

REPORT ON THE ON-SITE BIODORA TRAINING COURSE: MUNICH 10/07/23-14/07/23

The on-site BIODORA Training Course took place in Bundesamt für Strahlenschutz (BFS) in Munich during the 10th – 14th of July 2023. During this course all the Biological Dosimetry methods were demonstrated by the experts in the field and hands-on training was provided to the participants for the following techniques: the Dicentric assay (DCA), Radiation induced foci assay (γ -H2Ax), Cytokinesis block micronucleus (CBMN) assay, cell fusion induced Premature Chromosome Condensation (PCC) assay, chemical induced ring PCC assay, Fluorescence in Situ Hybridization (FISH) assay and gene expression assay.

MONDAY 10/07: At the beginning of the Course all the participants we were welcomed, were introduced to the Campus and safety instructions were discussed before the initiation of the Lab sessions. The first assay that we performed was the γ -H2Ax foci assay. Firstly, we discussed the procedure and some technical details and then we practiced the slide preparations for this assay using the cytopsin and centrifuging the samples onto the slides. Afterwards, we practiced in the analysis and scoring of the γ -H2Ax foci using the IKAROS Metasystems software. The second part of the session included the cultivation of blood, a procedure that is common for the DCA, MB and FISH assays. We had practical training in cultivating blood in RPMI medium using Phytohemagglutinin (PHA) for the stimulation of peripheral blood lymphocytes and their entrance to the cell cycle. Then we performed the fixation procedure and the slide preparation for the DCA and the FISH techniques.

TUESDAY 11/07: On Tuesday the first session was dedicated to the MN assay. We prepared the slides for this assay, and then we analysed blood samples using the IKAROS Metasystems software in three different ways: fully automatically, semi-automatically and manually. Then we estimated the radiation dose received by the sample using the Biodose Tools software for all the three different analyses. The second session, included the analysis and scoring of dicentric chromosomes for the DCA assay. Metaphases were provided and we were asked to analyse them and identify the dicentric chromosomes. Then we were introduced to a newly proposed automatic specific software for dicentrics identification provided by Metasystems (Germany) that enables fast analysis and dose evaluation.

WEDNESDAY 12/07: The whole day was dedicated to the cell fusion induced-PCC assay. We followed the whole procedure: the collection of mitotics Chinese Hamster Ovary (CHO) cells and their fusion with non-stimulated peripheral blood lymphocytes using Polyethylen Glycol (PEG). Then we were provided with fusion images of CHO cells and mononuclear white blood cells irradiated at different radiation doses. We practiced in identifying the fusions, counting the Prematurely Condensed Chromosomes PCCs and we created a “draft” calibration curve. Then we were asked to analyse three different samples and categorize them in a) control group, b) low irradiation dose group c) high irradiation dose group.

THURSDAY 13/07: On Thursday, during the first session we were introduced to the FISH technique. We practiced in the chromosome painting for FISH: denaturation of DNA, hybridization treatment using three different probes (three different chromosomes painting), and then the post-hybridization treatment and the finalization of slides preparation. Then we analysed some metaphases and scored the existing translocations using the IKAROS Metasystems software. We then used the results for dose estimation using the BIDOSE Tools. The second session refers to the ring PCC assay. The protocol was explained and then we prepared the slides for analysis. Images of lymphocyte PCCs were provided and we were asked to identify ring chromosomes and based on our analysis to identify the radiation dose.

FRIDAY 14/07: Friday was the last day of the Course and was dedicated to the gene expression method. We discussed all the steps of this method and its possible applications with its pros and cons. We were demonstrated all the equipment and the capabilities that this method offers in case of a large scale radiation accident. We then practiced in creating calibration curves using the FDXR gene and compared our curves with the existing ones in the Lab. After this session, we were gathered for a summary, to discuss what we learned during the whole week and then departed for the airport.

Overall, I would like to report that this course was outstanding! We were provided the protocols for all the techniques, we had the techniques demonstrated by the experts in the field and a full week of hands-on training. After the completion of this course, I feel I have not only the knowledge but also the confidence to apply most of the biological dosimetry methods in my Lab. I am really grateful for that and I would definitely recommend the course to every student, young scientist or researcher who wishes to obtain or maintain competence in biological dosimetry in the radiation protection field.

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