# Cell irradiation experiment using laser driven protons at ultra high dose rate

K.F. Kakolee[1], D. Doria[1], S. Kar[1], S. Litt[1], M. Zepf[1], M. Borghesi[1], D. Kirby[2], F. Fiorini[2], S. Green[2], K. Kirby[3], J.C. Jeynes[3], M. Merchant[3], J.Kavanagh[4], G. Schettino[4]

*[1] School of Mathematics and Physics, Queen's University Belfast, UK. [2] School of Physics , Department of Medical Physics, Birmingham University, UK. [3]Surrey Ion Beam Centre, University of Surrey, U.K .[4]Center for Cancer Research & Cell Biology, Queen's University Belfast, U.K.*

**Abstract:** The effect of proton irradiation of biological cells, on timescales orders of magnitude shorter than with conventional accelerators, has been investigated by employing the TARANIS laser at Queen's University. Multiple cell-spots with different doses and proton energies were irradiated at the same time in a single laser shot at dose rates exceeding $10^9$ Gy/sec. The data show a clear dose-dependant lethal effect of laser-driven protons over V-79 cells. A comparison with the survival obtained with an X-Ray standard source has been done and the resulting relative biological effectiveness (RBE) is about 1.3 at 10%.

**Introduction:** It has been widely recognized that the use of ion beams in cancer radiotherapy has the major advantage of highly localised dose deposition in the target due to the Bragg peak feature in their energy deposition curve [1]. Ions are able to deliver lethal amount of doses into the tumor, while limiting harm to the surrounding healthy tissues (Fig.1).

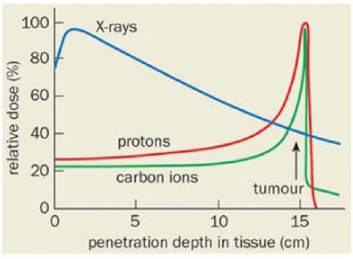


Figure 1. Comparison of the depth dose profiles of proton, carbon and photon (Xray).

A number of facilities dedicated to hadron-therapy using conventional accelerators are currently in operation in the world. However, the cost and size of these facilities limit a real diffusion of this treatment. The idea of future facilities based on laser-driven ions has been proposed as a way of reducing the complexity and cost. The rapid development of laser technology and related areas continues to encourage this hypothesis. At present, Petawatt lasers can produce proton beam

energies up to several tens of MeV, while proton energies of 250MeV are needed to reach deep-seated tumours in the human body. A significant amount of scientific effort is currently ongoing for achieving the ion beam parameters desired for the therapy [2]. Recently, some preliminary work on methodology and viability of implementing a laser-driven ion source for cell irradiation [3,4] has been published. Here, we report a quantitative measurement of relative biological effectiveness (RBE) of acute proton dose (up to 5 Gy) in the ultra-high dose-rate regime ($10^9$Gy/s) on V79 cancer cells (radiosensitive chinese hamster lung cancer cells).

**Experiment:** The experiment has been conducted using the TARANIS laser at Queen's University of Belfast. It is a Ti:Sapphire-Nd:glass laser system which can deliver energy up to 30J at 1053nm wavelength in a pulse duration of 700fs. A focal spot of 10um diameter was attained by using an F/3 parabola of focal length 315mm. The beam was focused onto a thin aluminum foil at intensities of the order of $10^{19}$W/cm$^2$. The accelerated particles were made to pass through a collimator and a magnet. An aperture of 500μm was used to select both the energy resolution and the irradiation time. A dipole magnet of 0.9 T was used to resolve the proton energy spectrum. At a distance of 14cm from the magnet, a 50μm thick mylar foil was used as a chamber window. The dish containing a V-79 cell monolayer, and the radiochoromic films (EBT2) were placed in air and 6cm far from this window (Fig. 2). Electrons and the X-rays generated by the laser could not reach the cells: the electrons being deviated in the opposite direction of protons and the X-rays continuing straight.

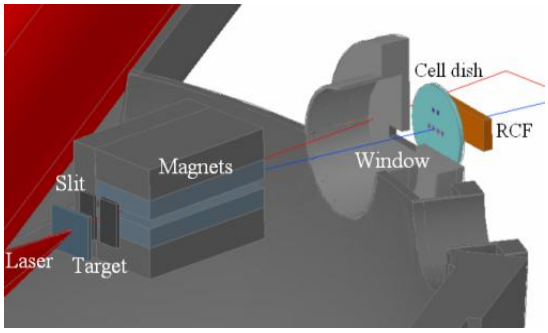


Figure 2. Experimental set-up used to irradiate the cancer cells. The red and blue lines represent the X-ray trajectory (no deflected) and the trajectory proton with middle energy of about 6MeV respectively.

**Procedures with cell line:** A laminar flow cabinet or a biosafety hood was used to avoid contamination from micro-organisms at the time of preparing the cells for irradiations as well as after the irradiation. The cells were cultivated in Dulbecco's minimum essential medium whereas Dulbecco's phosphate buffer solution (DPBS) containing 10% versane has been used to wash the

cells. Cells were seeded 10 hours before irradiation on a 150μm thick mylar sheet to give them time to multiply and stick on it. The density of the cells was found to be around $4\times10^4$ cells/mm$^2$. Cell dots of 2 mm diameter have been cut from that mylar sheet and placed over a 3μm mylar film. In each irradiated cell dish 6 cell dots were placed (fig. 3): 2 cell dots were used as controls, which were not irradiated during the laser shot; and the other 4 cell dots were irradiated by the protons. The plating efficiencies of the control points were nearly 60%. After irradiation plating densities were chosen to give 200 cells per petri dish with three dishes per dose point. Post–plating, all the petri dishes were maintained at $37^0$C in an atmosphere of 5%

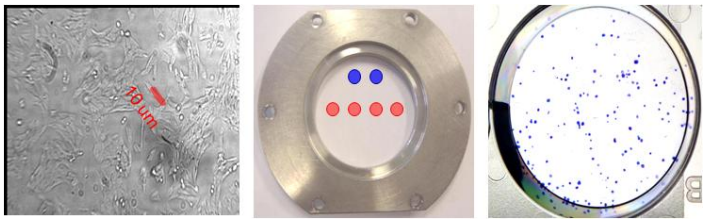


Figure 3. From left to right, cell line under the microscope and how cell dots are placed before irradiation and after irradiation how colony look like.

$CO_2$, 95% humidification in the incubator. There each living cell after irradiation finds the appropriate environment to multiply and form a cell colony (containing few hundred cells, Fig. 3). After five days the cells were stained with crystal violet to highlight the colonies and making them visible as violet dots. Each colony was counted by eye and the surviving fractions for different dose points were calculated.

**Result and Discussions:** The very low energetic protons were stopped by the mylar windows but those with an initial energy higher than 2 MeV could reach the dish and the EBT2 film stack. The films were calibrated using the 29 MeV monoenergetic beam accelerated by the cyclotron of the University of Birmingham, for doses up to 14 Gy. The counts in the control cell dots were used to calculate the plating efficiency (PE) for that particular shot. PE refers to the percentage of the cells seeded that grow into colonies [5]. The number of colonies counts for other dots were compared with the original number of cells placed for that particular shot to have a survival fraction. In general, the surviving fraction is given by, Surviving Fraction= Colonies Counted/ (Cells seeded * (PE/100)). A cell survival curve (fig 4) then describes the relationship between the radiation dose and the proportion of cells that survive.

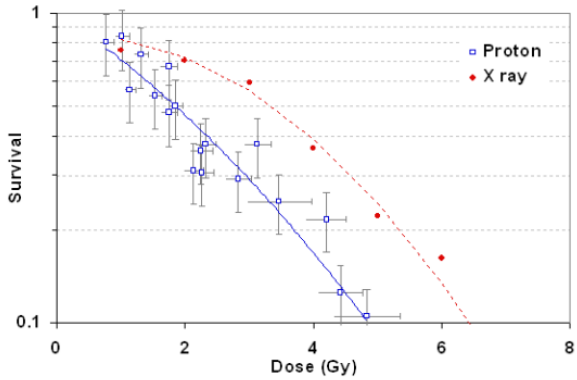


Figure 4. Survival curves of the V79 cancer cells obtained by proton and X-ray irradiation with different doses.

The survival curve obtained with protons shows the expected higher efficiency of protons in killing the cell with respect to X-rays. From the comparison between proton and X-ray data, a RBE of about 1.3 can be found at 10% of survival, which is consistent with the values present in literature [6].

**Acknowledgements:** This work was funded by EPSRC (LIBRA Consortium, Grant No: EP/E035728/1). We acknowledge the support and contribution of the Birmingham cyclotron team for the irradiation during film calibration.

**References:**

[1] J. M. Ruiz De Almodovar et al, Radiation Research 138 (1 SUPPL.), S93-S96 (1994)

[2] S. V. Bulanov et al, Phys. Lett. A 299, 240–247 (2002)

[3] S. D. Kraft et al., New Journal of Physics 12, 085003 (2010)

[4] A.Yogo et al., Appl. Phys. Lett. 94, 181502 (2009).

[5] W. P. Levin *et al*., *Proton Beam Therapy*, International Hospital Federation Reference Book 2006/ 2007.

[6] B. G. Wouters, et al., Radiat. Res. 146, 159-170 (1996)