

English Version

**Molecular in vitro diagnostic examinations - Specifications
for pre-examination processes for venous whole blood -
Isolated circulating cell free RNA from plasma**

Molekularanalytische in-vitro-diagnostische Verfahren
- Spezifikationen für präanalytische Prozesse für
venöse Vollblutproben - Isolierte zirkulierende
zellfreie RNA aus Plasma

This Technical Specification (CEN/TS) was approved by CEN on 14 February 2022 for provisional application.

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

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European foreword

This document (CEN/TS 17742:2022) has been prepared by Technical Committee CEN/TC 140 “In vitro diagnostic medical devices”, the secretariat of which is held by DIN.

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Introduction

Molecular *in vitro* diagnostics has enabled a significant progress in medicine. Further progress is expected by new technologies analysing profiles of nucleic acids, proteins, and metabolites in human tissues and body fluids. However, the profiles of these molecules can change drastically during the pre-examination process, including the specimen collection, transport, storage and processing.

Consequently, this makes the outcome from diagnostics or research unreliable or even impossible, as the subsequent examination might not determine the real situation in the patient, but an artificial profile generated during the pre-examination processes.

Besides circulating cell free DNA (ccfDNA), circulating tumour cells (CTCs) and other rare cells, circulating cell free RNA (ccfRNA) represents another key component of liquid biopsies. Therefore, there is a strongly increasing interest in emerging diagnostics and research in ccfRNA.

In blood, ccfRNA is composed of extracellular RNA present both in and outside of exosomes and other extracellular vesicles. The pre-examination process described in this document results in total ccfRNA isolated from blood plasma.

ccfRNA profiles and quantity can change significantly after blood collection, e.g. by release and/or uptake of RNA containing extracellular vesicles by cells present in the blood specimen as well as by ccfRNA degradation. Therefore, special measures have to be taken to secure good quality specimens for ccfRNA examination.

Standardization of the entire workflow from specimen collection to the ccfRNA examination is therefore needed. Studies have been undertaken to determine the important influencing factors. This document draws upon such work to codify and standardize the steps of ccfRNA examination in what is referred to as the pre-examination phase.

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 specifies requirements and recommendations for the pre-examination phase of circulating cell free RNA (ccfRNA) from venous whole blood specimens, including but not limited to the collection, handling, storage, processing and documentation of venous whole blood specimens intended for ccfRNA examination. This document covers specimens collected in venous whole blood collection tubes.

The pre-examination process described in this document results in circulating cell free RNA isolated from blood plasma without prior enrichment of exosomes and other extracellular vesicles.

This document is applicable to molecular *in vitro* diagnostic examinations 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.

Different dedicated measures need to be taken during the pre-examination phase for isolated RNA from enriched exosomes and other extracellular vesicles enriched from venous whole blood and for cellular RNA isolated from venous whole blood. These are not described in this document but are covered in CEN/TS 17747,¹ *Molecular in vitro diagnostic examinations - Specifications for pre-examination processes for exosomes and other extracellular vesicles in venous whole blood - Isolated DNA, RNA and proteins*, and in EN ISO 20186-1, *Molecular in vitro diagnostic examinations - Specifications for pre-examination processes for venous whole blood - Part 1: Isolated cellular RNA*.

NOTE 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, *Medical laboratories - Requirements for quality and competence (ISO 15189)*

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 apply.

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

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

¹ Under preparation. Stage at the time of publication: FprCEN/TS 17747.

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 the Compendium of Chemical Terminology Gold Book. International Union of Pure and Applied Chemistry. Version 2.3.3., 2014; the PAC, 1990,62,1193 (Nomenclature for sampling in analytical chemistry (Recommendations 1990)) p. 1206; and the PAC 1990, 62, 2167 (Glossary of atmospheric chemistry terms (Recommendations 1990)) p. 2173.

3.2

ambient temperature

unregulated temperature of the surrounding air

[SOURCE: EN ISO 20166-1:2018, 3.2]

3.3

analyte

component represented in the name of a measurable quantity

[SOURCE: ISO 17511:2020, 3.2, modified — Deleted Example.]

3.4

backflow

flow of a liquid opposite to the usual or desired direction

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 with a vacuum which forces blood from the vein through the needle into the tube

3.7

circulating cell free DNA

ccfDNA

extracellular human DNA present in blood and plasma

Note 1 to entry: ccfDNA includes DNA present in extracellular vesicles such as exosomes.

3.8

circulating cell free RNA

ccfRNA

extracellular human RNA present in blood and plasma

Note 1 to entry: ccfRNA includes RNA present in extracellular vesicles such as exosomes.