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Biological dosimetry following exposure to neutrons in a criticality accident

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Abstract

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In 2009, experiments were conducted for establishing both PCC ring and dicentric dose calibration curves. Neutron irradiation of human whole blood obtained from two volunteers was conducted in the Netherlands at the Petten reactor. Cell cultures and analysis of whole blood exposed to eight doses between 0 and 10 Gy were performed for both techniques. For the dicentric assay, excellent uniformity in dose calibration for data from both SU and STUK was observed. For PCC rings, the SU and STUK curves were not equally congruent, probably due to the less uniform scoring criteria. However, both curves displayed strong linearity throughout the dose range. In 2010, an exercise was conducted to simulate a criticality accident and to test the validity of the established dose calibration curves. For accident simulation, 16 blood samples were irradiated in Norway at the Kjeller reactor and analysed for dose estimation with both assays. The results showed that, despite a different com-position of the radiation beams in Petten and Kjeller, good dose estimates were obtained.
The activity has provided good experience on collaboration required in radiation emergency situations where the biodosimetry capacity and resources of one laboratory may be inadequate. In this respect, the project has strengthened the informal network between the Nordic countries: STUK, the Finnish Radiation and Nuclear Safety Authority, NRPA, the Norwegian Radiation Protection Authority and SU, the Stockholm University.

Key words

Biological dosimetry, dose assessment, neutron exposure, PCC ring assay, dicentric chromosome assay

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1. Introduction

In the majority of radiation accidents people are exposed to low LET radiation and therefore laboratories conducting biological dosimetry have established calibration curves mainly for sparsely ionizing radiation. Overexposure to neutron irradiation occurs very rarely. Laboratories lacking an appropriate calibration curve are compelled to use available curves for sparsely ionizing radiation and apply correction factors for neutrons. Or, they would need to use neutron calibration coefficients established in another laboratory. Such approaches will influence the accuracy of dose assessment.

The aim of the BIONCA project was to establish and validate the PCC ring and the dicentric techniques for fission neutrons. The project is a continuation of the earlier NKS-funded biodosimetry projects (BIODOS and BIOPEX; Lindholm et al. 2010), where the PCC ring technique was demonstrated to be an alternative for dicentric assay and especially useful after exposure to high doses of gamma-irradiation. A natural continuation of this collaboration was to expand both biodosimetic assays to cover exposure to fission neutrons. The aim of the BIONCA project was to strengthen the informal network between three organisations: STUK, the Finnish Radiation and Nuclear Safety Authority, NRPA, the Norwegian Radiation Protection Authority and SU, the Stockholm University. The main deliverable of BIONCA is the high competence and resources to perform biodosimetry evaluation in emergency cases where people would be exposed to fission neutrons and where the capacity of a single laboratory could possibly be exceeded.

2. Materials and methods

2.1. Irradiation with neutrons, culture and cytogenetic analysis for the dose calibration curve

Irradiations of blood samples were performed on May 19th 2009 at the High Flux Reactor (HFR) of the Institute of Energy, Joint Research Centre, Petten, Netherlands, where an irradiation facility was built for the purpose of boron neutron capture therapy (Rassow et al. 2001). The advantage of the facility is its excellent dosimetry. The experiment was possible thanks to the competent support of the HFR team Sander Nievaart and Ray Moss.

On the day of exposure blood was collected from two healthy male donors (38 and 49 years old) in 10 x 9-ml tubes per donor. A water phantom (figure 1) was heated to 37ºC and positioned 30 cm away from the neutron beam collimator. A pair of blood tubes (one tube from each donor) was positioned in the water phantom at a depth of 21 cm (2 cm below the water surface) and exposed

Figure 1. Photograph of water phantom with 2 tubes positioned for exposure. Laser light from the positioning system is visible. Opening of the beam can be seen behind.

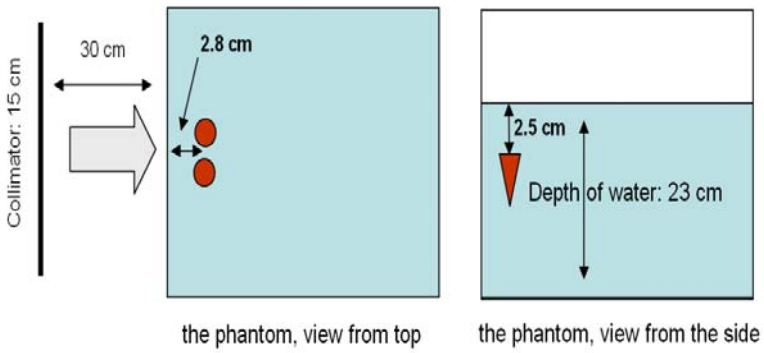


Figure 2. Scheme of the water phantom for irradiating blood samples. Blood samples were submerged in water to ensure dose homogeneity.

to radiation (figure 2). The diameter of the collimator was 15 cm. The doses and irradiation times are given in table 1. In order to increase the number of cells to be analysed at the low dose range, i.e. 0.1 and 0.25 Gy, two blood tubes were irradiated per dose.

Table 1. Doses, exposure times and temperature of water during exposure.

Dose Gy	Exposure time	Time on (rounded)	Time off (rounded)	Temp on Celcius	Temp off Celcius
0	0 min	14:50	15:00	36.5	36.5
0.1	1 min 10 s	11:44	11:46	33.8	33.8
0.1	1 min 10 s	11:53	11:54	34.3	34.3
0.25	3 min	11:57	12:00	34.5	34.5
0.25	3 min	12:04	12:07	34.8	34.8
0.5	6 min	14:15	14:21	36.5	36.5
1.0	12 min	14:23	14:36	36.5	36.2
2.5	30 min	09:38	10:09	33.4	32.5
5.0	60 min	10:18	11:18	33.1	30.3
10.0	120 min	12:10	14:11	35.2	36.5

The neutron energy spectrum consisted of epithermal neutrons with energies mainly between 1 eV and 10 keV, with two small peaks around 30 keV and 70 keV. The beam had a very small divergence (2º) and was circular due to the beam collimator. The thermal neutron flux caused by moderation of the epithermal neutrons had a maximum at 3 cm in a water phantom. The MNCP Monte Carlo code was used to calculate dose distributions (Nievaart et al. 2006). The calculated doses were verified by the foil activation method (Liu et al. 2009).

The beam components are given, as percent values, in table 2. The doses listed in table 1 are the total doses. Induced gamma radiation is that induced in the sample and water by the (n,γ) reaction. The beam gamma component is that included in the neutron beam.

Table 2. Composition of the beam, expressed as percent of the total dose.

Neutron radiation			Gamma radiation			Total
thermal	epithermal	total	induced	beam	total	
13.8	9.0	22.8	61.7	15.5	77.2	100

Following irradiation blood samples were transported to Stockholm where cultures were set up. Three series of culture were set up, with two cultures per point: Series A: Colcemid for the last 4 h, harvest at 50 h culture time; Series B: Colcemid for the last 4 h, harvest at 64 h culture time; Series C: Colcemid for the last 4 h, Calyculin A/Okadaic acid added for the last 1 h, harvest at 50 h culture time. Slides from each donor and dose were delivered to STUK.

At SU, selected samples exposed to 0.1 Gy were stained by the fluorescence plus Giemsa method and the percentage of M2 cells was determined from the PCC samples (Series C): Donor A – 9%, donor S – 4 %. At STUK, samples from several doses were similarly analysed with respect to the M2 cells; the percentage of second division cells was less than 5 %.

2.2 Irradiations with neutrons, culture and cytogenetic analysis for simulation of a neutron accident

A simulated accident irradiation was performed on May 3rd 2010 at the IFE (Institute for Energy Technique) JEEP II research reactor in Kjeller, north of Oslo. The experiment was possible thanks to the competent support and collaboration with Sverre Hval and John Arild Tellevik from IFE. A description of the experiment with photographs can be found at:

http://www.ife.no/ife_nyheter/2010/ife_nks_eksperiment_jeep_ii.

On that day blood was collected in the morning from 3 male and 1 female healthy donors aged between 30 and 38. 4 x 9 ml of blood was collected from each donor and transported to Kjeller. Exposure started at ca 13:00 and stopped at ca 14:00.

A photograph of the exposure facility is shown in figure 3. A hollow, water tight aluminium tube was positioned vertically inside the reactor pool. The temperature of the water pool was ca 20^0C. Each blood sample was lowered into the tube for different time periods, corresponding to doses given in table 3. The depth of blood sample position during exposure was 5 m, corresponded to the maximum neutron flux.

The exposure times and the doses absorbed by the blood samples are shown in table 3. The beam gamma component was measured by a UNIDOS E TM30010 ionisation chamber (PTW, Freiburg, Germany) which was held in the aluminium tube at 5m depth.

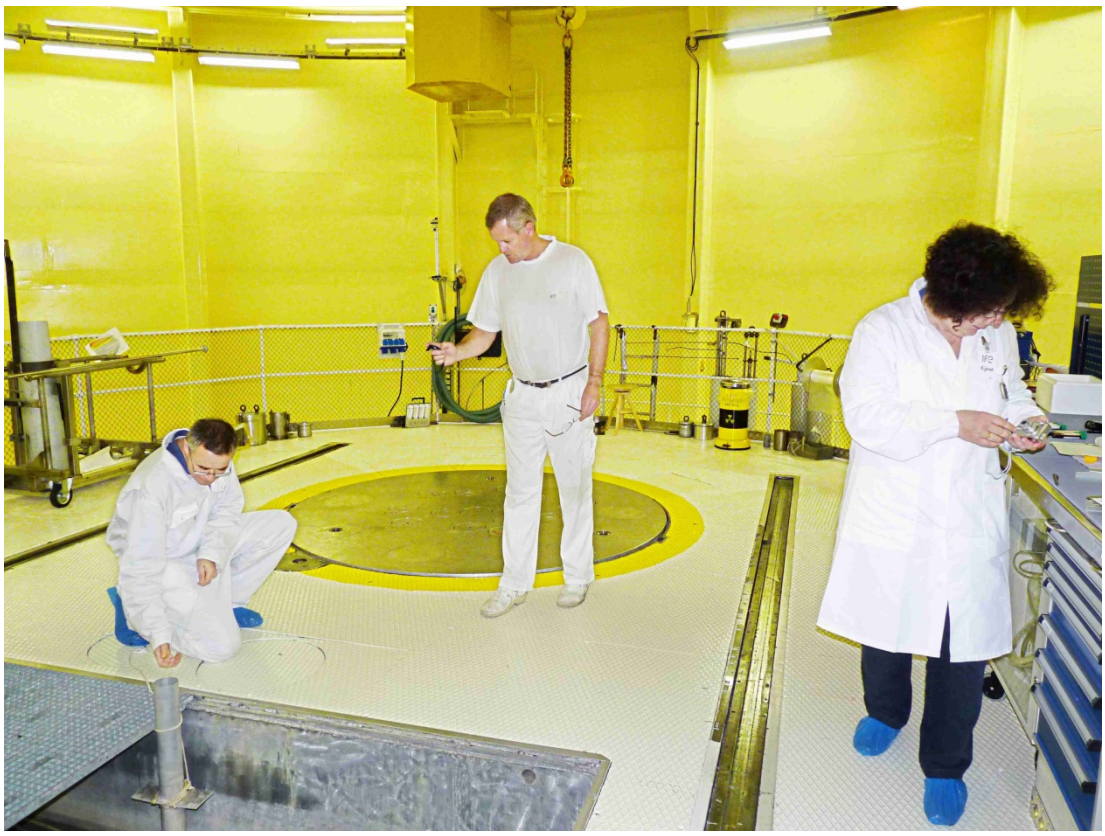


Figure 3. A photograph of the IFE exposure facility. Andrzej Wojcik (left) is kneeing and holding a blood tube inside the aluminium tube. Alicja Jaworska (right) is preparing the next sample for exposure. John Arild Tellevik (middle) is taking the time with a stopwatch.

The neutron doses were calculates by the IFE team and the induced gamma doses were calculated by Justin Brown and Mark Dowdall, radiation chemists at NRPA.

Table 3. Doses and exposure times.

Time of exposure in seconds	Beam gamma Gy	Induced gamma Gy	Neutron dose Gy	Total dose Gy
5	1,38	0,027	0,05	1,46
10	2,77	0,027	0,1	2,89
20	5,53	0,027	0,2	5,76
40	11,07	0,027	0,4	11,49

The composition of the beam is shown in Table 4. The neutron beam is composed of a mixture of epithermal and thermal neutrons.

Table 4. Composition of the beam expressed as percent of the total dose.

Beam gamma	Induced gamma	Neutron dose	Total dose
96,31	0,23	3,48	100,00

Following irradiation blood samples from donors 3 and 4 were sent by DHL to STUK and blood samples from donors 1 and 2 were transported to SU. Whole blood cultures were set up. Two series of cultures were set up, with two cultures per point: Series A: Colcemid for the last 4 h, harvest at 50 h culture time; Series B: Colcemid for the last 4 h, Calyculin A/Okadaic acid added for last 1 h, harvest at 50 h culture time.

Selected samples were stained by the fluorescence plus Giemsa method and the percentage of M2 cells was determined and found to be less than 5 %.

2.3 Statistical analyses

The fitting of dose-response curves for dicentrics and PCC rings was performed both at STUK and at SU for the own scoring results. STUK applied the DoseEstimate software (Ainsbury and Lloyd 2010) and SU applied the CABAS software (Deperas et al. 2007). Both software tools rely on the maximum likelihood method (Papworth 1974) of curve fitting, as recommended for the purpose of biological dosimetry (IAEA 2001). PCC results were fitted by a linear function, whereas a linear-quadratic function was used for dicentrics (IAEA 2001).

Testing of Poisson distribution was performed by a chi-square test (Edwards et al. 1979). Dose estimates for the simulation cases were obtained using calibration curves established for dicentrics and PCC rings.

3. Results

Tables 5 - 6 contain the scoring results of dicentrics and rings in lymphocytes irradiated at the Petten reactor. Data from the two donors were pooled. Results are presented seperately for STUK, SU and NRPA.

The data shown in tables 5a, 5b, 6a and 6b were used to generate calibration curves. The parameters of the calibration curves generated in STUK and in SU are shown in table 7 as well as graphically in figure 4. It can be observed that for the dicentric data almost equal curves were obtained for both SU and STUK. Moreover, the linear term dominates over the quadratic term, rendering the dose calibration curve for dicentric chromosomes almost linear with a small curvature as the dose increases. Surprisingly, no obvious saturation of dicentric frequency was demonstrated despite doses up to 10 Gy. For PCC rings, larger differences in SU vs. STUK calibration curves were observed probably due to the lack of standardized scoring criteria and less experience in PCC ring scoring.

Tables 8 - 13 contain the scoring results of dicentrics and PCC rings in lymphocytes irradiated at the Kjeller reactor. Also shown are the doses with 95% confidence limits estimated based on the

scoring results. SU calibration curves were used to calculate the doses based on the NRPA data. The results are shown in a graphic form in figures 5 and 6. As a general trend, the dicentric data provided more accurate dose estimates than PCC rings. At the two lowest dose points (1,46 and 2,89 Gy) in the PCC ring data (Figure 6 and Tables 9 and 11), none of the 95% confidence limits in the STUK or SU estimates include the dose. However, in the NRPA scoring for donor 3, the estimate was well within the limits (Table 13).

Table 5a. Dicentric scoring data obtained by STUK in lymphocytes irradiated at the Petten reactor. An asterisk next to a value of the product of variance/mean points towards a significant deviation from Poisson distribution. U: results of test-of-fit to a Poisson distribution.

Dose	Number of	Number of	Dicentrics	Dicentric distribution							Variance/	U
Gy	cells	dicentrics	per cell	0	1	2	3	4	5	6	mean	
0	1000	1	0	999	1	0	0	0	0	0	1	0
0,1	2003	42	0,02	1963	38	2	0	0	0	0	1,07	2,4
0,25	1340	91	0,07	1261	68	10	1	0	0	0	1,22	5,69
0,5	347	50	0,14	300	44	3	0	0	0	0	0,98	-0,28
1	150	51	0,34	110	32	6	1	1	0	0	1,26*	2,24
2,5	69	52	0,75	33	25	7	3	1	0	0	1,11	0,64
5	30	61	2,03	2	10	8	6	3	1	0	0,76	-0,91
10	20	77	3,85	0	0	4	3	7	4	2	0,42	-1,81

Table 5b. PCC ring scoring data obtained by STUK in lymphocytes irradiated at the Petten reactor. An asterisk next to a value of the product of variance/mean points towards a significant deviation from Poisson distribution. U: results of test-of-fit to a Poisson distribution.

Dose	Number of	Number of	PCC rings	PCC rings distribution					Variance/	U
Gy	cells	PCC rings	per cell	0	1	2	3	4	mean	
0	1279	1	0,00	1278	1	0	0	0	1	0
0,1	3173	12	0,00	3161	12	0	0	0	1	-0,14
0,25	2295	27	0,01	2268	27	0	0	0	0,99	-0,39
0,5	1185	25	0,02	1162	21	2	0	0	1,14*	3,47
1	840	22	0,03	818	22	0	0	0	0,98	-0,53
2,5	757	70	0,09	694	56	7	0	0	1,11*	2,13
5	668	122	0,18	565	86	15	2	0	1,16	3
10	159	59	0,37	114	32	12	1	0	1,14	1,3

Table 6a. Dicentric scoring data obtained by SU in lymphocytes irradiated at the Petten reactor. An asterisk next to a value of the product of variance/mean points towards a significant deviation from Poisson distribution. U: results of test-of-fit to a Poisson distribution.

Dose	Number of	Number of	Dicentrics	Dicentric distribution							Variance/	
Gy	cells	Dicentrics	per cell	0	1	2	3	4	5	6	mean	U
0	447	1	0,00	446	1	0	0	0	0	0	1	0
0,1	1001	18	0,02	984	16	1	0	0	0	0	1,09	2,1
0,25	997	51	0,05	950	44	2	1	0	0	0	1,15*	3,26
0,5	100	29	0,29	81	12	5	1	1	0	0	1,69*	4,88
1	200	49	0,25	158	35	7	0	0	0	0	1,3*	3,01
2,5	100	74	0,74	43	34	17	3	2	1	0	1,18	1,26
5	60	115	1,92	9	15	14	18	0	5	2	1,1	0,53
10	12	36	3,00	0	2	2	4	2	2	0	0,61	-1,07

Table 6b. PCC ring scoring data obtained by SU in lymphocytes irradiated at the Petten reactor. An asterisk next to a value of the product of variance/mean points towards a significant deviation from Poisson distribution. U: results of test-of-fit to a Poisson distribution.

Dose	Number of	Number of	PCC rings	PCC rings distribution					Variance/	
Gy	cells	PCC rings	per cell	0	1	2	3	4	mean	U
0	3388	3	0,00	3385	3	0	0	0	0,99	-0,02
0,1	3009	10	0,00	2999	10	0	0	0	0,99	-0,12
0,25	3003	35	0,01	2968	35	0	0	0	0,99	-0,44
0,5	1004	24	0,02	981	22	1	0	0	1,06	1,35
1	1000	46	0,05	956	42	2	0	0	1,04	0,94
2,5	732	107	0,15	436	58	6	1	0	1,1	1,62
5	501	137	0,27	384	95	19	3	0	1,11	1,78
10	300	148	0,49	196	71	23	9	1	1,27	3,27

Table 6c. PCC ring scoring data obtained by NRPA in lymphocytes irradiated at the Petten reactor. An asterisk next to a value of the product of variance/mean points towards a significant deviation from Poisson distribution. U: results of test-of-fit to a Poisson distribution. U: results of test-of-fit to a Poisson distribution.

Dose	Number of	Number of	PCC rings	PCC rings distribution						Variance/	
Gy	cells	PCC rings	per cell	0	1	2	3	4	5	mean	U
0,1	1125	11	0,01	1115	9	1	0	0	0	1,17*	4,10
0,25	960	16	0,02	944	16	0	0	0	0	0,98	-0,34
5	160	51	0,32	119	32	8	1	0	0	1,12	1,07
10	80	50	0,63	45	26	5	3	0	1	1,35*	2,22

Table 7. Coefficients of the calibration curves generated by STUK and SU for dicentrics and PCC rings based on the irradiation in Petten and used for assessing the doses applied at the Kjeller reactor. S.E.: standard error.

Assay		Baseline frequency +/- S.E.	Linear term (α) +/- S.E.	Quadratic term (β) +/- S.E.
Dicentric	STUK	0.0007 +/- 0.0010	0.2660 +/- 0.0222	0.0153 +/- 0.0059
	SU	0.00118 +/- 0.00158	0.2480 +/- 0.0212	0.01510 +/- 0.0056
PCC ring	STUK	0.0008 +/- 0.0006	0.0361 +/- 0.0021	0.000
	SU	0.0006 +/- 0.0004	0.0497 +/- 0.0038	0.000

Figure 4. Dose response data and the fitted calibration curves for dicentrics (left panel) and PCC rings (right panel). The data points from NRPA were not fitted, as they were generated to test the scoring compatibility in the three laboratories.

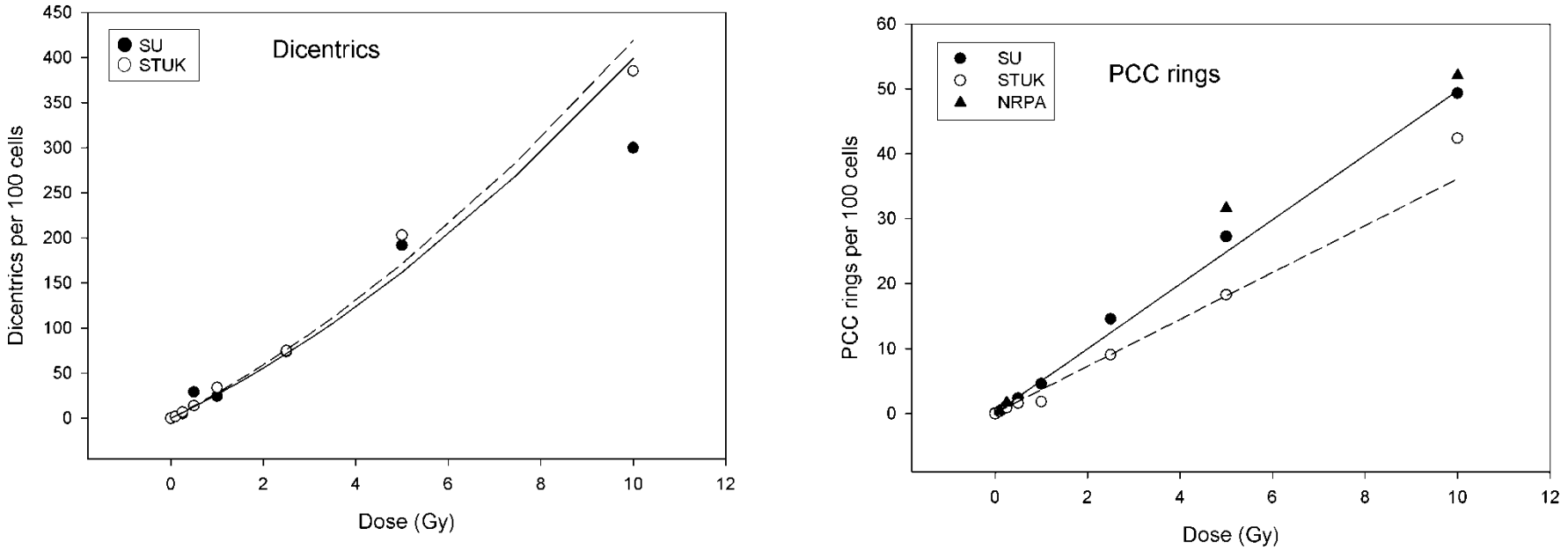


Table 8. SU results of dicentric (dic) scoring and dose estimation for neutron accident simulation. Var: variance, LCL: lower 95% confidence limit, UCL: upper 95% confidence limit. An asterisk next to a value of the product of variance/mean points towards a significant deviation from Poisson distribution. U: results of test-of-fit to a Poisson distribution. High numbers of cells were scored (150 cells or 60 dicentrics).

Donor	Dose	Number of cells	Number of dics	Dic frequency	Distribution												var/ mean	U	Estimated dose	LCL	UCL
					0	1	2	3	4	5	6	7	8	9	10	11					
1	1,46	150	48	0,32	116	29	5	0	0	0	0	0	0	0	0	0	1,00	0,03	1,19	0,89	1,55
1	2,89	100	63	0,63	52	36	9	3	0	0	0	0	0	0	0	0	0,95	-0,35	2,23	1,75	2,77
1	5,76	90	169	1,877	9	31	22	20	6	2	0	0	0	0	0	0	0,76	-1,59	5,63	4,96	6,34
1	11,49	22	95	4,318	0	1	4	5	2	4	2	3	0	0	1	0	1,13	0,48	10,58	9,07	12,20
2	1,46	150	39	0,26	115	31	4	0	0	0	0	0	0	0	0	0	0,95	-0,42	0,98	0,71	1,32
2	2,89	100	81	0,81	44	36	15	5	0	0	0	0	0	0	0	0	0,94	-0,42	2,78	2,27	3,36
2	5,76	90	176	1,955	11	23	32	11	10	2	1	0	0	0	0	0	0,87	-0,86	5,81	5,14	6,53
2	11,49	40	207	5,175	1	0	5	4	7	6	6	4	3	2	1	1	1,04	0,20	12,03	10,89	13,23

Table 9. SU results of PCC ring (Ring) scoring and dose estimation for neutron accident simulation. Var: variance, LCL: lower 95% confidence limit, UCL: upper 95% confidence limit. An asterisk next to a value of the product of variance/mean points towards a significant deviation from Poisson distribution. U: results of test-of-fit to a Poisson distribution. High numbers of cells were scored (500 cells or 100 PCC rings).

Donor	Dose	Number of cells	Number of rings	Ring frequency	Distribution						var/ mean	U	Estim. dose	LCL	UCL
					0	1	2	3	4	5					
1	1,46	500	16	0,032	484	16	0	0	0	0	0,97	-0,47	0,63	0,35	1,02
1	2,89	500	23	0,046	477	23	0	0	0	0	0,96	-0,70	0,91	0,57	1,36
1	5,76	420	147	0,35	302	95	19	2	2	0	1,16	2,27*	6,82	5,78	7,98
1	11,49	280	209	0,746	151	76	35	11	5	2	1,39	4,59*	14,13	12,36	16,05
2	1,46	500	11	0,022	489	11	0	0	0	0	0,98	-0,32	0,43	0,20	0,77
2	2,89	500	51	0,102	457	35	8	0	0	0	1,21	3,42*	2,02	1,50	2,65
2	5,76	420	107	0,254	331	70	18	1	0	0	1,13	1,87	5,00	4,11	6,02
2	11,49	350	260	0,743	170	116	51	10	3	0	1,02	0,29	14,06	12,48	15,77

Table 10. STUK results of dicentric (dic) scoring and dose estimation for neutron accident simulation. Var: variance, LCL: lower 95% confidence limit, UCL: upper 95% confidence limit. An asterisk next to a value of the product of variance/mean points towards a significant deviation from Poisson distribution. U: results of test-of-fit to a Poisson distribution. Minimal numbers of cells were scored (50 cells or 30 dicentrics).

Donor	Dose	Number	Number	Dic	Distribution									var/		Estimated		
	Gy	of cells	of dics	frequency	0	1	2	3	4	5	6	7	8	mean	U	dose	LCL	UCL
3	1,46	50	26	0,52	30	14	6	0	0	0	0	0	0	0,96	-0,2	1,77	1,19	2,5
3	2,89	25	24	0,96	10	8	5	2	0	0	0	0	0	1	-0,01	3,07	2,07	4,3
3	5,76	9	33	3,67	1	1	0	2	2	1	1	1	0	1,36	0,74	9,06	6,82	11,61
3	11,49	5	29	5,8	0	0	0	1	0	0	2	2	0	0,47	-0,77	12,63	9,46	16,21
4	1,46	50	25	0,5	30	17	1	2	0	0	0	0	0	1,08	0,41	1,71	1,14	2,5
4	2,89	34	23	0,68	17	11	6	0	0	0	0	0	0	0,87	-0,54	2,25	1,48	3,22
4	5,76	9	25	2,78	0	1	3	2	3	0	0	0	0	0,43	-1,16	7,34	5,2	9,84
4	11,49	5	32	6,4	0	0	0	0	0	2	0	2	1	0,28	-1,03	13,53	10,32	17,11

Table 11. STUK results of PCC ring (Ring) scoring and dose estimation for neutron accident simulation. Var: variance, LCL: lower 95% confidence limit, UCL: upper 95% confidence limit. An asterisk next to a value of the product of variance/mean points towards a significant deviation from Poisson distribution. U: results of test-of-fit to a Poisson distribution. Minimal numbers of cells were scored (300 cells or 50 PCC rings).

Donor	Dose	Number	Number	Ring	Distribution				var/		Estimated		
		of cells	of rings	frequency	0	1	2	3	mean	U	dose	LCL	UCL
3	1,46	300	8	0,03	292	8	0	0	0,98	-0,31	0,72	0,3	1,43
3	2,89	300	17	0,06	285	14	0	1	1,3	3,79	1,55	0,89	2,49
3	5,76	301	53	0,18	258	35	6	2	1,28	3,47*	4,86	2,84	8,14
3	11,49	119	56	0,47	79	28	8	4	1,25	1,97*	13,01	7,83	21,2
4	1,46	300	8	0,03	292	8	0	0	0,98	-0,31	0,72	0,3	1,43
4	2,89	300	14	0,05	286	14	0	0	0,96	-0,55	1,27	0,69	2,15
4	5,76	300	43	0,14	260	37	3	0	1	-0,01	3,95	2,85	5,33
4	11,49	189	69	0,37	135	42	9	3	1,16	1,59	10,09	7,85	12,78

Table 12. NRPA results of dicentric (dic) scoring and dose estimation for neutron accident simulation. Var: variance, LCL: lower 95% confidence limit, UCL: upper 95% confidence limit. An asterisk next to a value of the product of variance/mean points towards a significant deviation from Poisson distribution. U: results of test-of-fit to a Poisson distribution. High numbers of cells were scored (150 cells or 60 dicentrics).

Donor	Dose	Number of cells	Number of dics	Dic frequency	Distribution						var/ mean	U	Estimated dose	LCL	UCL
					0	1	2	3	4	5					
2	2,89	30	10	0,33	21	8	1	0	0	0	0,90	-0,40	1,24	0,62	2,18
3	2,89	27	21	0,78	10	14	2	1	0	0	0,73	-1,01	2,69	1,75	3,87

Table 13. NRPA results of PCC ring (Ring) scoring and dose estimation for neutron accident simulation. Var: variance, LCL: lower 95% confidence limit, UCL: upper 95% confidence limit. An asterisk next to a value of the product of variance/mean points towards a significant deviation from Poisson distribution. U: results of test-of-fit to a Poisson distribution. High numbers of cells were scored (500 cells or 100 PCC rings).

Donor	Dose	Number of cells	Number of rings	Ring frequency	Distribution				var/ mean	U	Estim. dose	LCL	UCL
					0	1	2	3					
2	2,89	300	21	0,07	280	19	1	0	1,03	0,35	1,39	0,86	2,12
3	2,89	300	39	0,13	262	37	1	0	0,92	-0,93	2,57	1,83	3,51

Figure 5. Results of dose estimates based on scoring of dicentrics in lymphocytes of 4 donors exposed to radiation at the Kjeller reactor. Horizontal lines show the applied doses. The dose estimates are shown for each donor along with 95% confidence limits. Dose estimates in lymphocytes of donors 3 and 4 were based on scoring minimal numbers of cells, hence the broad 95% confidence limits. More information about this is given in the discussion.

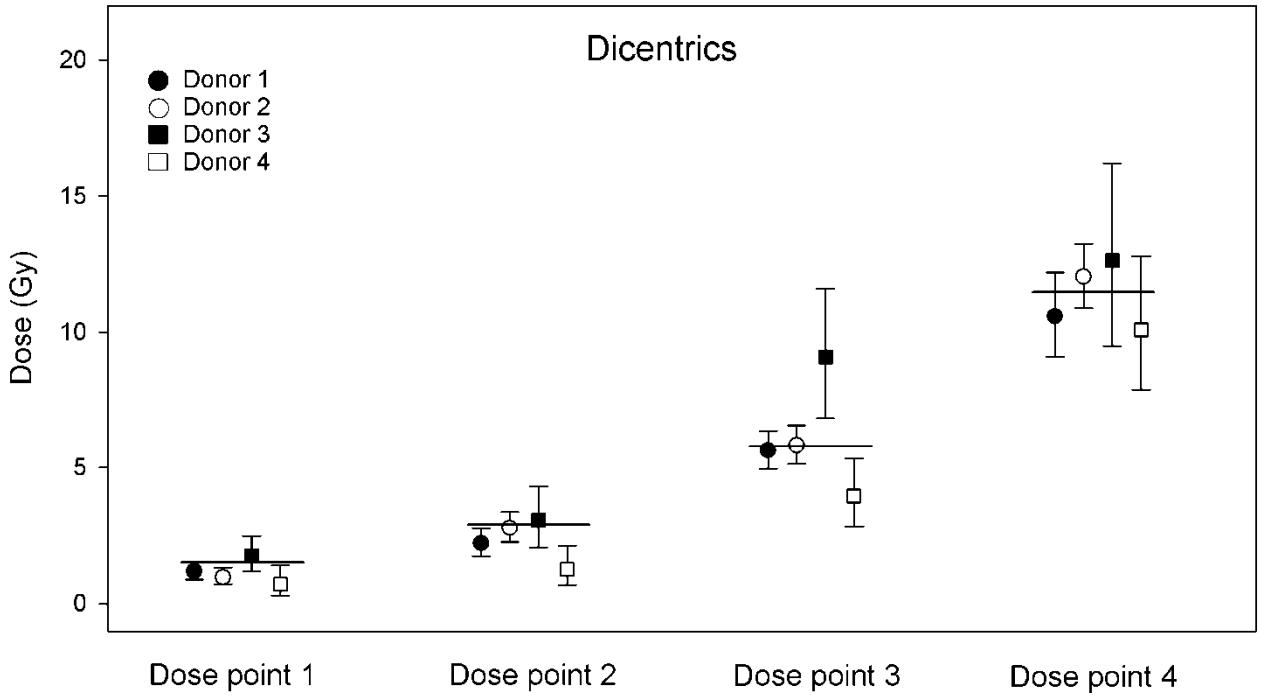
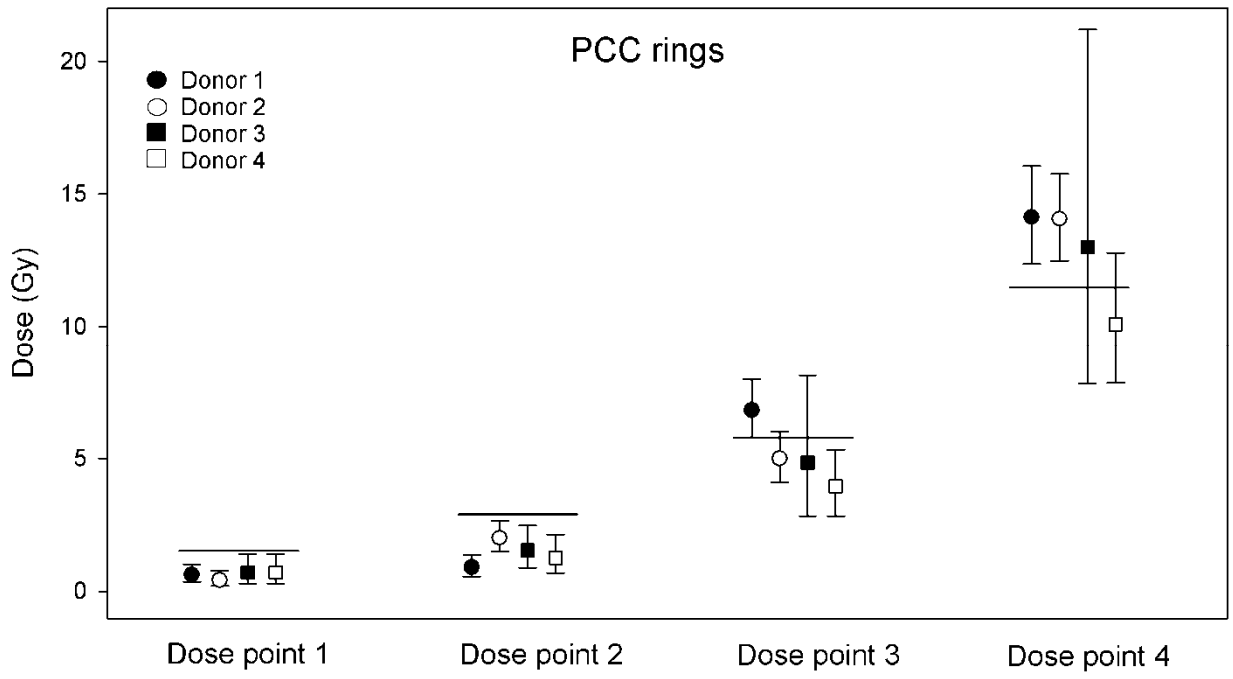


Figure 6. Results of dose estimates based on scoring of PCC rings in lymphocytes of 4 donors exposed to radiation at the Kjeller reactor. Horizontal lines show the applied doses. The dose estimates are shown for each donor along with 95% confidence limits. Dose estimates in lymphocytes of donors 3 and 4 were based on scoring minimal numbers of cells, hence the broad 95% confidence limits. More information about this is given in the discussion.



4. Discussion

The scoring of dicentric chromosomes in lymphocytes irradiated at the Petten reactor lead to nearly identical calibration curves generated in STUK and SU. This points towards highly coherent scoring criteria used in both laboratories. In consequence, both laboratories could use a common calibration curve. This is an important finding for the case that both laboratories would have to jointly score samples from a radiation accident.

The dicentric frequency recorded at SU after the dose of 10 Gy is much below the fitted curve. This was due to a poor quality of chromosomes and few mitotic cells that could be found after this dose. This dose-point has, however, a minor impact on the fitted calibration curve, which clearly shows the advantage of using the method of maximum likelihood (Papworth 1974) for curve fitting, where the data points are weighted based on the number of scored cells.

The calibration curves for PCC rings deviate more strongly. This is not surprising in view of the less uniform criteria for identifying PCC rings than for dicentrics. Obviously, for the PCC ring technique, each laboratory should base dose estimation on its own calibration curve. Despite the differences in PCC ring curves for SU and STUK, both curves showed strong linearity at least up to 10 Gy and no saturation of ring formation was observed. Lamadrid et al. (2006) reported no increase of PCC rings beyond a neutron dose of 9.4 Gy, however, here the neutron component was larger and the mean energy higher than in the Petten irradiations.

The calibration curves were used to assess the doses measured in lymphocytes exposed in the Kjeller reactor. A very interesting aspect of this experiment was that the Petten and the Kjeller beams differ in quality. As the Petten reactor beam comprised of gamma to approximately 77% , the IFE beam is composed to over 95% of gamma radiation. This difference is due to the fact that the gamma radiation in the Petten beam is blocked by a dedicated argon filter. Such filtering is not available in Kjeller, where the blood samples were exposed in a tube that was positioned directly in the reactor cooling water. Despite these differences, good dose estimates could be achieved, whereby the estimates based on dicentrics were superior to those based on PCC rings. This result is in accordance to our earlier findings with gamma radiation (Lindholm et al. 2010).

It was interesting to compare the precision of dose-estimates following scoring of different cell numbers. Hence, high numbers of cells were scored in lymphocytes of donors 1 and 2: 150 cells or 60 dicentrics and 500 cells or 100 PCC rings. Low numbers were scored in lymphocytes of donors 3 and 4: 50 cells or 30 dicentrics and 300 cells or 50 PCC rings. The aim of this was to explore how accurate dose estimates can be achieved with a triage approach in an acccident scenario, where limited time is available to score many cells. The results clearly show the lower precision of dose estimate in samples where the number of scored cells was low. Nevertheless, the triage scoring appears

to be sufficient for preliminary evaluation of dose categories for medical care purpose. For a more precise dose assessment, it is recommended to score high number of cells, whenever possible.

In conclusion, the BIONCA project has been a very good collaborative exercise between STUK, NRPA and SU in many aspects. The laboratories now have neutron dose calibration curves available for both dicentrics and PCC rings that can be used for dose assessment of overexposure cases. The results clearly demonstrated that laboratory procedures and scoring criteria are highly coherent, especially for the dicentric assay. Thus, the laboratories are well prepared to share work in assessing doses by biological dosimetry in any future radiation accident or incidence that require a capacity that goes beyond what one laboratory can provide. The coherent outcome should be maintained by continued collaboration between the laboratories.

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Abstract	The aim of the BIONCA project was to implement cytogenetic techniques for biodosimetry purposes in the Nordic countries. The previous NKS-funded biodosimetry activities (BIODOS and BIOPEX) concentrated on experiments using gamma-irradiation and on developing the PCC ring assay for biodosimetry. Experiments conducted during the present BIONCA project has broadened the biodosimetry capacity of the Nordic countries to include dose estimation of exposure to neutrons for both PCC ring and dicentric chromosome techniques. In 2009, experiments were conducted for establishing both PCC ring and dicentric dose calibration curves. Neutron irradiation of human whole blood obtained from two volunteers was conducted in the Netherlands at the Petten reactor. Cell cultures and analysis of whole blood exposed to eight doses between 0 and 10 Gy were performed for both techniques. For the dicentric assay, excellent uniformity in dose calibration for data from both SU and STUK was observed. For PCC rings, the SU and STUK curves were not equally congruent, probably due to the less uniform scoring criteria. However, both curves displayed strong linearity throughout the dose range. In 2010, an exercise was conducted to simulate a criticality accident and to test the validity of the established dose calibration curves. For accident simulation, 16 blood samples were irradiated in Norway at the Kjeller reactor and analysed for dose estimation with both assays. The results showed that, despite a different com-position of the radiation beams in Petten and Kjeller, good dose estimates were obtained. The activity has provided good experience on collaboration required in radiation emergency situations where the biodosimetry capacity and resources of one laboratory may be inadequate. In this respect, the project has strengthened the informal network between the Nordic countries: STUK, the Finnish Radiation and Nuclear Safety Authority, NRPA, the Norwegian Radiation Protection Authority and SU, the Stockholm University.
Key words	Biological dosimetry, dose assessment, neutron exposure, PCC ring assay, dicentric chromosome assay

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