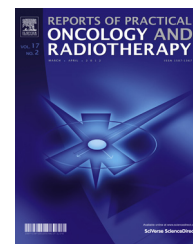




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## Original research article

# Experimental set up for the irradiation of biological samples and nuclear track detectors with UV C



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## ABSTRACT

**Aim:** In this work we present a methodology to produce an “imprint” of cells cultivated on a polycarbonate detector by exposure of the detector to UV C radiation.

**Background:** The distribution and concentration of <sup>10</sup>B atoms in tissue samples coming from BNCT (Boron Neutron Capture Therapy) protocols can be determined through the quantification and analysis of the tracks forming its autoradiography image on a nuclear track detector. The location of boron atoms in the cell structure could be known more accurately by the simultaneous observation of the nuclear tracks and the sample image on the detector.

**Materials and Methods:** A UV C irradiator was constructed. The irradiance was measured along the lamp direction and at different distances. Melanoma cells were cultured on polycarbonate foils, incubated with borophenylalanine, irradiated with thermal neutrons and exposed to UV C radiation. The samples were chemically attacked with a KOH solution.

**Results:** A uniform irradiation field was established to expose the detector foils to UV C light. Cells could be seeded on the polycarbonate surface. Both imprints from cells and nuclear tracks were obtained after chemical etching.

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Conclusions: It is possible to yield cellular imprints in polycarbonate. The nuclear tracks were mostly present inside the cells, indicating a preferential boron uptake.

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## 1. Background

Boron Neutron Capture Therapy (BNCT) is a cancer treatment modality based on the high thermal neutron capture cross section of  $^{10}\text{B}$  and the high linear energy transfer (LET) radiation of the particles emitted in the reaction. Nontoxic tumor-seeking boron compounds are administered to patients and the region to be treated is exposed to a thermal/epithermal neutron flux. Due to the short path of the emitted particles, a lethal damage is produced on tumor cells, preserving surrounding normal tissue.<sup>1</sup>

The precise knowledge about the location of boron compounds in tumor and surrounding tissue is essential in order to optimize the treatment protocol. The variety of tissue structures will accumulate different amounts of boron and this will affect dose distribution. Dosimetric effect of tissue heterogeneity has also been reported for other radiation therapies.<sup>2,3</sup>

The neutron autoradiography can provide this essential information.<sup>4</sup> This methodology is based on the superposition of a biological sample and a solid state nuclear track detector (SSNTD). When the assembly sample-detector is irradiated with thermal neutrons, the detector registers the damage (nuclear tracks) produced by the alpha and lithium particles originated in the neutron capture reaction with the boron atoms present in the sample. As alpha and lithium particles are ejected in opposite directions, only one of them is registered in the detector per each  $^{10}\text{B}$  reaction making it possible to localize the emission place in the sample. In this way the distribution and concentration of  $^{10}\text{B}$  atoms in biological samples can be determined through the quantification and analysis of the tracks forming its autoradiography image on the SSNTD.<sup>5</sup> As a chemical attack is necessary to enlarge the damage and convert it to observable tracks, the biological sample is lost in the etching process. For this reason, a methodology based on the use of a reference system have been developed,<sup>6</sup> to correlate the histological and the autoradiographic images for a better determination of local boron concentration values.

However, the location of boron atoms in the cell structure could be known even more precisely by the simultaneous observation of the nuclear tracks and the sample image on the detector. Sophisticated techniques of high resolution autoradiography<sup>7</sup> can be found in the literature, but an attractive alternative is the detector sensitization with UV C, in order to create reliefs in the SSNTD. This work had been proposed for culture cells in polyallyldiglycol carbonate and combined with special observation methods.<sup>8–10</sup>

## 2. Aim

The advantages of polycarbonate for neutron autoradiographic studies of samples coming from BNCT protocols were

previously demonstrated,<sup>11,12</sup> so we present here the experimental setup to produce an “imprint” of cells cultivated on a polycarbonate detector by exposure of the detector to UV C radiation.

## 3. Material and methods

Cells of a human metastatic line of melanoma (MELJ) were seeded on polycarbonate foils (Lexan<sup>TM</sup>) of 250  $\mu\text{m}$  thickness. The detectors were cut with circular shapes in order to fit in the base of Petri dishes (diameter: 60 mm). They were incubated with borophenylalanine (BPA, 10  $\mu\text{g}/\text{L}$ ) for 4 h, and then washed and fixed. Glutaraldehyde and Methanol were tested as fixation solutions. Previous studies reported  $^{10}\text{B}$  concentrations in blood during BNCT treatments calculated as the mean of measured values just before and after BNCT. The values were among 3 and 9 ppm of boron in blood.<sup>13</sup> The concentration of 10 ppm of boron in the medium of cells cultures adopted in our study represents a value close to those above mentioned values determined in blood samples during clinical treatments.

As shown in Fig. 1, the samples were attached to Lexan films of 9 cm  $\times$  13 cm and fixed to an acrylic container, in order to expose them to the thermal column of the biomedical facility of the RA-3 Reactor (Ezeiza, Argentina).<sup>14,15</sup> Two neutron fluences were evaluated: 10<sup>12</sup> n cm<sup>-2</sup> and 10<sup>13</sup> n cm<sup>-2</sup>.

The neutron flux was assumed to be uniform. Neutron flux was previously measured using a SPND (Self Powered Neutron Detector) and the uncertainty in the delivered neutron fluence is 8%. A UV C irradiator was built with a 254 nm wave length lamp (G15T8, General Electric, USA) of 15 W. A wooden container with a sample holder was fabricated and painted with opaque black coating, in order to absorb light. The dose was evaluated as a function of the distance from the lamp. The uniformity of the irradiance along the lamp and in the transversal direction was also analyzed in order to establish the field where the samples would receive a uniform dose. A UV C radiometer (Extech 40736C) was used for this purpose. A scheme of the UV C irradiation facility is presented in Fig. 2.

The samples were divided in sections of about 2 cm  $\times$  2 cm, exposed to the UV C lamp for 8 h, stained with hematoxylin and explored with increasing magnification (1.25 $\times$ , 10 $\times$ , 20 $\times$ , 40 $\times$  and 100 $\times$ ) with a light microscope (Olympus DP70). Reference points were established on different regions in order to localize them after the chemical etching and correlate the cellular images with its corresponding autoradiographies. For this purpose, the foils were drilled with a drill bit (diameter: 0.5 mm). The lateral resolution using the reference points is  $\leq 10 \mu\text{m}$ .

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