INTERNATIONAL STANDARD

ISO 21243

First edition
2008-09-01

Radiation protection — Performance criteria for laboratories performing cytogenetic triage for assessment of mass casualties in radiological or nuclear emergencies — General principles and application to dicentric assay

Radioprotection — Critères de performance pour les laboratoires pratiquant le tri par cytogénétique en cas d'accident radiologique ou nucléaire affectant un grand nombre de personnes — Principes généraux et application aux dicentriques

Reference number
ISO 21243:2008(E)

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Published in Switzerland

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Foreword

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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 21243 was prepared by Technical Committee ISO/TC 85, *Nuclear energy*, Subcommittee SC 2, *Radiation protection*.

Introduction

The potential for nuclear and radiological emergencies involving mass casualties from accidental or malicious acts or terrorism requires generic procedures for emergency dose assessment to help the development of medical response capabilities. A mass-casualties incident is defined here as an event that exceeds the local medical resources. Biological dosimetry, based on cytogenetic analysis using the dicentric assay, typically applied for accidental dose assessment, has been defined in ISO 19238. Cytogenetic triage is the use of chromosome damage to evaluate and assess approximately and rapidly radiation doses received by individuals in order to supplement the clinical categorization of casualties. This International Standard focuses on the use of the dicentric assay for rapid cytogenetic triage involving mass-casualty incidents.

After a large-scale radiation emergency or malevolent act with involvement of radioactive materials, physicians are primarily concerned with preserving life and evaluating medical signs and symptoms for early treatment decisions. It is expected that patients have already been assessed clinically and triaged on the basis of any prodromal signs and symptoms of overexposure plus available information concerning their involvement in the incident. In this early-response phase of a radiological emergency, the initial purpose of cytogenetic triage is to rapidly estimate the dose for each referred patient to supplement this early clinical assessment.

The role of a secondary triage by cytogenetics is to confirm whether displayed symptoms can really be attributed to radiation rather than being a false positive response to some other cause. It is expected that the cytogenetic report be sufficiently informative to provide guidance to medical staff as they proceed to clinical management of the patients. This management can range from rapid identification of concerned but not radiation-exposed public (worried well), giving patients advice and reassurance before sending home lightly irradiated patients who do not need out-patient observation (i.e. dose below 0,5 Gy) or clinical intervention (i.e. dose below 1,0 Gy), through to active treatment of potentially life-threatening injury and optimized use of limited medical resources.

Several clinical triage systems have been developed where, based on severity of prodromal reactions, irradiated patients are allocated to one of 4 dose ranges (1 Gy to 2 Gy, 2 Gy to 4 Gy, 4 Gy to 6 Gy and $>$ 6 Gy) or acute-radiation-sickness (ARS) response categories (RC-01, RC-02, RC-03, RC-04) representing mild to very severe injuries. Enough experience with using clinical triage schemes (e.g., from Chernobyl) has been gained to show that the early sorting of persons into these dose or response category cohorts was adequate for the initial planning of the patients' management. However, as time progresses clinicians are looking for more accurate estimations of doses both in the low-dose range, where irradiated persons require counselling on risks of late stochastic effects, and also for higher doses, for anticipating the shorter-term sequelae of severe tissue reactions.

It should be noted that the initial clinical triage interprets the symptoms in terms of response to acute, more-or-less whole-body exposure. Protracted and fractionated exposures, of course, require higher doses in order to produce the same severity of responses.

It is expected that the cytogenetic triage achieve a rapid estimate of dose or response categories, quantitatively more precise than the four clinically derived categories, and also take account of any evidence that the exposure might not have been received acutely or involved the whole body. It is expected that the need for precision be set against the competing requirement for rapid results and it is necessary that this judgement be made at the time, depending on the anticipated number of patients, the surge capacity of the laboratory and the rate at which the blood samples are received at the laboratory.

Expert cytogenetic biodosimetry laboratories typically function to support national radiation-protection programmes and emergency-response schemes. Several of these reference cytogenetic biodosimetry laboratories have independently and successfully performed rapid dose assessment in actual and simulated mass-casualty incidents. Their approaches included pre-planning, reagent stockpiling, simplified sample processing, automation, as well as modifying some of the ISO 19238 scoring criteria. Recently, several of these national reference cytogenetic biodosimetry laboratories have also established networks of

supplementary, satellite cytogenetic laboratories, both nationally as well as internationally. Building upon their experience, this International Standard is intended to define criteria for performing quality-assured cytogenetic triage.

The primary purpose of this International Standard is to provide a guideline to all laboratories in order to perform the dicentric-bioassay - cytogenetic triage for dose assessment using documented and validated procedures. Secondly, it can facilitate the application of cytogenetic biodosimetry networks to permit comparison of results obtained in different laboratories. Finally, it is expected that laboratories newly commissioned to carry out the cytogenetic triage conform to this International Standard in order to perform the triage reproducibly and accurately.

This International Standard is written in the form of procedures to adopt for dicentric-bioassay - cytogenetic triage biological dosimetry for overexposures involving mass radiological casualties. The criteria required for such measurements usually depend on the application of the results: medical management when appropriate, radiation-protection management, record keeping and medical/legal requirements. For example, selected cases can be analysed to produce a more accurate evaluation of high partial-body exposure; secondly, doses can be estimated for persons exposed below the threshold for deterministic effects, by using the ISO 19238 criteria. These latter data also assist in counselling for the risk of late stochastic disease.

Part of the information in this International Standard is contained in other international guidelines and scientific publications, primarily in ISO 19238 and the International Atomic Energy Agency's Technical Report No.405, on Biological Dosimetry [4]. However, this International Standard details and standardizes the quality assurance and quality control of performance criteria for cytogenetic assessment of individual exposures in radiological or nuclear mass casualties. This International Standard is generally compliant with ISO/IEC 17025, with particular consideration given to the specific needs of rapid, emergency biological dosimetry. The expression of uncertainties in dose estimations given in this International Standard complies with the ISO Guide 98 and ISO 5725 (all parts).

Radiation protection — Performance criteria for laboratories performing cytogenetic triage for assessment of mass casualties in radiological or nuclear emergencies — General principles and application to dicentric assay

1 Scope

The purpose of this International Standard is to give an overview of the minimum requirements of process and quality-control components of the cytogenetic response for triage of mass casualties. Cytogenetic triage is the use of chromosome damage to evaluate approximately and rapidly radiation doses received by individuals in order to supplement the early clinical categorization of casualties. This International Standard concentrates on organizational aspects of applying the dicentric assay for operation in a triage mode. The technical aspects of the dicentric assay can be found in ISO 19238. This International Standard is applicable either to an experienced biological dosimetry laboratory working alone or to a network of collaborating laboratories (as defined in Clause 9).

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 19238, *Radiation protection — Performance criteria for service laboratories performing biological dosimetry by cytogenetics*

3 Terms and definitions

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

3.1
acute radiation syndrome or sickness
ARS
acute illness caused by irradiation of the entire body (or most of the body) by a high dose of penetrating radiation in a very short period of time (usually a matter of minutes)

3.2
associate laboratory
laboratory that has previously been validated and is prepared to be contacted for assistance when the capacity of the reference laboratory is exceeded

3.3
bias
statistical sampling or testing error caused by systematically favouring some outcomes over others

3.4
biological dosimetry
assessment of the absorbed dose of ionizing radiation using indicators found in biological material

3.5
confidence limits
CL
statistical range about an estimated quantity within which the value of the quantity is expected to occur, with a specified probability

3.6
chromosome
structure that carries genetic information

NOTE Normally, 46 such structures are contained in the human cell nucleus. During nuclear division, they condense to form characteristically shaped bodies.

3.7
cytogenetics
study of the structure of chromosomes

3.8
deterministic effect
effect from radiation that is absent below a certain threshold dose but its severity increases with the absorbed dose in human tissues due to ionizing radiation

EXAMPLES Cataract, radiation burn in the form of erythema or more serious local consequences, or acute radiation sickness/syndrome.

3.9
dicentric chromosome
chromosome aberrant in having two centromeres derived from the joining of parts from two broken chromosomes

NOTE A dicentric chromosome is generally accompanied by an acentric fragment.

3.10
dicentric assay
assay which measures radiation damage based on the frequency of dicentric or dicentric plus ring chromosomes found in metaphase cells

3.11
fractionated exposure
exposure to ionizing radiation that has been divided into small exposures separated in time

3.12
inhomogeneous exposure
exposure that is not received uniformly over the whole body or is received only by part of the body

3.13
inter-comparison
comparison between several laboratories on the accuracy and precision of their methods and dose estimates

3.14
intra-comparison
comparison within a laboratory on the accuracy and precision of their dose estimates (using different methods)

3.15
in vitro
technique performed in a controlled environment outside of a living organism

3.16
medical responders
professionals responding to an emergency situation who are dealing with providing medical care to the casualties

3.17
metaphase
stage of mitosis when the nuclear membrane is dissolved and the chromosomes are condensed to their minimum lengths and aligned for division

3.18
minimum detection level
MDL
smallest measurable amount (e.g. activity-concentration or dose) that can be detected with a probability of non-detection (type II error) while accepting a probability of erroneously deciding that a positive (non-zero) quantity is present in an appropriate background sample (type I error)

3.19
network
group of reference and associate cytogenetic laboratories trained and prepared to jointly respond to a large-scale radionuclear emergency requiring biological dosimetry

3.20
network laboratory
laboratory included in the network, both reference and associate

3.21
partial body exposure
exposure to ionizing radiation of a certain part of the body as opposed to the whole-body exposure

3.22
precision
dispersion of measurements with respect to a measure of location or central tendency

3.23
prodromal
⟨early signs and symptoms⟩ indicative of the imminent development of a disease or illness

EXAMPLES Erythema, nausea, vomiting.

3.24
protracted
⟨dose⟩ received over a long period of time

3.25
quality assurance
planned and systematic actions necessary to provide adequate confidence that a process, measurement or service satisfies given requirements for quality specified in a license

3.26
quality control
part of quality assurance intended to verify that systems and components conform with predetermined requirements

3.27
reference laboratory
laboratory primarily responsible for activating the network, communicating with emergency organizations and delivering the dose estimation results in an emergency situation

3.28
sequelae
condition resulting from prior injury or attack

3.29
stochastic effect
effect from exposure to radiation that has no threshold dose and is characterized by increasing probability of occurrence with increase of the dose

EXAMPLE Cancer.

3.30
triage
rapid process of sorting people depending on their need for immediate medical treatment (as is usually done in emergencies)

3.31
whole-body exposure
exposure to ionizing radiation of most of the body, involving the major part of hematopoetic tissues

4 Abbreviated terms

ARS Acute radiation sickness

MDL Minimum detection level

CL Confidence limits

5 Pre-planning

5.1 Awareness of the standard

It is necessary that local, state, and federal governments' health care providers and facilities be aware of the existence of the cytogenetic biodosimetry programme for individual dose assessment in radiological or nuclear mass casualties as established in this International Standard. This is critical for the laboratory to be able to receive blood samples promptly and thereby provide a rapid biodosimetry response within the time frame that is clinically useful in order to mitigate the acute health effects. Qualified laboratories and health care facilities should know their organization, roles and responsibilities, and the concept of operations in an emergency.

5.2 Roles and responsibilities of health care facilities

The health care facilities at the local, state, and/or the federal levels are responsible for the following:

a) evaluating the medical consequences for individuals exposed to radiation;

b) requesting qualified biodosimetry laboratories to provide individual dose assessments;

c) selecting the cohort of individuals who require biological dosimetry for immediate treatment, in consultation with qualified biological dosimetry laboratories;

d) obtaining informed consent from cases before requesting biological dosimetry assessment;

e) sampling blood for cytogenetic biodosimetry as soon as practical after exposure in specified blood-sampling tubes for cytogenetic biodosimetry; the health care facilities may request sampling kits from their respective national stockpiles or from a qualified cytogenetic laboratory, or use their own, if appropriate;

f) making arrangements to courier samples to the cytogenetic laboratory facility for dose assessment.

See Annex B for an example of an initial contact information form.

5.3 Roles and responsibilities of the biodosimetry laboratories

Each laboratory shall be organized and operate in such a way that upon receiving a request from the state/health care facility/hospital for biodosimetry response, they can rapidly and efficiently provide individual dose assessments. The laboratory's organization shall be clearly predefined and documented.

Qualified laboratories shall provide guidance to local, state and/or federal health-care facilities on

— the appropriateness of the biodosimetry assay,

— the laboratory's capabilities in order to select an appropriate cohort of individuals whose treatment can benefit from the cytogenetic biodosimetry.

Each laboratory shall be responsible for the following:

a) maintaining a stockpile of its own reagents or having immediate access to reagents and supplies from a local, state or national stockpile or commercial entity for receiving blood samples, culturing lymphocytes, preparing metaphase spreads and analysing samples for cytogenetic biodosimetry; these include general laboratory supplies as well as reagents and supplies specific to cytogenetic protocols;

b) maintaining established communication links with the local/state/federal health care facilities;

c) specifying and documenting the responsibilities, roles and interrelations of all personnel whose laboratory functions affect the quality of emergency biodosimetry response;

d) receiving appropriate samples, preparing and analysing samples, estimating dose, reporting and archiving samples or slides;

e) tracking, prioritizing (based upon rapid screening or input from physicians), determining the appropriate tests and reprioritizing as the tests progress, and reporting results;

f) knowing its maximal capability for samples processing (time versus number);

g) maintaining its own quality control and quality-assurance programme;

h) participating, as appropriate, in relevant educational, training and exercise programmes;

i) participating in periodic inter-comparison studies;

j) maintaining a safety plan; the laboratory head shall define written safety procedures for protection against viral, microbial, chemical and optical hazards.

6 Communication and information

6.1 Biological dosimetry request and confidentiality

Biological dosimetry investigations made by reference and/or associate laboratories shall be undertaken in accordance with the national regulations regarding confidentiality. This normally includes the maintenance of confidentiality of the patient's identity, medical data and social status.

This requirement extends to

a) written, electronic or verbal communications between the laboratory and the person/organisation requesting the analysis and receiving the report,

b) protection of confidential information held within the organization where the laboratory is located,

c) electronic record management.

Users with different access restrictions should have different privileges within the system. The laboratory chief assigns rights and access restrictions to the rest of the laboratory.

The laboratory head shall have established protocols for maintaining the anonymity of samples. To avoid the identification of the patient while guaranteeing the traceability of the analysis, the blood samples should be coded upon arrival in the laboratory. The coding is performed in an unambiguous way according to a standard procedure. The same code shall be used for all the stages of the analysis. The code is assigned by an authorized person. The decoding, interpretation of results and compiling of the report shall also be performed by an authorized person. If it is required to share a sample, the same code shall be used by all associate laboratories and for communication between them.

6.2 Educational programme — Formation, training and exercises

The laboratory should have clearly defined educational and training programmes addressing the following:

a) qualification of staff employed;

b) training in cytogenetics, radiation cytogenetics, analysis of radiation-induced structural chromosomal aberrations, biological dosimetry, good laboratory procedures, human-use protocols and general laboratory safety; training should also cover laboratory instrumentation and standard operating procedures.

The laboratory head is responsible for maintaining the performance criteria and the qualifications of the individual scorers. All scorers shall participate in periodic intra- and inter-laboratory comparisons.

7 Biological dosimetry process in radiological or nuclear mass-casualty incidents

See Annex A for a flow diagram indicating the interactions between physicians and biological dosimetry laboratories.

8 Emergency response of the reference laboratory

The head of the laboratory should have a prepared emergency response plan, so that all members of the staff know their role.

The biodosimetry assay for use in the emergency procedure shall be decided before processing the blood samples. This decision is based on information about the accident that is available to the head of the laboratory and discussions between the laboratory and other medical personnel involved in the management of the emergency situation. By default and/or without additional information, the dicentric assay is the method-of-choice.

The laboratory processes the blood samples in the order of arrival. Where possible, priority is given to microscopic analysis in the most severe cases. Generally, blood samples from the most seriously irradiated persons based on clinical symptoms and/or physical dosimetry (when such information is available) are prioritized.

The biological dose estimate is obtained from appropriate calibration curve(s) pre-defined by the network.

The results of biological dosimetry should be made available as soon as possible.

According to the country and the emergency situation, blood samples either

— arrive directly at the laboratory, where all of the processing for cytogenetic analysis occurs; or alternatively,

— are initially processed in a specialized associate laboratory in, for example, a hospital or research institute that is already network linked to the central reference laboratory.

9 Design of laboratory network

9.1 Overview

A cytogenetic biodosimetry laboratory network may consist of a reference laboratory with associate laboratories as well as a consortium of reference laboratories.

The constitution of the laboratory network is based on a voluntary and consensual participation of expert laboratories qualified in the selected cytogenetic techniques.

The number of associate and/or reference laboratories in a network depends on each country. The associate and/or reference laboratories can be both from within as well as outside that country. The reference laboratory is primarily responsible for network activation. The associate laboratories are called on for collaboration by the reference laboratory when the assignment is above its workload.

In an emergency situation, when the decision to activate the network is taken, the reference laboratory in the country where the radiation incident occurs becomes the nucleus for communication among the network.

The reference laboratory is primarily responsible for communication with emergency organizations and for issuing dose-estimation results.

In cases where the event takes place in a country lacking a biodosimetry laboratory, the emergency officials may contact any of the laboratories in the network and that laboratory will become the reference laboratory.

9.2 Preparedness of the laboratory network

As soon as the network is constituted, the following items shall be clearly defined:

a) response of the network to the emergency situation in terms of techniques used, countries/laboratories activated and data interpretation;

b) logistic organization planning, starting with how the blood-sample collection is organized in each country and then efficiently transported to the other countries/laboratories activated by the network;

c) standardization of techniques involved in order to pool data in a combined response to a large-scale radiological emergency;

d) information exchange and training organization between network staff, for all scientific and technical questions related to the network tasks.

All these parameters shall be established and maintained through meetings of consensus, which are organized as appropriate by the reference laboratory and at least every two years.

The network shall make its existence and capabilities known to national front-line emergency-response organizations of each country in which a laboratory of the network is involved, and in particular, inform the physicians in charge of the patients with whom the network directly liaises. Furthermore, professionals in other countries that are not participating in this network should also know what to do in case of a large-scale incident and how to activate this network.

The laboratories of the network prepare and circulate to the associate laboratories the following documents:

— an information sheet containing full contact details (addresses, phones, telefax, e-mail) of the laboratory staff who are involved in the network; this shall be updated as necessary;

— a detailed information sheet, in the language of the country, for medical staff that carry out blood sampling; this includes contact information, specification of sample size, correct anticoagulant and packing/shipping requirements and shipment tracking information;

— detailed laboratory protocols used for the cytogenetic analysis, microscope analysis and data interpretation.

Each laboratory always maintains sufficient stocks of necessary consumables or an immediate access to a national or regional stockpile to be able to process at least 100 blood samples at short notice.

Stocks of specimen tubes together with information sheets shall be stored in the laboratory and/or deposited in appropriate locations in the country.

The network organizes periodically an inter-comparison of the detailed laboratory protocols used to ensure the consistency of their analytical procedures and dose estimates. This also can include developing a panel of reference microscope slides that can be used for evaluating individual scorers when there are staff changes and also when other laboratories join the network.

9.3 Laboratory network operation

The reference laboratory responsible for the dose estimation calls for collaboration of network laboratories when the number of cases for examination is above its workload.

When the decision to activate the network is made, the reference laboratory becomes the nucleus for communication with the network. The reference laboratory informs the partners of the circumstances of the incident and together they establish the extent of cooperation needed.

Cytogenetic examination for dose estimation is performed at the request of physicians. Selection of cases for examination is made by discussion among experts in cytogenetic dose estimation, scene managers and physicians.

The reference laboratory and the network laboratories discuss the details of work sharing in biodosimetry.

An informed written consent shall be submitted by each individual or a treating physician, as applicable, prior to taking blood. Special care shall be taken to protect privacy throughout the assignment.

The reference laboratory organizes the blood sampling and dispatching of specimens to the partners, when and if possible, or designates another suitable laboratory to be responsible. To minimize delay, this can entail that the local/regional hospital dealing with the patients dispatches the blood specimens directly to the foreign network laboratories. An alternative procedure may be adopted by the reference laboratory, by which it processes the specimens and then sends fixed cell suspensions or microscope slides to the network partners. These alternatives are decided by a discussion among the partners.

It should also be decided among the partners whether the scoring of cytogenetic indicators is performed in each laboratory or the local laboratory just helps with culturing and then giving fixed cells to the experts.

The results of scoring (and sometimes also dose estimation) are reviewed by more than one laboratory and dose estimation for each person is made based on the reviewed results.

The associate laboratories send the reference laboratory the raw data and the aberration-distribution data. They also send the dose estimates, adjusted when necessary for dose protraction or heterogeneity, obtained from their own calibration curve and most appropriate for the type of radiation involved.

The reference laboratory receives the results from the network partners and acts as the central point of communication/liaison with the physicians.

Following a review with the medical staff, some patients may be selected for increased cell scoring in order to improve statistical uncertainties on dose estimates and better discrimination of inhomogeneous overexposure. Such further examination shall be made in accordance with the performance criteria described in ISO 19238.

10 Expected results

10.1 General

The aim for cytogenetic triage shall be to achieve rapidly a quantitatively more precise estimate of dose than the initial sorting based on clinical prodromal sickness and also to take account of any evidence that the exposure might not have been received acutely or involved the whole body.

The need for precision shall be set against the competing requirement for rapid results; this judgement shall be made at the time, depending on the anticipated number of patients, the surge capacity of the laboratory and/or the network and the rate at which the blood samples are received from the front-line medical services. See Annex C for a guidance table.

10.2 Whole-body exposure

Unless there is evidence to the contrary, the cytogenetic data should be converted to doses on the basis that the exposure was a single acute event. "Acute," in terms of cytogenetics, means received in less than 30 min. The dicentric yield, Y, should then be referred to an *in vitro* dose response curve for an appropriate quality of radiation, as given in Equation (1):

$$Y = c + \alpha D + \beta D^2 \tag{1}$$

where

D is the dose absorbed;

c is a constant (background noise);

α and β are linear and quadratic constants.

If there are sufficient grounds to believe that continuous exposure was protracted beyond 30 min and/or fractionated with break(s) in exposure of more than about 15 min, the dicentric data should be interpreted with a time-dependent G-function correction to the β yield coefficient (see IAEA, 2001). Simple linearity, as given in Equation (2), can be assumed where the duration of continuous exposure is $>$ 24h and also for intermittent irradiations, provided that no dose fraction is received acutely:

$$Y = c + \alpha D \tag{2}$$

Doses shall be calculated by reference to the dose response curve and uncertainties, expressed as 95 % confidence limits (CL) on the dose, and shall be calculated only from the statistical uncertainty on the scoring data from the patient. Thus, standard errors on the curve coefficients shall be ignored.

The laboratory shall aim to score 50 metaphases per patient or to stop sooner if 30 dicentrics have been found. The laboratory should prepare, in advance, tables or a computer program based on their own appropriate dose response curve that shows the numbers of dicentrics observed in 50 cells (or 30 dicentrics in fewer than 50 cells), and calculates the resultant dose estimates and the associated statistical errors.

Annex D shows such a table, constructed using a generic, acute γ-ray curve, with coefficients typical of values published by several leading laboratories as given in Equation (3):

$$Y = 0{,}001 + 0{,}02D + 0{,}06D^2 \tag{3}$$

Doses and CLs have been calculated for acute exposures using the full equation, Equation (1), and for chronic exposures without the quadratic term, Equation (2). For simplicity, not all of the 80 possible dicentric frequencies are illustrated. It should be stressed that the table shown in Annex D is an example. It shall not be used as a substitute for calculations made by a laboratory with its own calibration data.

10.3 Inhomogeneous exposure

Inhomogeneous exposure can be detected from an examination of the distributions of dicentric aberrations among the scored cells. It is characterized by over-dispersion with respect to the Poisson distribution that occurs with homogeneous exposure (see IAEA, 2001). Protracted inhomogeneous exposure is smoothed out by the movement and circulation of lymphocytes in the body and, therefore, it is feasible to detect inhomogeneity reliably only in a triage situation where the exposure is acute.

The laboratory shall have available the necessary computer program in order to analyse the distributions of the dicentrics among the 50 or fewer cells scored, by calculating the ratio of variance to mean, σ^2/y, and using the unit mean deviate test (u-test) to determine whether the distribution departs significantly from the Poisson distribution. There are two methods available: the contaminated Poisson and the Qdr, to interpret dose distributions that deviate from the Poisson form. For both, it is necessary to make a simplifying assumption that the inhomogeneity consists of a part-body exposure with one irradiated fraction having been irradiated uniformly. The calculations (see IAEA, 2001) should be made with the laboratory's own acute-dose response curve and the cell survival parameter, D_0, equal to 3,0 Gy. This, then, provides an estimate of the size of the irradiated fraction of the body and its average dose.

It is unlikely that localized doses below ~ 3,0 Gy require clinical treatment for managing late tissue reactions (deterministic injuries) but, for patients with estimated doses higher than this, there is an urgent need for more precise data to better inform the clinicians. Such patients constitute high-priority subjects for increasing the number of cells scored in order to give better statistical discrimination of their inhomogenous irradiation. One useful parameter in such cases is a simple calculation of the percentage of undamaged metaphases observed (i.e. without a dicentric, a centric ring or a fragment). These are indicative of unexposed cells and, by implication, un-irradiated parts of the body that can include active bone marrow.

11 Quality assurance and quality control

11.1 Overview

As a minimum, the quality-assurance and quality-control practices cited in 11.2 and 11.3 apply to the network laboratories performing cytogenetic assessment of individual exposures in radiological or nuclear mass casualties. If the reference laboratory enlists the services of other laboratories, either as a contracting or a sub-contracting alternative for expediting any portion of this service, the reference laboratory shall maintain total oversight of the quality-assurance and quality-control responsibility for all participants.

11.2 Quality control

11.2.1 General

Performance checks shall be conducted to ensure the conformance of analytical processes, measurement equipment and procedures and the facilities to predetermined operational requirements.

11.2.2 Quality control procedures

The laboratory shall verify that the estimation of absorbed-dose measurements complies with the accuracy requirements. Procedures should include quality control performance checks on the following:

a) measurement systems and use of traceable reference standards;

b) review of procedures, specifications and operating logs;

c) observation of operations and evaluation of quality-control data to ensure the long-term consistency of analytical results;

d) evaluating conformance to the performance criteria of this International Standard;

e) verification of minimum detection levels.

11.2.3 Performance checks of sample transport integrity

In many cases, blood collection occurs at sites distant from the processing laboratory and transportation is necessary. A minimum-maximum thermometer in the shipping container provides information on the temperature range during transport. To minimize temperature changes, it is recommended that samples be shipped in styrofoam containers containing room-temperature gel-packs. If air transportation is used, avoid X-irradiation at the security checkpoints. For international transport, the appropriate permits shall be obtained in advance and be included in the shipment to avoid delays at customs. All details concerning blood collection and storage should be recorded. Because of the risk for infectious diseases (hepatitis, HIV), appropriate precautions shall be followed when handling the blood samples. See Annex E for an example of instructions for customers.

The delay between blood sampling and arrival at the laboratory should not exceed 48 h.

Only blood samples in a solution containing lithium heparin are acceptable for dicentric assay.

11.2.4 Performance checks of sample integrity by the laboratory

A system for recording the collection and storage of the blood samples should be established by the laboratory receiving the samples so that sample integrity is guaranteed. If it is necessary that one biological dosimetry laboratory send some blood samples to other laboratories, the sender is responsible for the shipping and shall follow the same rules as in 11.2.3.

The use of coded samples is critical for medical confidentiality. This coding scheme should also be used for communications between laboratories.

11.2.5 Performance checks of instrumentation

The essential instruments and apparatus should be clearly identified. They should be calibrated and controlled periodically.

11.2.6 Performance checks of sample protocol

The culture, fixation and staining procedures shall be described in detail in the quality handbook. It is recommended that the same batch of media and reagents be used throughout the study. The composition of all reagents shall be described as accurately as possible in the quality handbook.

11.2.7 Performance checks of sample scoring

Uniform criteria for scoring shall be used. Scoring shall be performed by trained and experienced observers.

The ability of each observer to perform the assay should be controlled periodically. The laboratory head is responsible for maintaining the scoring criteria and the qualifications of the individual scorers in accordance with the criteria defined in the ISO 19238. The list of qualified scorers should be held by the network.

The microscope slides and fixed cell suspensions should be stored such that their quality is preserved for several years.

11.2.8 Performance checks of dose and confidence limits estimation

The dose-effect calibration curve should be well documented, i.e. radiation dose, radiation quality, dose rate, number of patients used to set up the curve, number of cells scored and statistical method of curve fitting.

The way uncertainties are calculated should also be clearly documented for the dose effect curve and for the samples analysed.

11.2.9 Performance checks of the generation of reports results

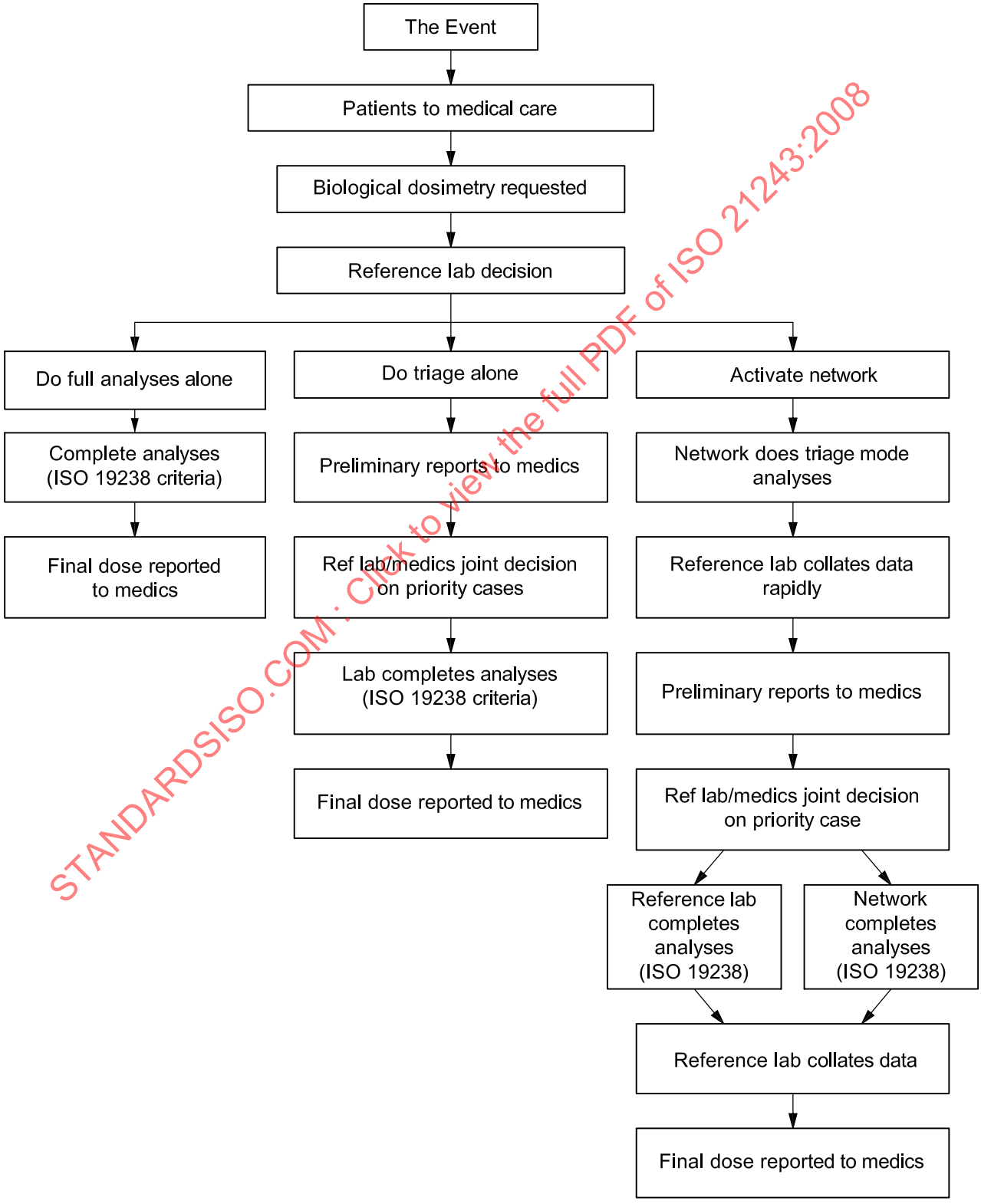
The study reports to customers shall be examined to ensure that they contain the necessary information defined in this International Standard, namely subject and customer identifiers, exposure information, sampling dates, the scoring results, the interpretation of the results in terms of dose and its uncertainty and information on how this was derived. See Annex F for an example of a group sample report.

11.2.10 Performance of the network

Samples containing known exposures of specific radiation doses and quality of interest shall be analysed to determine bias and precision of the analytical procedures, in each reference laboratory and within the network as well. Replicate samples should also be processed periodically. Statistical techniques, such as quality control charts, shall be used to evaluate biological dosimetry cytogenetic procedure performance data.

Annex A
(normative)

Interactions between physicians and biological dosimetry laboratories



Annex B
(informative)

Initial contact information form

A form of this type can be used for initial contact informing the laboratory of the event, requesting information about the assay, and identifying the requirements for sample collections.

Date/Time call received: _____

Person receiving the call: _____

Contact person/establishment: _____

_____

Telephone number for the contact person: _____

Fax number for the contact person: _____

e-mail for the contact person: _____

Send the preliminary reports to: Name: _____

Fax #: _____

e-mail: _____

Send final report to: Name: _____

Fax #: _____

Address: _____

_____

_____

Are sample kits required: Yes: _____ No: _____

If yes, how many: _____

Is prioritization of samples possible? Is medical/physical information available or will it be available?

_____

_____

_____

Summary of communications:

_____

_____

_____

_____

Annex C
(informative)

Guidance for threshold of detection

This annex provides guidance on the minimum number of spreads that need to be prepared and analyzed in the event that many people may require radiological exposure assessment. In addition, the time required for conducting the analysis is estimated using the assumptions described below.

Table C.1 — Minimum number of analyses and minimum time required as a function of number of samples

Number of samples [a]	Metaphases to score [b]	Dicentrics to score [c]	Maximum total spreads	Total scoring time	
				h	days [d]
⩽ 10	500 to 1 000	100	500 to 10 000	—	—
10 to 25	50	30	500 to 1 250	29 to 71	0,6 to 1,5
25 to 50	50	30	1 250 to 2 500	71 to 142	1,5 to 3
50 to 75	40	30	2 000 to 3 000	117 to 175	2,5 to 3,7
75 to 100	30	30	2 250 to 3 000	138 to 184	2,9 to 3,9
100 to 150	20	30	2 000 to 3 000	134 to 200	2,8 to 4,2
⩾ 151	20	30	> 3 020	> 202	> 4,3

[a] It is assumed that more than 10 samples is a triage scenario.

[b] It is assumed that the analysis time for the metaphase is 3 min.

[c] It is assumed that scanning and set-up time per slide is 20 min.

[d] The total working days is calculated as 24 h/working day, with 3 shifts of 8 h, 2 people/per shift and 2 metaphase finders.

Annex D
(informative)

Estimates of dose and 95 % confidence limits for selected observations of numbers of dicentrics and cells

Table D.1 — Estimates of dose as a function of the numbers of dicentrics and cells

Dicentrics	Cells	Acute exposure [a] Gy			Chronic exposure [a] Gy		
		LL [b]	Mean	UL [c]	LL [b]	Mean	UL [c]
0	50	0	0	0,9	0	0	3,6
1	50	0	0,4	1,2	0	1,0	5,5
2	50	0,1	0,7	1,4	0,2	2,0	7,2
3	50	0,3	0,8	1,5	0,6	3,0	8,7
4	50	0,4	1,0	1,7	1,0	4,0	10,2
5	50	0,6	1,1	1,8	1,6	5,0	11,6
10	50	1,1	1,7	2,3	4,8	10	18
15	50	1,5	2,1	2,7	8,4	15	25
20	50	1,9	2,4	3,0	12	20	31
25	50	2,2	2,7	3,3	16	25	37
30	50	2,4	3,0	3,6	20	30	43
30	45	2,6	3,2	3,8	22	33	48
30	40	2,7	3,4	4,1	25	37	53
30	35	2,9	3,6	4,4	29	43	61
30	30	3,2	3,9	4,7	34	50	71
30	25	3,5	4,3	5,2	40	60	85
30	20	3,9	4,8	5,8	51	75	110
30	15	4,6	5,6	6,7	67	100	140
30	10	5,8	6,9	8,3	100	150	210
30	5	8,0	10	12	200	300	430

[a] The data in the table are calculated using a generic acute gamma-ray curve using the coefficients given in Equation (3).

[b] LL: Lower confidence limit.

[c] UL: Upper confidence limit.