SLOVENSKI STANDARD
SIST EN ISO 21427-2:2009

01-maj-2009

Kakovost vode - Vrednotenje genotoksiļ nosti z merjenjem indukcije mikronukleusov - 2. del: Metoda z nesinhronizirano populacijo celic V79 celiļ ne linije (ISO 21427-2:2006)

Water quality - Evaluation of genotoxicity by measurement of the induction of micronuclei - Part 2: Mixed population method using the cell line V79 (ISO 21427-2:2006)

Wasserbeschaffenheit - Bestimmung der Gentoxizität mit dem In-vitro-Mikrokerntest - Teil 2: Verwendung einer nicht-synchronisierten V79-Zellkulturlinie (ISO 21427-2:2006)

Qualité de l'eau - Évaluation de la génotoxicité par le mesurage de l'induction de micronoyaux - Partie 2: Méthode de la population mélangée à l'aide de la lignée de cellules V79 (ISO 21427-2:2006)

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13.060.70	Preiskava bioloških lastnosti vode	Examination of biological properties of water

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English Version

Water quality - Evaluation of genotoxicity by measurement of the induction of micronuclei - Part 2: Mixed population method using the cell line V79 (ISO 21427-2:2006)

Qualité de l'eau - Évaluation de la génotoxicité par le mesurage de l'induction de micronoyaux - Partie 2: Méthode de la population mélangée à l'aide de la lignée de cellules V79 (ISO 21427-2:2006)

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

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Contents

Page

Foreword

The text of ISO 21427-2:2006 has been prepared by Technical Committee ISO/TC 147 “Water quality” of the International Organization for Standardization (ISO) and has been taken over as EN ISO 21427-2:2009 by Technical Committee CEN/TC 230 “Water analysis” the secretariat of which is held by DIN.

This European Standard shall be given the status of a national standard, either by publication of an identical text or by endorsement, at the latest by September 2009, and conflicting national standards shall be withdrawn at the latest by September 2009.

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Endorsement notice

The text of ISO 21427-2:2006 has been approved by CEN as a EN ISO 21427-2:2009 without any modification.

INTERNATIONAL STANDARD

ISO 21427-2

First edition
2006-11-15

Water quality — Evaluation of genotoxicity by measurement of the induction of micronuclei —

Part 2: Mixed population method using the cell line V79

Qualité de l'eau — Évaluation de la génotoxicité par le mesurage de l'induction de micronoyaux —

Partie 2: Méthode de la population mélangée à l'aide de la lignée de cellules V79

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Contents

Page

Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

International Standards are drafted in accordance with the rules given in the ISO/IEC Directives, Part 2.

The main task of technical committees is to prepare International Standards. Draft International Standards adopted by the technical committees are circulated to the member bodies for voting. Publication as an International Standard requires approval by at least 75 % of the member bodies casting a vote.

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ISO 21427-2 was prepared by Technical Committee ISO/TC 147, *Water quality*, Subcommittee SC 5, *Biological methods*.

ISO 21427 consists of the following parts, under the general title *Water quality — Evaluation of genotoxicity by measurement of the induction of micronuclei*:

— *Part 1: Evaluation of genotoxicity using amphibian larvae*

— *Part 2: Mixed population method using the cell line V79*

Water quality — Evaluation of genotoxicity by measurement of the induction of micronuclei —

Part 2: Mixed population method using the cell line V79

WARNING — Persons using this part of ISO 21427 should be familiar with normal laboratory practice. This standard does not purport to address all of the safety problems, if any, associated with its use. It is the responsibility of the user to establish appropriate safety and health practices and to ensure compliance with any national regulatory conditions.

IMPORTANT — It is absolutely essential that tests conducted according to this part of ISO 21427 be carried out by suitably trained staff.

1 Scope

This part of ISO 21427 specifies a method for the determination of genotoxicity of water and waste water using a mammalian *in vitro* test which detects damage, induced by water-soluble substances, to the chromosomes or the mitotic apparatus of V79 cells from the Chinese hamster.

The micronucleus test allows the identification of substances that cause cytogenetic damage which results in the formation of micronuclei containing lagging chromosome fragments and/or whole chromosomes.

The assay is based on the increase in the frequency of micronucleated cells after incubation with and without metabolic activation.

2 Normative references

The following referenced documents are indispensable for the application 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.

ISO 5667-16, *Water quality — Sampling — Part 16: Guidance on biotesting of samples*

3 Terms and definitions

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

3.1
cell lines
distinct families of cells grown in culture originated from a single clone

3.2
cofactor solution
aqueous solution of chemicals (e.g. NADP, Glucose-6-phosphate and inorganic salts) needed for the activity of the enzymes in the S9 fraction

3.3
dilution level D
denominator of the dilution coefficient (using the numerator 1) of a mixture of water or waste water with dilution water as integral number

NOTE For undiluted water or waste water, this coefficient is per definition 1:1. The corresponding smallest possible D value is 1.

3.4
D value
smallest value of D at which, under the conditions of this part of ISO 21427, no increase in the number of micronuclei per culture is detected

NOTE In the case of more than one D value (at maximum two are possible, see 9.2), the highest D value is decisive.

3.5
karyotype
characteristic of the nucleus of a cell, including its size, form and chromosome number

3.6
micronuclei
small particles consisting of acentric fragments of chromosomes and/or entire chromosomes which lag behind at anaphase stage of cell division and form, after telophase, single or multiple micronuclei in the cytoplasm

3.7
mitotic index
percentage of cells of a cell population under division at a particular time of observation

3.8
plating efficiency
measure of the number of colonies originated from single cells

3.9
proliferation index
rate at which cells are dividing within the culture

3.10
proliferation rate
rate with which cells replicate, calculated by a formula which takes into account 1, 2, 4 and 8 cell stages of clones

3.11
S9 fraction
9 000 g supernatant of a tissue homogenate in 0,15 mol/l KCl, obtained from livers of male rats (200 g to 300 g) pretreated with an appropriate substance or substance combination for enzyme induction

3.12
S9 mix
mixture of the S9 fraction and the cofactor solution

3.13
stock culture
frozen culture for the preservation of the characteristics of V79 cells

3.14
survival index
percentage of surviving cells compared to all cells, used as index of toxicity

3.15
test culture
culture of cells used for the study