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All happy families resemble each other, each unhappy family is unhappy in its own way

LEO TOLSTOY, *Anna Karenina*

We can paraphrase this: Every radiation accident is unique and it is not possible to predict how the next accident will look like

What do we do if there is a large event with thousands of people potentially exposed?
First question: who was exposed?

TODAY'S WORLD:
➡ Expansion of use of nuclear power and use of radiation sources
➡ 21st century threats

Multi-disciplinary biodosimetric tools to manage high scale radiological casualties MULTIBIODOSE

Were they exposed to radiation?

Aim: to analyse a variety of biodosimetric tools and to adapt them to different mass casualty scenarios.

The main objective is to develop triage tools. Consequently, we do not aim at PRECISION of dose-estimate, but at SPEED of performance.

www.multibiodose.eu

The partners

Consortium Member institutions:

Stockholm University	Stockholm University (SU), Sweden
Bundesamt für Strahlenschutz	Bundesamt für Strahlenschutz (BfS), Germany
UNIVERSITEIT GENT	Universiteit Gent (UGent), Belgium
Health Protection Agency	Health Protection Agency (HPA), United Kingdom
IRSN	Institut de Radioprotection et de Sûreté Nucléaire(IRSN), France
	Istituto Superiore di Sanità (ISS), Italy
Statens strålevern	Norwegian Radiation Protection Authority (NRPA), Norway
STUK	Radiation and Nuclear Safety Authority (STUK), Finland
WESTLAKES	Westlakes Scientific Consulting (WSC), United Kingdom*
UAB	Universitat Autonoma de Barcelona (UAB), Spain
inct	Institute of Nuclear Chemistry and Technology (INCT), Poland
HelmholtzZentrum münchen	Helmholtz Zentrum München (HMGU), Germany
	Bundeswehr Institut für Radiobiologie in Verbindung mit der Universität Ulm (BIR), Germany
OXFORD	Gray Institute for Radiation Oncology and Biology, University of Oxford (UOXF), United Kingdom
EURADOS	European Radiation Dosimetry Group (EURADOS), European network registered in Germany

*) Former member. In August 2010 the Westlakes Research Institute ceased to exist and, consequently, left the consortium.

CONTACT INFO

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Stockholms universitet

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FP7 Security

Duration:
05.2010 – 04.2013

The biodosimetric tools

The dicentric assay

The automated micronucleus assay

The gamma H2AX foci assay

The skin speckle assay

The serum protein assay

**The mobile phone as
a dosimeter**
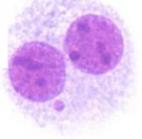
     **display glass – EPR
     semiconductors – OSL**
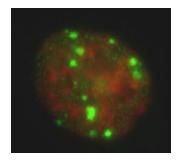

Glass

Electronic
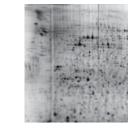components

Chip cards   Plastics

The concept of MULTIBIODOSE
The aims are reflected in the structure of each task

No	Task	2	4	6	8	10	12	14	16	18	2
		Year 1						Year			
WP1	Dicentric assay										
1.1	To validate the conventional dicentric assay and provide training										
1.2	To validate the automatic dicentric scoring and provide training										
1.3	To validate telescoring of dicentrics and provide training										
1.4	To compare the dicentric assay with other biodosemeters										
1.5	To improve the performance of the Metasystems scoring tools										
1.6	To develop a future training programme for the dicentric assay										

A repeating pattern…
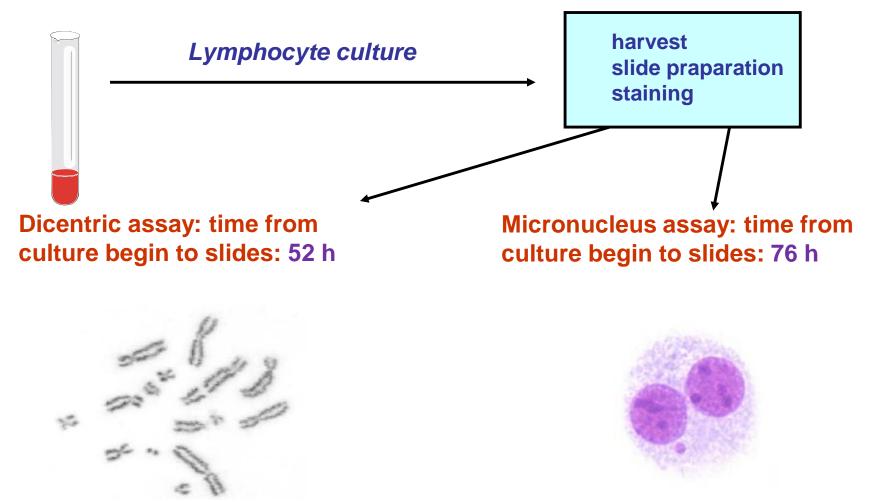
May 2012

- To validate
- To improve/develop
- To compare with other assays
- To automate/ improve performance/ pursue commercialisation
- To develop a training programme

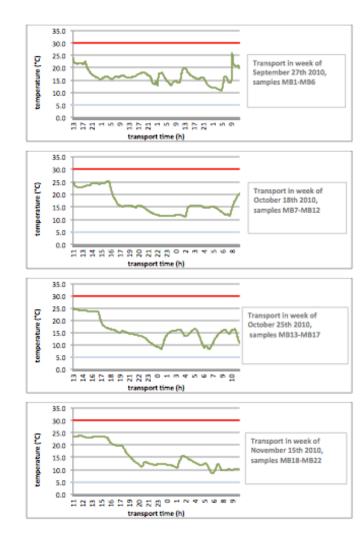# Some remarks about the speed of analysis of dicentrics and micronuclei in lymphocytes

Lymphocyte culture

harvest
slide praparation
staining

Dicentric assay: time from culture begin to slides: 52 h

Micronucleus assay: time from culture begin to slides: 76 h

Sending blood samples between laboratories

Transports of irradiated blood samples

- Transport by DHL

- Biological material code: UN 3373 BIOLOGICAL SUBSTANCE CATEGORY B

- 8 transports, problems with 1 transport => packages were lost !!

- TLD readings were generally 0 Gy with highest dose recorded 0.5 mGy, which can be neglected compared to the doses administered *in vitro*.

- Temperature loggers (see figures): temperatures during transport remained between 5-30 °C

Transport in week of September 27th 2010, samples MB1-MB6

Transport in week of October 18th 2010, samples MB7-MB12

Transport in week of October 25th 2010, samples MB13-MB17

Transport in week of November 15th 2010, samples MB18-MB22

WP 1 Dicentric chromosome assay

1. Conventional dicentric assay	2. Automation	3. Telescoring
Month 1 to 12	Month 13 to 24	Month 25 to 30
Cell culture (standard)	Scoring criteria	Scoring criteria
Conventional scoring	Cell culture (24h colcemide)	Conventional scoring
Dose estimation	Dic candidate scoring	Quick scoring
Triage validation & Training	Dose effect curve	

WP 1: current conclusions regarding the automated analysis

➢ The two classificators give comparable results

➢ The dicentrics follow poison distribution (dose effect curves)

➢ Semi-automated scoring of dicentric candidates gives acceptable results

➢ Semi-automated scoring detects inhomogenous exposures (u-test)

➢ Semi-automated scoring can be used in a large scale accident as pre-screening

The analysis of 1 sample (150 cells) can be perfomed in 25 minutes and needs only few human interaction

Metaphase finding:	5 min
Auto Capturing of 150 cells:	17 min
Automatic scoring :	1 min
Evaluation of dic candidates:	2 min
Total:	25 min
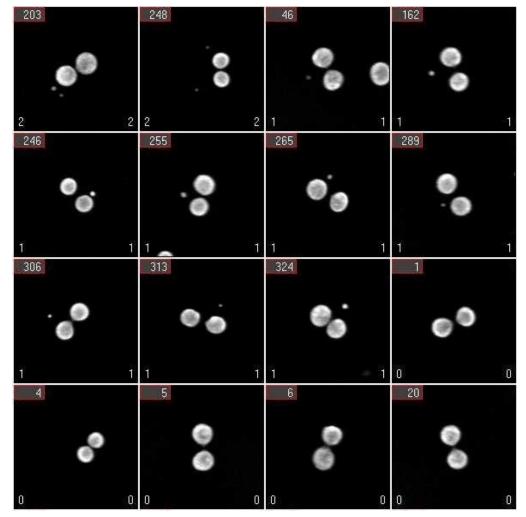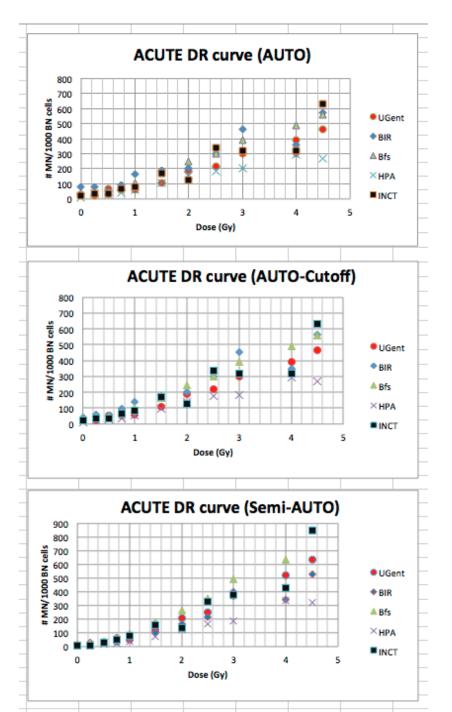
WP2 Automated micronucleus assay

Scoring is performed at low optical resolution.

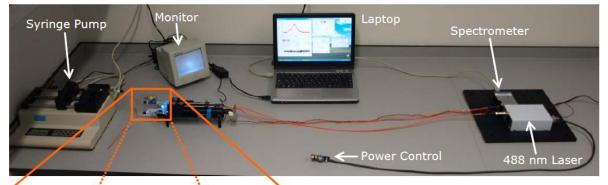**2000 cells can be scored within 8 minutes.**

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General conclusion

The combination of the spontaneous MN and the MN dose response data obtained within this work package point to the superiority of the semi-automated scoring procedure for large scale biodosimetry in a multicenter setting to manage high scale radiological casualties.
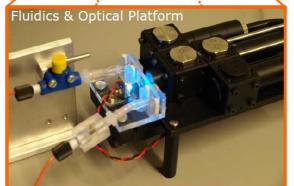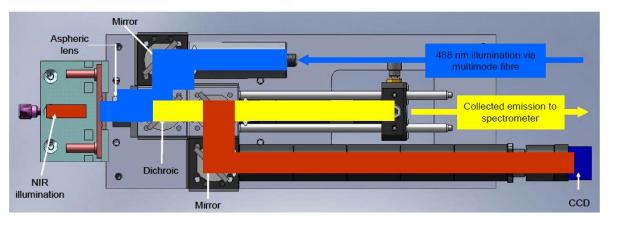
WP 3 Gamma-H2AX assay
WP leader: HPA

Image of two, 10 μm, beads displayed on the monitor screen. The blue circle represents the focused laser spot size.

Image of the laser beam profile revealed using a cuvette containing a green fluorescent dye.

Portable fluidic system

Results highly variable Not suitable for on-site triage

Metafer/MetaCyte-based foci analysis

Issues:
- Microscope objectives (x40 or x63)
- Compatibility between Metafer versions (3.2.2 vs 3.5 vs 3.8.4)

General conclusion

General problem of the assay: weak signal stability but doses can be detected until 24h post radiatio.

With uniform Standard Operational Procedures (SOP) for blood handling and image analysis settings good reproducibility can be achieved.

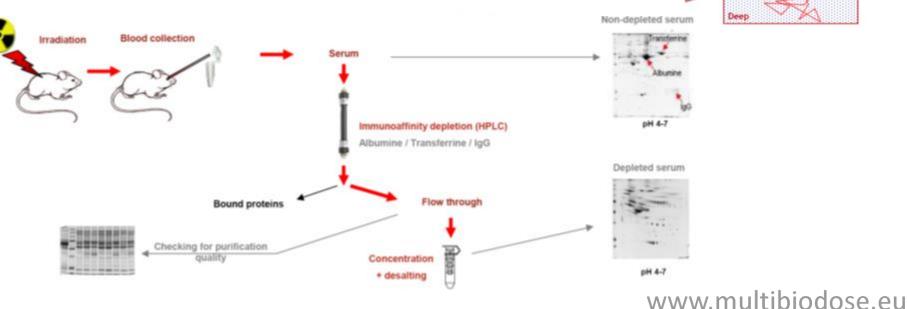**Time of analysis from blood collection to results: ca 2-3h.**

WP4: Skin Speckle Assay (SSA) and Serum Protein Assay (SPA)

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WP leader: IRSN

SSA

Main aims: To identify and validate a dose detection assay based on radiation-induced optical changes of skin (SSA) and changes in the level of selected serum proteins (SPA).
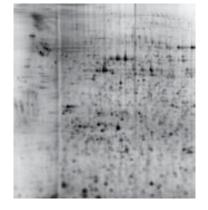

SPA

WP 4: current status

Skin Speckle Assay:
Not suitable for triage because radiation-induced skin changes become manifest after ca 140 days post radiatio.

Serum Protein assay
8 proteins were identified in a mouse model. Currently being tested in serum of patients treated by external beam radiotherapy.
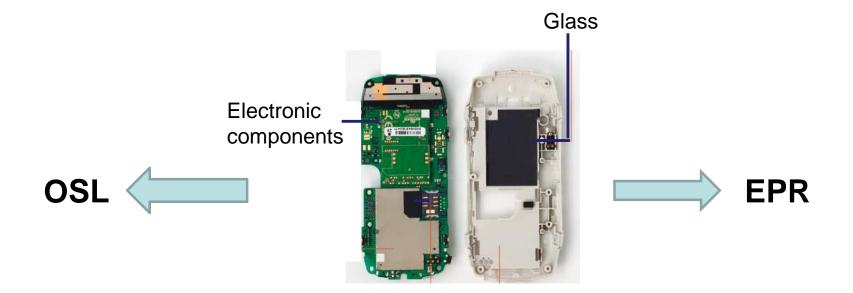
Time between sampling and result: ca 4-6 h

WP5: Electron Paramagnetic Resonance (EPR) and Opticaly Stimulated Luminescence (OSL) in portable electronic devices

Glass

Electronic components

OSL ⬅️ 　　　➡️ **EPR**

Main aim: To identify components of portable electronic devices that can be used to read off the absorbed dose EPR or OSL.

Optically Stimulated Luminescence (OSL)

Electron Paramagnetic Resonance (EPR)

Advantage: high sensitivity (mGy)
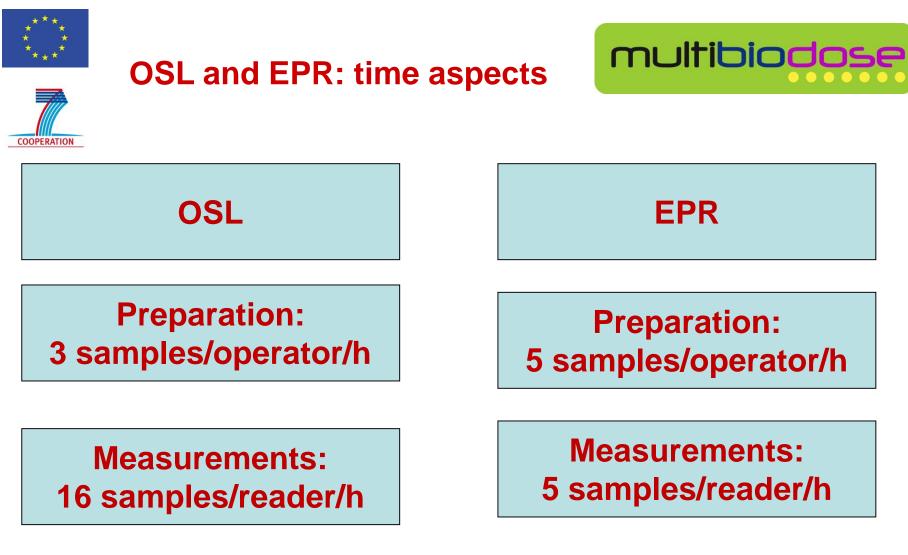
Disadvantage: signal fading – 50% loss during 10 days

Advantage: high signal stability

Disadvantage: low sensitivity (1-2 Gy)

OSL and EPR: time aspects

OSL	EPR
Preparation: 3 samples/operator/h	**Preparation:** 5 samples/operator/h
Measurements: 16 samples/reader/h	**Measurements:** 5 samples/reader/h

1 operator: ca. 200 samples a week
5 operators ca. 1000 samples a week

1 operator: ca. 250 samples a week

Bottle neck: sample preparation

Bottle neck: instrument

WP leader: HPA

WP6: Statistical software package

Reminder:
    Purpose of MULTIBIODOSE – Triage !!
    Need - Dose categorisation and not a dose estimate

Three dose categories:

Red – 'High exposure' – > 2 Gy

Yellow – 'Intermediate exposure' – 1–2 Gy

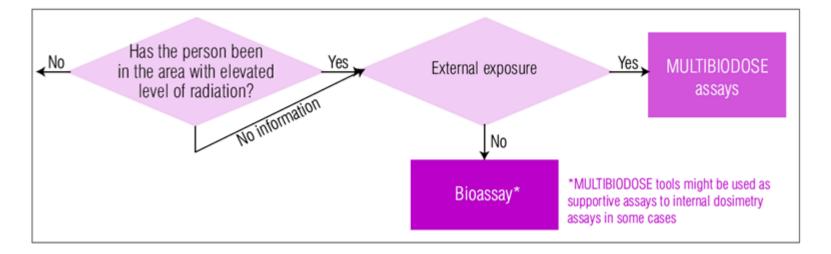**Green – 'Low exposure' – < 1 Gy**

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WP7: Guidance for using MULTIBIODOSE tools in emergencies and dissemination of results

Main aims: (1) To provide information on the project and its results to the emergency preparedness organisations and radiation dosimetry community across Europe. (2) To develop operational guidance for using the tools developed by the MULTIBIODOSE project.

The MULTIBIODOSE team