

STUDY OF THE INSTALLATION OF A SMALL ANIMAL EXPERIMENT IN A MC-50 CYCLOTRON LEPT BEAM LINE*

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Abstract

Proton therapy has a feature of minimal effect on tumor surrounding healthy tissue and huge damage on tumor volumes specifically. Due to these characteristics of proton therapy the number of patients with receiving proton therapy is increasing every year. Proton therapy is useful for tumor treatment but still not know mechanism of proton beam that how to kill the tumor cells. In Korea, a lot of current research progressed at the cellular level by using a proton accelerator, the animal experiments was not held virtually because of the absence of the device. In this study, we installed animal experiment device for proton beam.

INTRODUCTION

The proton beam is known as a one of the powerful tools for the treatment of tumor tissues. The proton has a better dose distribution compared to photons [1, 2]. This physical advantage of proton can be used to reduce radiotherapy-induced side effects by sparing normal tissue. Because of this advantage, proton therapy has gained increasing attention in the last decade. Several researches were recently published advocating the effectiveness of proton therapy [3-5].

Proton beam-induced cell death is identified as apoptosis [6]. However mechanisms of proton beam-induced cell death is not fully clear. At the present, in-vivo experiment is more and more important and an essential factor. In Korea, mechanism study of proton beam-induced apoptosis has stayed at the in-vitro level. Because it is not enough as the Korea has machine for a research and in vivo experimental devices. In this research, we composed in-vivo experiment device adding ridge filter type modulator, range shifter, collimator, bolus etc. at LEPT (Low Energy Proton Therapy) beam line of MC-50 cyclotron (Korea Institute of Radiological and Medical Sciences). In this paper, we confirmed that size of mouse tumor decreases with in-vivo experiment device by proton beam. And we confirmed the possibility of the developed in-vivo experiment device for its application to in-vivo experiments in the field of biomedical sciences.

EXPERIMENT

Animal experiment device at MC-50 cyclotron

Animal experiment devices were installed at MC-50 cyclotron LEPT (Low Energy Proton Therapy) beam line of KIRAMS (Korea Institute of Radiological & Medical Sciences). It is composed of mouse holder, ridge filter type modulator, collimator, bolus and range shifter as shown in Fig. 1. Mouse holder was made from acryl and thickness is 30mm for protect healthy tissue. We supposed that the target volume is spherical shape and produced collimator, bolus and range shifter. These was designed that may can assemble easily and have 4~13mm hole.

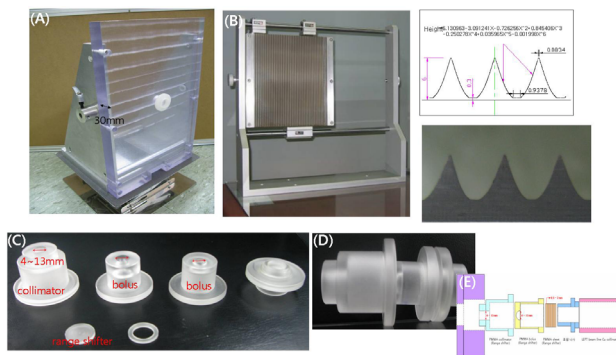


Figure 1: Manufacture of animal experiment devices. A. Mouse holder, B. Ridge filter type modulator, C.D.E. Collimator, bolus and range shifter.

In-vivo experiment

5×10^5 of LLC (Lewis Lung Carcinoma) cells were mixed with Matrigel in final volume 200 μ l (ratio media: Matrigel = 1:1) and immediately inoculated subcutaneously at the flank of C57BL.6 mice. Tumor growth were measured with calliper every 2 days using a formula, volume (V) = height x length x depth (cm^3). 8 days after inoculation, mice were irradiated with a proton beam by using animal experiment devices installed at the MC-50 cyclotron (Fig. 2). We aligned the center of the tumor tissue to the beam line center using the mouse holder. And the bolus was used for control of the penetration depth according to the shape of the tumor. Mice were irradiated at single dose level of 20 and 40 Gray.

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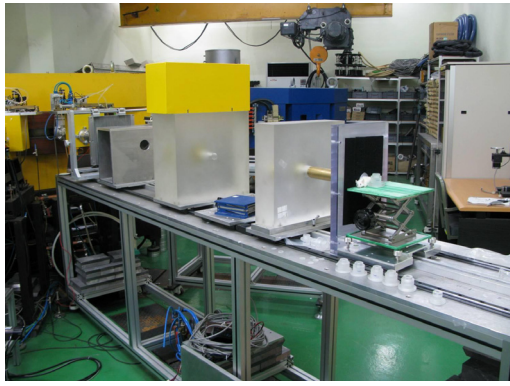


Figure 2: Photograph of animal experiment.

Srim calculation and experiment

Srim simulation results shows that 41.7 MeV proton beam was penetrated water (2.5mm) after passed PMMA (15.5mm) (Fig. 3A). We experimented in LEPT beam line to prove srim simulation. Because we use Md-55 Gafchromic film, measured penetration depth of proton beam against target volume. Fig. 3B shows that the result is similar to Srim simulation. And we measured SOBP created by ridge filter type modulator by ion chamber and confirmed ion chamber data using MD-55 Gafchromic film (Fig. 3C,D)

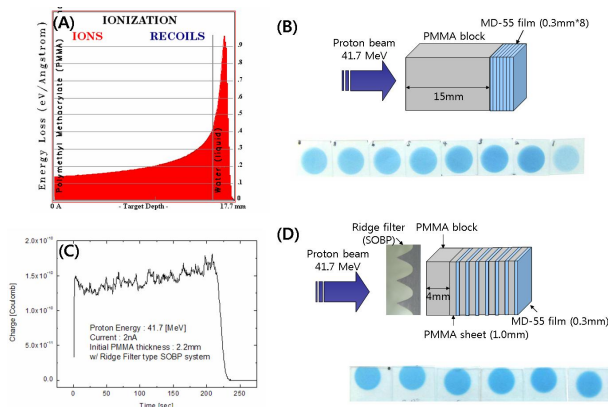


Figure 3: Srim calculation and measurement

RESULTS

Lung carcinoma cell viability is decreased by proton beam irradiation

To determine cytotoxicity of proton beam in tumor cells, we seeded tumor cells on 96-well culture plates. After 24h later, proton beam irradiated at a dose of 1 to 50 Gray. Fig. 4 shows that cell viabilities were decreased from 48h after proton beam irradiation by dose-dependent manners. With this result, we decided on total dose of in-vivo experiment.

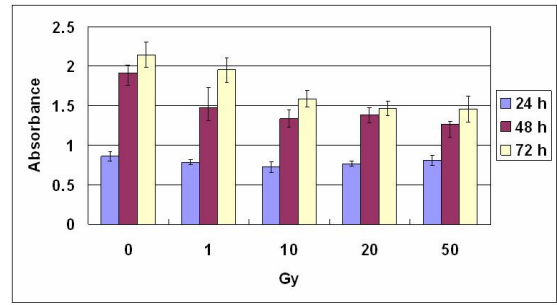


Figure 4: Effect of proton beam irradiation about cell apoptosis.

Inhibition of tumor growth in C57Bl/6 mice models by proton beam irradiation

Mice were irradiated at single dose level of 20 and 40 Gray. Control mice showed rapid increase of tumor volume. However, proton beam inhibits tumor growth with dose of 20 and 40 Gray (Fig. 5). This result indicates that animal experiment devices were successfully applicable in-vivo experiment.

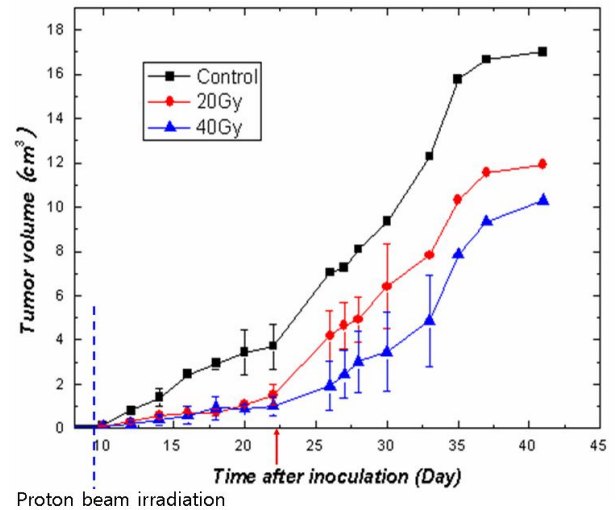


Figure 5: Data of tumor growth after proton beam irradiation

DISCUSSION AND CONCLUSION

The data show that proton beam inhibit tumor growth by dose dependent manner using animal experiment devices. However we observed that tumor growth rate is accelerated in proton irradiated groups in process of time (Fig. 5 arrow). This is conjectured that cell does not die on proton beam irradiation and survival cells grow again. But, we verified effectiveness of animal experiment system and we think that it can apply to a lot of researches of bio-medical field.

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