Participate in the RENEB inter-laboratory comparison exercise 2021

Cytogenetic assays for biological dosimetry: dicentric chromosome assay [DCA] and cytokinesis-block micronucleus assay [CBMN] following telomere and centromere hybridization

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Goal

RENEB 2021 inter-laboratory comparison of three blinded samples exposed to x-rays had the purpose of simulating an emergency scenario in which early dose estimates are required for immediate medical management support. The participating labs were asked to provide fast analysis in a triage mode and report results in a quickly manner.

Methods

Single male donor (age 32)

2.6 mL of Lithium-heparinized whole blood / sample

3 blind doses (Gy) (x-rays)

- **0.0 (low exposed)**
- **1.2 (medium exposed)**
- **3.5 (highly exposed)**

2 Endpoints:

Cytokinesis-block Micronucleus Assay

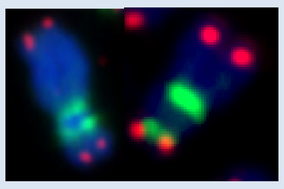
- ✓ 14 Laboratories
- ✓ 13 different countries
- ✓ 1000 BN cells in semi-automated mode

Dicentric Chromosome Assay

- ✓ 33 Laboratories
- ✓ 22 different countries
- ✓ 150 metaphases scored semi-automatically

Dicentric Assay with Telomeres and Centromeres FISH improves Chromosomal Aberrations Detection

Giema staining

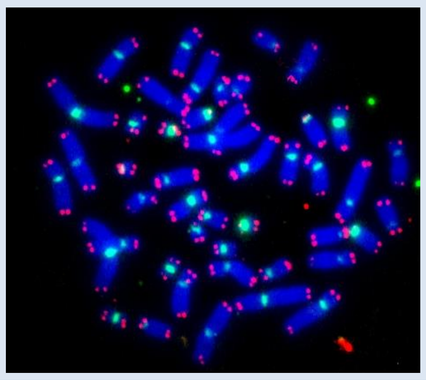


Underestimated dicentrics

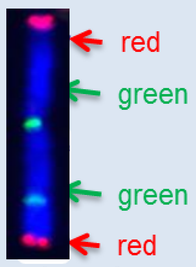
Evaluation of chromosomal aberrations :

DIFFICULT requiring a high level of expertise

TC staining



Fish PNA

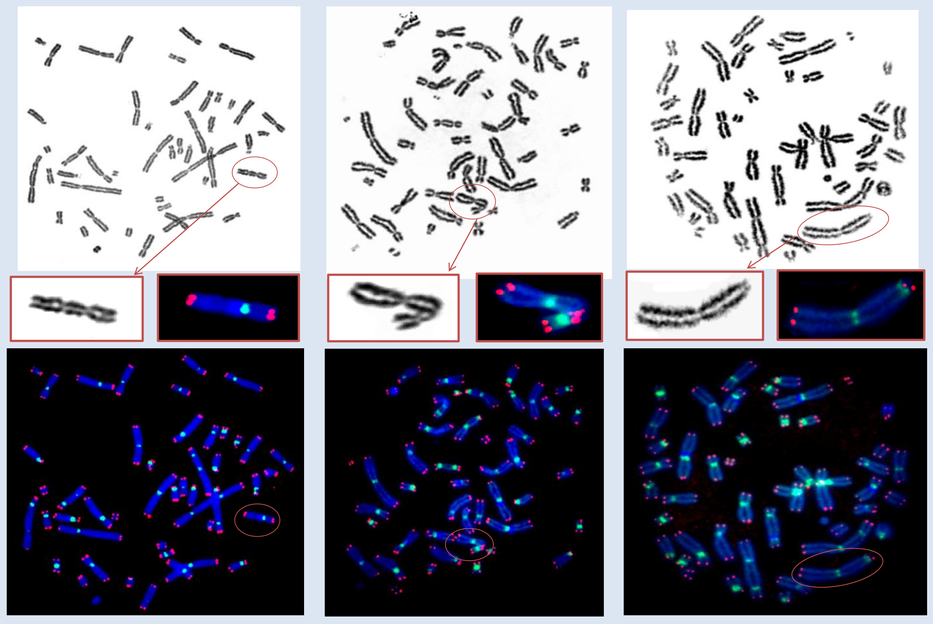


Dicentric

Evaluation of chromosomal aberrations :

EASY not requiring a high level of expertise

Improvement of chromosomal aberration detection

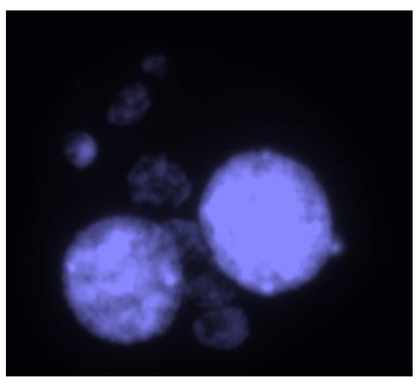


Dicentric chromosome assay was performed using telomere and centromere staining for chromosomal abnormalities scoring, allowing the improvement of the technique compared to uniform staining generally used by other participating laboratories (Giemsa or DAPI), mainly due to an underestimation by conventional techniques of the dicentric having a centromere and a telomere adjacent or with very close two centromere.

M'kacher R, El Maalouf E, Ricoul M, Heidingsfelder L, Laplagne E, Cuceu C, Hempel WM, Colicchio B, Dieterlen A, Sabatier L. Mutation Research/ Fundamental and Molecular Mechanisms of Mutagenesis 770 (2014) 45–53

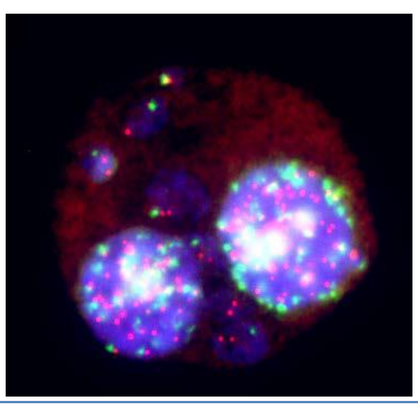
Micronucleus Assay following FISH with Telomere and Centromere

Uniform staining

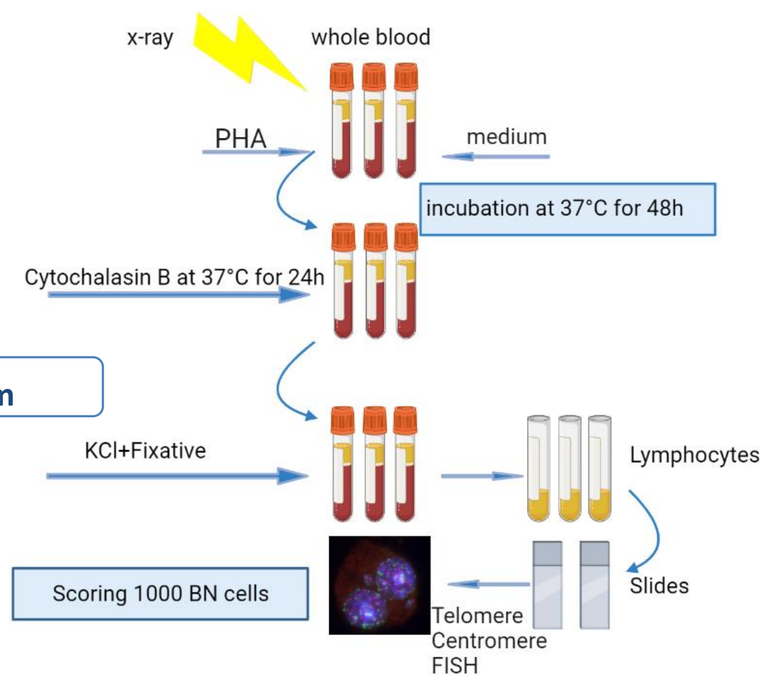


DIFFICULT to detect cytoplasm

TC staining

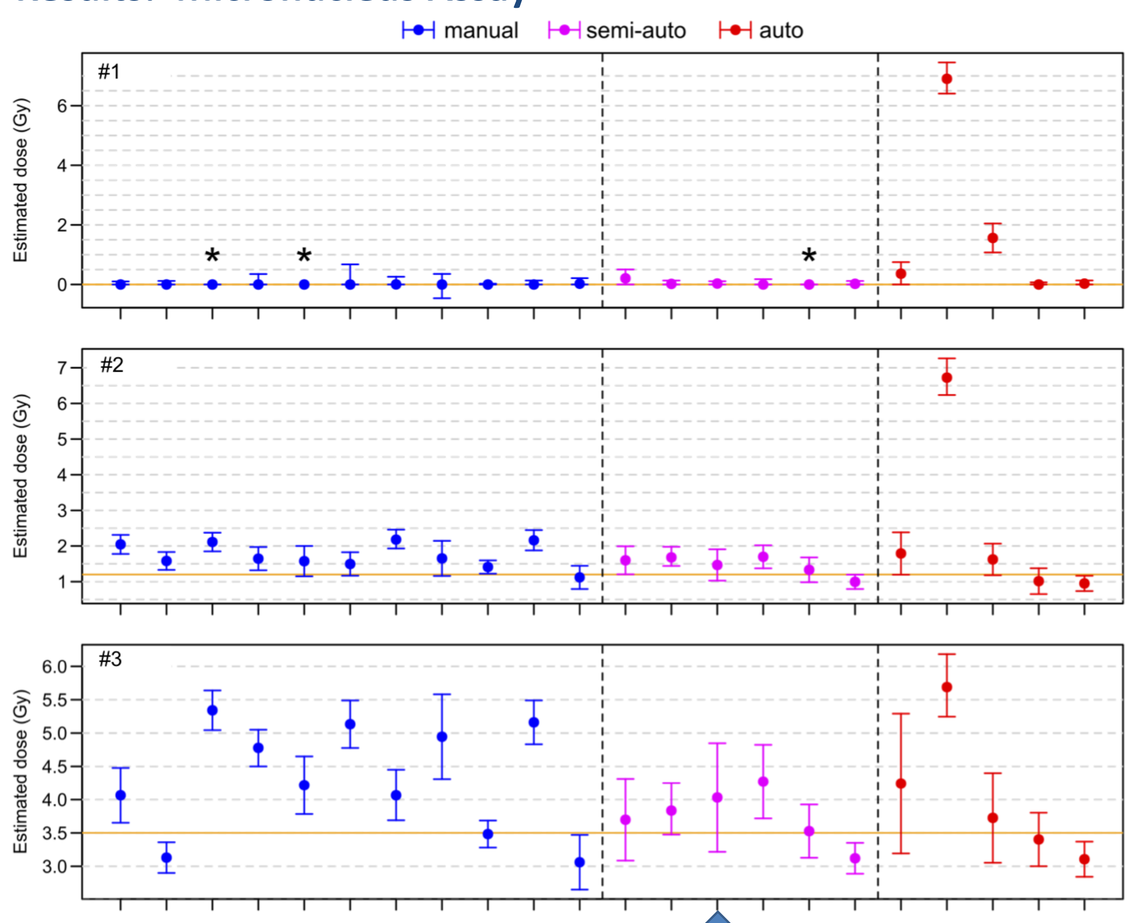


EASY to detect cytoplasm



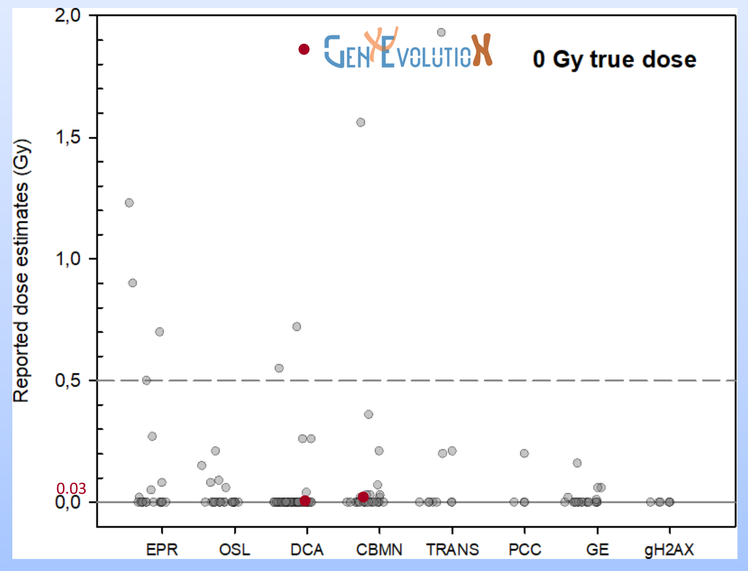
Telomere centromere FISH staining allows discrimination between clastogen or aneugen effect

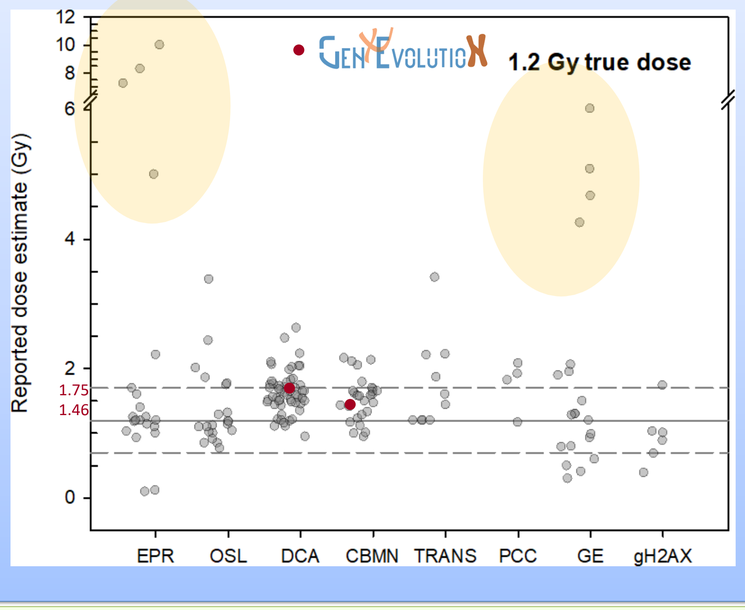
Results: Micronucleus Assay

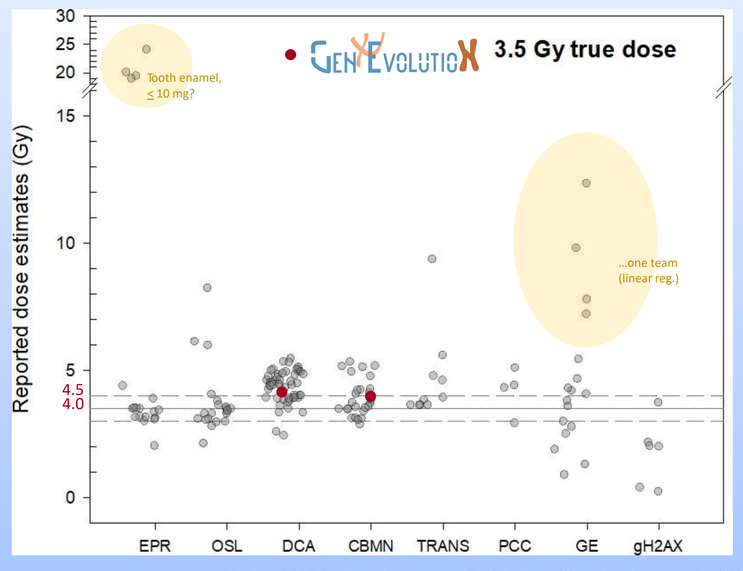


Vral A et al. RENEB Inter-Laboratory Comparison 2021: The Cytokinesis-Block Micronucleus Assay, in preparation June 2023

Reported dose estimates from all participants including DCA and CBMN Assays







Matthias PORT, Bundeswehr Institute of Radiobiology affiliated to the University of Ulm, RENEB inter-laboratory comparison 2021, study on biological and physical dosimetry employing eight assays

Conclusion

Both The CBMN assay and the DCA assay allowed classification of samples in the correct triage categories. These new approaches can be used for biological dosimetry in radiation emergency medicine and to achieve high sample throughput for the processing of large cohorts of exposed populations to genotoxic agents and their follow-up.

Acknowledgement

project RENEB ILC 2021

Contact

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