*.

This document does not cover the isolation of specific white blood cells and subsequent isolation of genomic DNA therefrom.

This document does not cover pre-analytical workflow requirements for viable CTC cryopreservation and culturing.

NOTE 1 The requirements given in this document can also be applied to other circulating rare cells (e.g. foetal cells).

NOTE 2 International, national or regional regulations or requirements can also apply to specific topics covered in this document.

2 Normative references

The following documents are referred to in the text in such a way that some or all of their content constitutes requirements of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

EN ISO 15189:2012, *Medical laboratories - Requirements for quality and competence (ISO 15189:2012, Corrected version 2014-08-15)*

ISO 15190, *Medical laboratories — Requirements for safety*

3 Terms and definitions

For the purposes of this document, the terms and definitions given in EN ISO 15189 and the following terms and definitions apply.

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

- IEC Electropedia: available at http://www.electropedia.org/
- ISO Online browsing platform: available at http://www.iso.org/obp

3.1
aliquot
portion of a larger amount of homogenous material, assumed to be taken with negligible sampling error

Note 1 to entry: The term is usually applied to fluids. Tissues are heterogeneous and therefore cannot be aliquoted.

Note 2 to entry: The definition is derived from References [6], [7] and [8].

[SOURCE: EN ISO 20166-3:2019, 3.1]

3.2
ambient temperature
unregulated temperature of the surrounding air

[SOURCE: EN ISO 20166-3:2019, 3.2]

3.3
analyte
component represented in the name of a measurable quantity

[SOURCE: EN ISO 17511:2003, 3.2, modified — EXAMPLE has been removed.]

3.4
analytical test performance
accuracy, precision, specificity and sensitivity of a test to measure the analyte of interest

Note 1 to entry: Other test performance characteristics such as robustness, repeatability can apply as well.

[SOURCE: EN ISO 20184-1:2018, 3.4, modified — "specificity" was added.]

3.5
blood collection set
intravenous device specialized for venipuncture consisting of a stainless steel bevelled needle and tube (tubing) with attached plastic wings and fitting connector

Note 1 to entry: The connector attaches to an additional blood collection device, e.g. a blood collection tube.

3.6
blood collection tube
tube used for blood collection, usually in a vacuum which forces blood from the vein through the needle and into the tube

3.7
backflow
flow of a liquid opposite to the usual or desired direction

3.8
circulating tumor cells
CTCs
cells present in blood, originating from a primary and/or metastatic site of a tumor

3.9
CTC enrichment
any method that is able to increase the ratio of CTCs to other cells

3.10
CTC isolation
any method resulting in a sample containing CTCs without any other cell type

3.11
DNA
deoxyribonucleic acid
polymer of deoxyribonucleotides occurring in a double-stranded (dsDNA) or single-stranded (ssDNA) form

[SOURCE: EN ISO 22174:2005, 3.1.2]

3.12
diagnosis
identification of a disease from its signs and symptoms

Note 1 to entry: Where the diagnostic process can involve examinations and tests for classification of an individual's condition into separate and distinct categories or subclasses that allow medical decisions about treatment and prognosis to be made

[SOURCE: EN ISO 20184-1:2018, 3.6]

3.13
examination
analytical test
set of operations having the object of determining the value or characteristics of a property

Note 1 to entry: Processes that start with the isolated analyte and include all kinds of parameter testing or chemical manipulation for quantitative or qualitative examination.

[SOURCE: EN ISO 15189:2012, 3.7, modified — Notes to entry 1 to 3 have been removed, Note 1 to entry has been added and “analytical test” has been added as a preferred term.]

3.14
examination performance
analytical test performance
analytical performance
ability of an examination procedure to measure or detect a particular analyte

Note 1 to entry: Analytical performance is determined from analytical performance studies used to assess the ability of an *in vitro* diagnostic examination procedure to measure or detect a particular analyte.

Note 2 to entry: Analytical performance includes such characteristics as analytical sensitivity, detection limit, analytical specificity (interference and cross-reactivity), trueness, precision and linearity.

[SOURCE: ISO/TS 17822-1:2014, 3.2, modified — “analytical test performance” and “analytical performance” have been added as preferred terms.]

3.15
examination manufacturer
analytical test manufacturer
group or company that provides the specific analytical test

3.16
needle holder
barrel used in routine venipuncture procedures to hold the blood collection tube in place and to protect the phlebotomist from direct contact with blood

[SOURCE: EN ISO 20186-1:2019, 3.24]

3.17
pre-examination processes
preanalytical phase
preanalytical workflow
processes that start, in chronological order, from the clinician's request and include the examination request, preparation and identification of the patient, collection of the primary sample(s), transportation to and within the medical or pathology laboratory, CTC enrichment, CTC isolation where applicable, isolation of analytes, and end when the analytical examination begins

Note 1 to entry: The pre-examination phase includes preparative processes that influence the outcome of the intended examination.

[SOURCE: EN ISO 15189:2012, 3.15, modified — "pre-analytical workflow" has been added as a preferred term, Note 1 to entry has been added and the definition has been extended.]

3.18
primary sample
specimen
discrete portion of a body fluid, breath, hair or tissue taken for examination, study or analysis of one or more quantities or properties assumed to apply for the whole

[SOURCE: EN ISO 15189:2012, 3.16, modified — Notes to entry 1 to 3 have been removed.]

3.19
proficiency testing
evaluation of participant performance against pre-established criteria by means of inter-laboratory comparisons

[SOURCE: EN ISO/IEC 17043:2010, 3.7, modified — Notes to entry 1 to 3 have been removed.]

3.20
DNA proficiency testing program
proficiency testing for DNA based examinations

3.21
room temperature
for the purposes of this document, temperature in the range of 18 °C to 25 °C

Note 1 to entry: Local or national regulations can have different definitions.

3.22
sample
one or more parts taken from a primary sample

[SOURCE: EN ISO 15189:2012, 3.24, modified — EXAMPLE has been removed.]

3.23
stability
ability of a sample material, when stored under specified conditions, to maintain a stated property value within specified limits for a specified period of time

Note 1 to entry: The analyte for the purpose of this document is DNA.

[SOURCE: ISO Guide 30:2015, 2.1.15, modified — The words "reference material" were replaced by "sample material".]

3.24
storage
prolonged interruption of the preanalytical workflow of a sample or analyte respectively, or of their derivatives e.g., stained sections or tissue blocks, under appropriate conditions in order to preserve their properties

Note 1 to entry: Long-term storage typically occurs in laboratory archives or in biobanks.

[SOURCE: EN ISO 20166-3:2019, 3.24]

3.25
validation
confirmation, throughout the provision of objective evidence, that the requirements for a specific intended use or application have been fulfilled

Note 1 to entry: The term "validated" is used to designate the corresponding status.

[SOURCE: EN ISO 9000:2015, 3.8.13, modified — Notes to entry 1 to 3 have been removed.]

3.26
verification
confirmation, through provision of objective evidence, that specified requirements have been fulfilled

Note 1 to entry: The term "verified" is used to designate the corresponding status.

Note 2 to entry: Confirmation can comprise activities such as:

— performing alternative calculations,

— comparing a new design specification with a similar proven design specification,

— undertaking tests and demonstrations, and

— reviewing documents prior to issue.

[SOURCE: EN ISO 9000:2015, 3.8.12, modified — Notes to entry 1 and 2 have been removed.]

3.27
workflow
series of activities necessary to complete a task

[SOURCE: EN ISO 20166-3:2019, 3.29]