

DESIGN OF BIOLOGICAL RESPONSE STUDIES TO FAST NEUTRONS AT THE DONES FACILITY

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Second DONES Users Workshop



NEUTRONES PARA MEDICINA
Cátedra Universitaria



UNIVERSIDAD
DE GRANADA

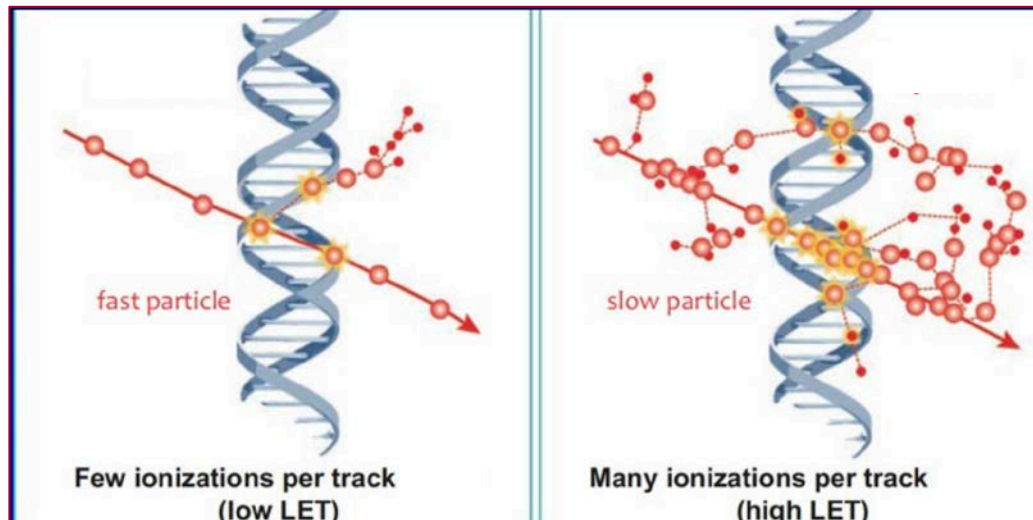
Neutrons: Biomedical applications

Particle therapy for cancer: Protons, **NEUTRONS**, helium, carbon ions

Beneficial properties:

- ❑ Decrease the radiation dose
- ❑ Decrease the irradiated volume

Ionizing radiation → Cell death
→ Genetic alterations



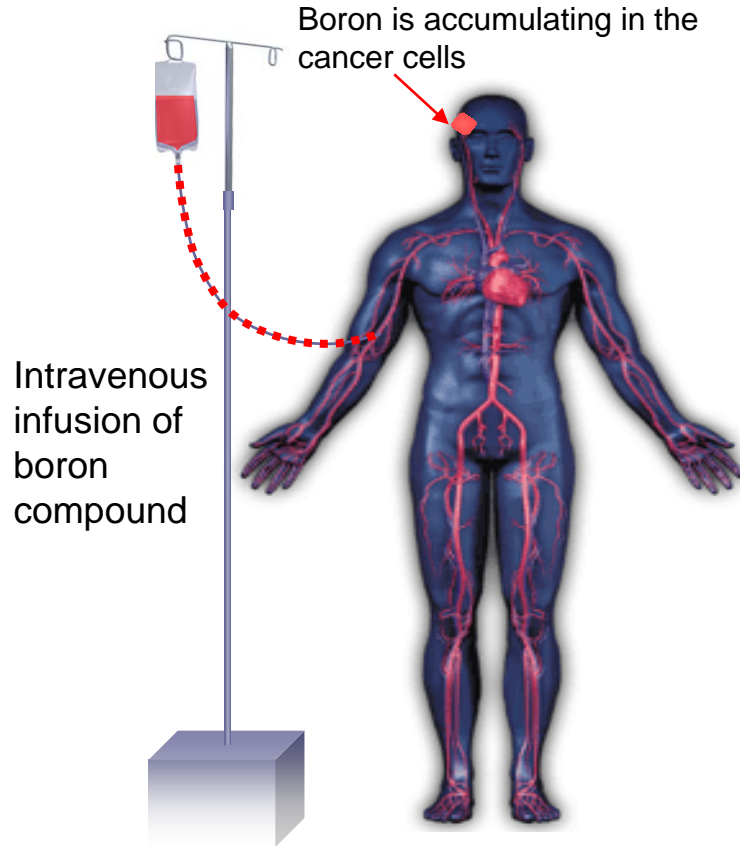
High LET (Linear Energy Transfer)

↓
Complex DSB lesions in DNA

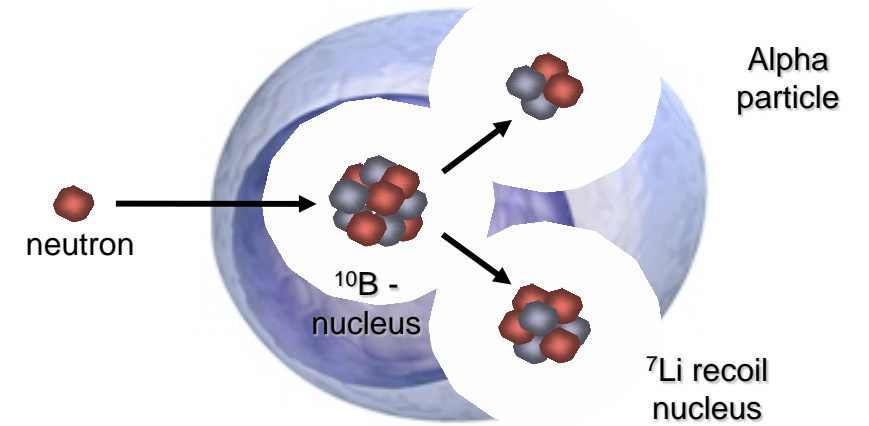
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Genomic instability, carcinogenesis

Neutrons: Biomedical applications

Boron Neutron Capture Therapy (BNCT)



Treatment Room at FIR1
Research Reactor, Helsinki



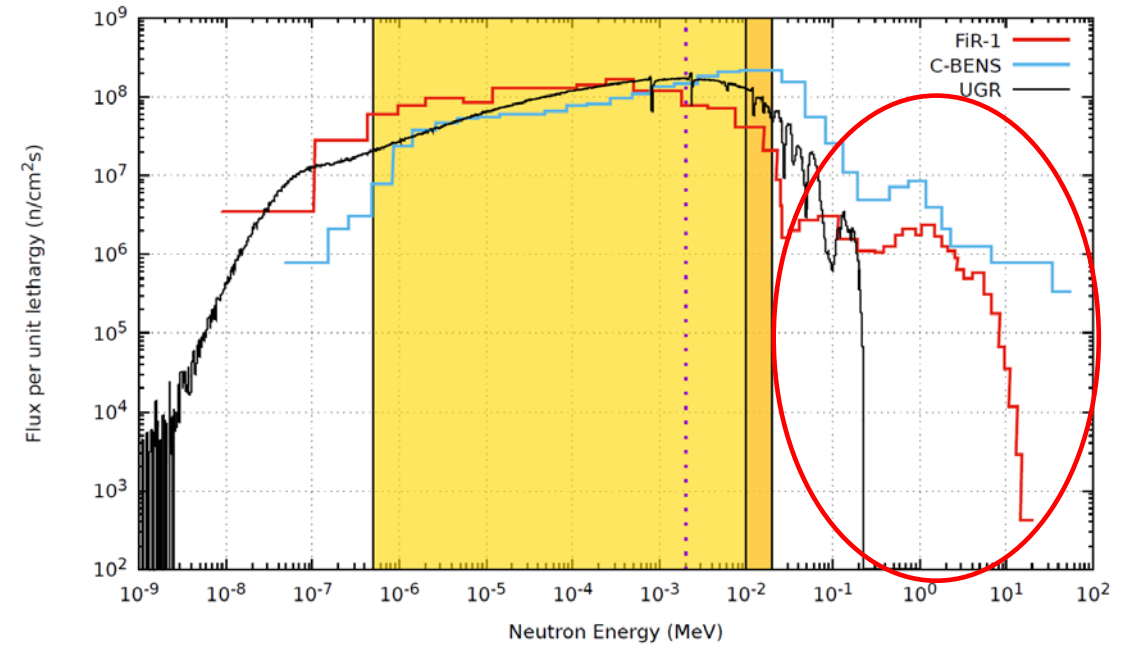
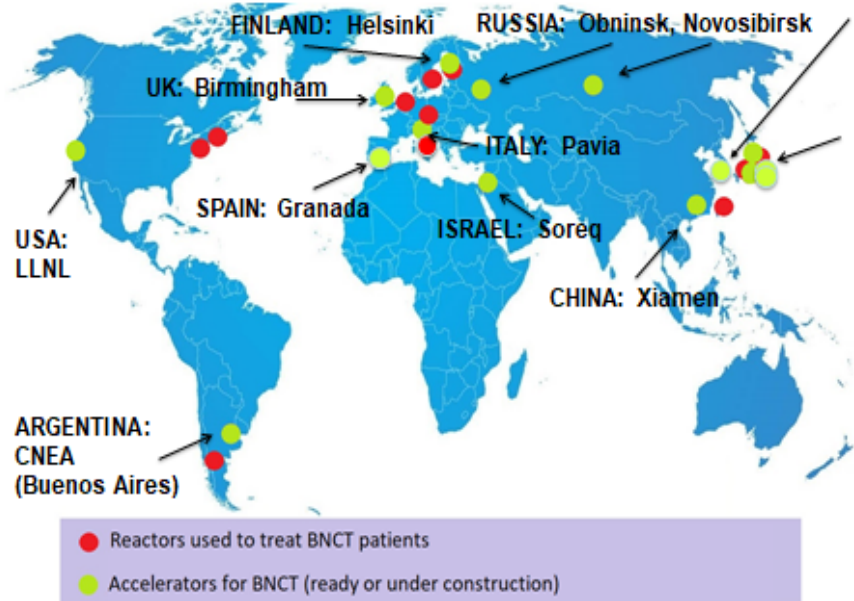
The boron atoms capture neutrons, leading to nuclear reaction emitting alpha and lithium recoil particles that cause extensive radiation damage in the cancer cells

High LET particles
Very short range → Selective at cellular level

Neutrons: Biomedical applications

Boron Neutron Capture Therapy (BNCT)

Boost of BNCT in new AB-facilities



Neutron spectra at different Accelerator-Based facilities

Neutrons: Space missions

Sources of space radiation in the solar system:

- ❑ Solar particle events (SPE)
- ❑ Galactic cosmic rays (GCR)



High-energy neutrons

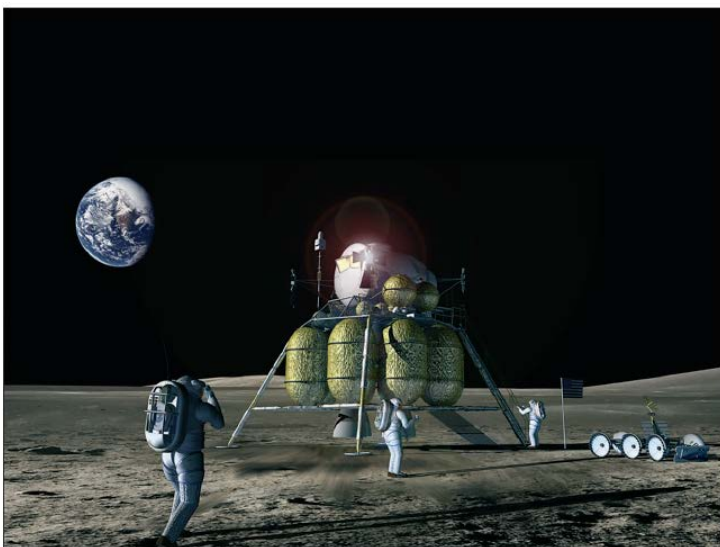
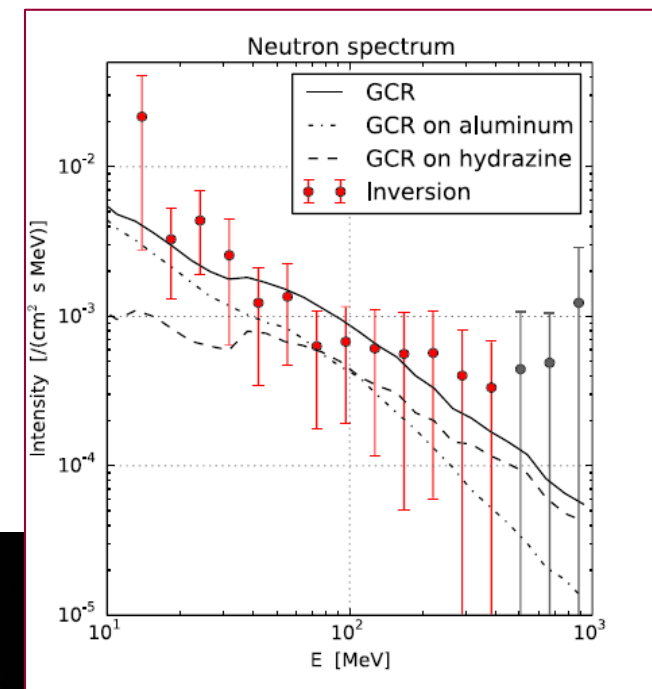


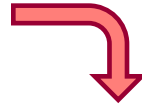
Figure 1: A future moon landing

According to the new Vision for Space Exploration (January, 2004), the National Aeronautics and Space Administration (NASA) plans to return to the moon in 2020. The present project anticipates four to six crew members who will complete lunar-surface exploration for 60–180 days. The Earth-moon cruise lasts about 4 days.



Relative Biological Effectiveness for Neutrons

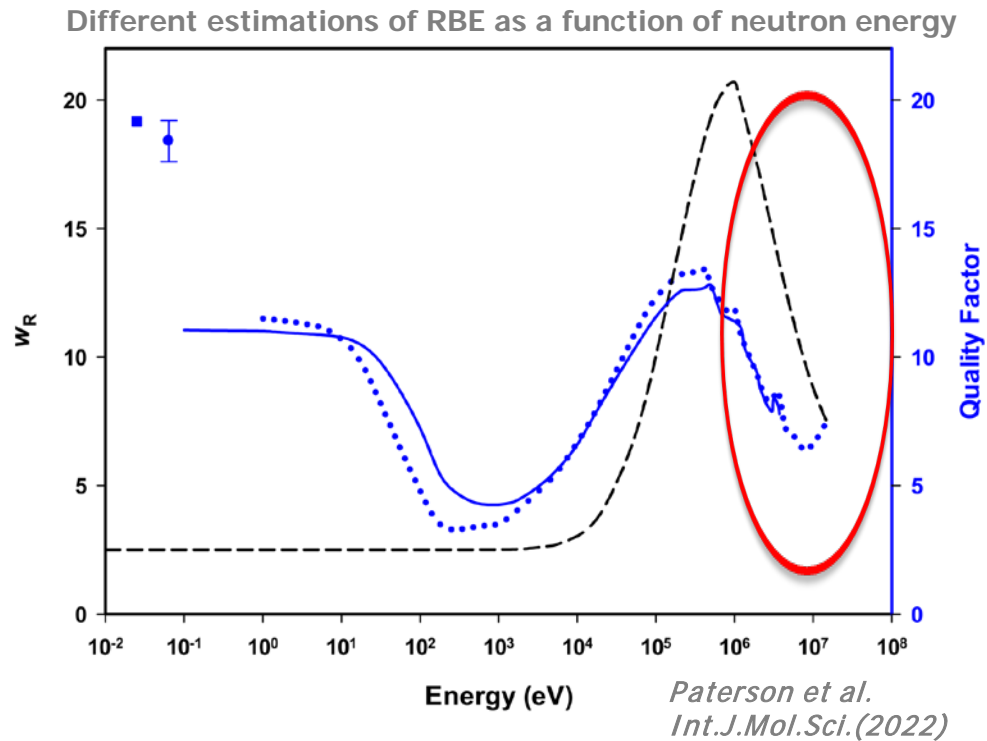
- Effects in living organism
 - Radiation protection
- RBE (Relative Biological Effectiveness) factor**



- Direct measurements, neutron irradiation
- Indirect, LET of secondary particles

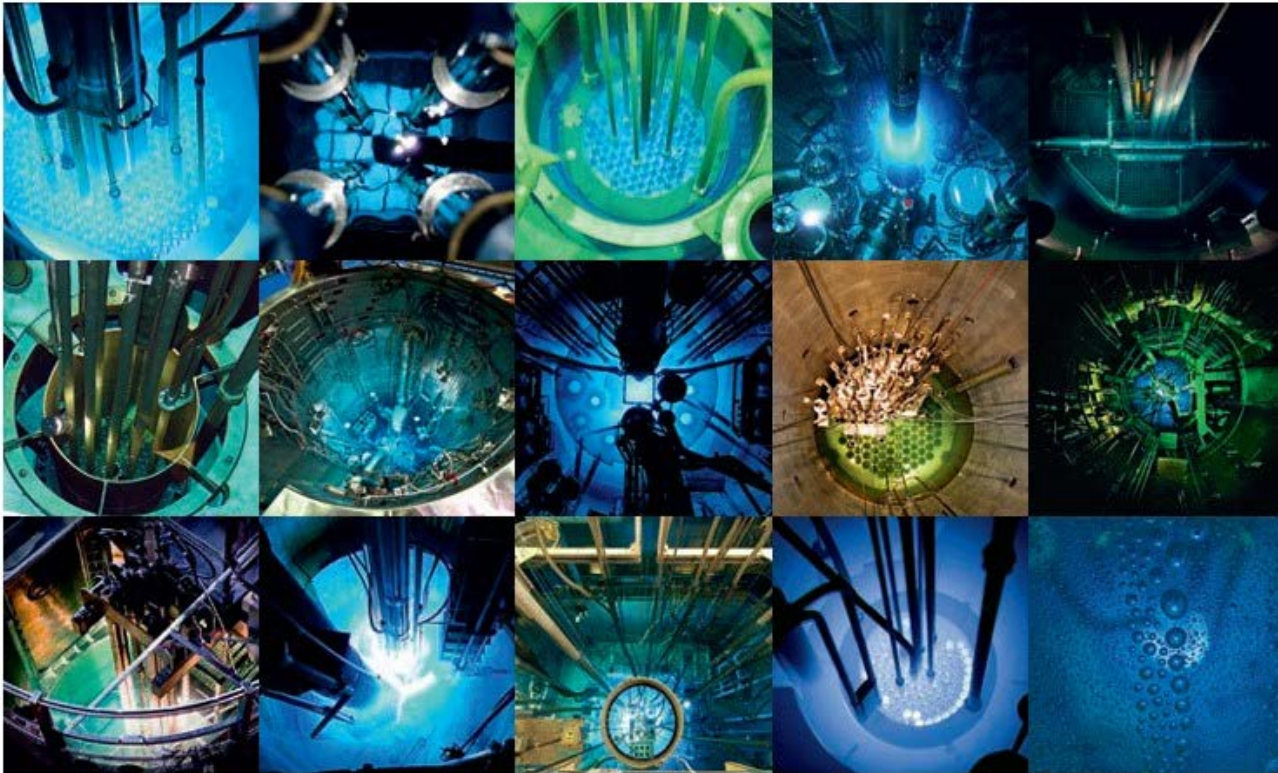
RBE for neutrons is highly energy-dependent

High uncertainties beyond 10 MeV



Neutron sources for biological response studies

Few reactors remain operational to address this type of research



The potential of **IFMIF-DONES** as a source of a **high flux of neutrons** with a **broad energy spectrum** opens an important way towards progress and improvement in the study of the biological effects of neutrons (such as those produced by cosmic radiation) and the applications they may have in several human pathologies, such as cancer.

DONES facility: Biological response studies

- ❑ Biological effects at the cellular and molecular level of radiation with different particles.
- ❑ Energy dependence studies of RBE factors of interest for neutron treatment planning and radiation protection.
- ❑ Effects of high radiation dose rate on biological organisms and obtaining data to determine the risk to humans in space missions.
- ❑ Test possible chemical compounds (e.g. sulfur) as neutron radiation shields.
- ❑ Relationships between RBE and LET, of interest in ion therapies.



Radiobiology laboratory



DONES facility: Biological response studies



Irradiation of biological samples (cell lines)

■ Cell culture laboratory



■ Equipment for a variety of techniques:

- Colorimetric assays
- Clonogenic assays
- Invasion assays
- Comet assay
- Flow cytometry
- Immunofluorescence & confocal microscopy
- Western-blot
- RT-PCR
- Fluorescence *in situ* hybridization (FISH)...

DONES facility: Biological response studies

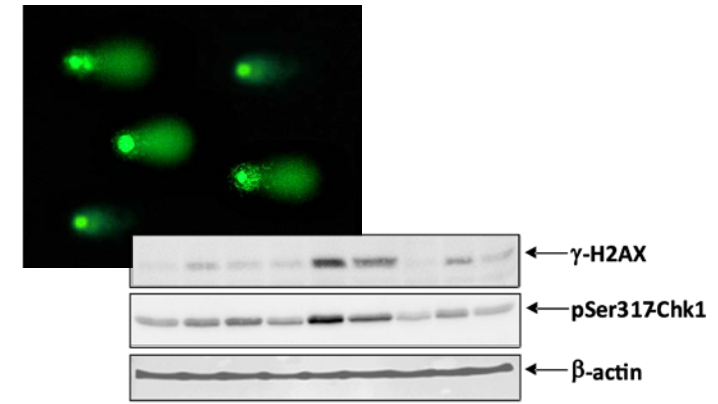
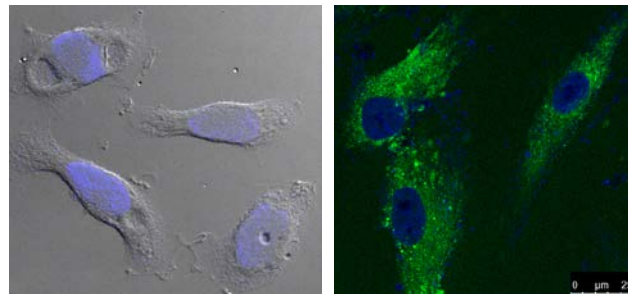
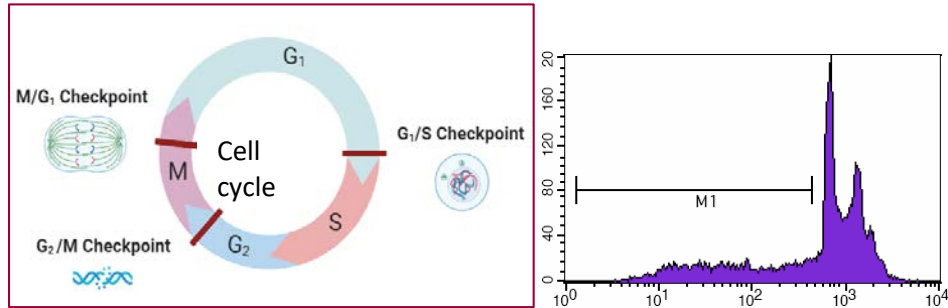
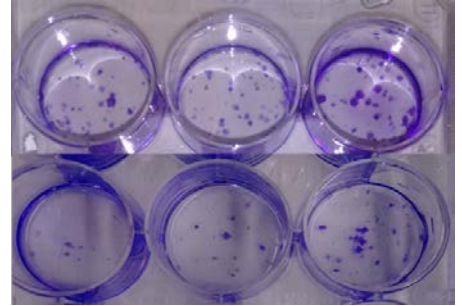
End-points after irradiation:

- ❑ Proliferation
- ❑ Survival / Cell death
- ❑ Clonogenic ability
- ❑ Invasion capacity
- ❑ DNA damage
- ❑ Activation of DNA repair pathways
- ❑ Chromosome aberrations
- ❑ ROS production
- ❑ Exosomes production
- ❑ Cytokines expression

➔ Data for calculation of the **RBE factors**

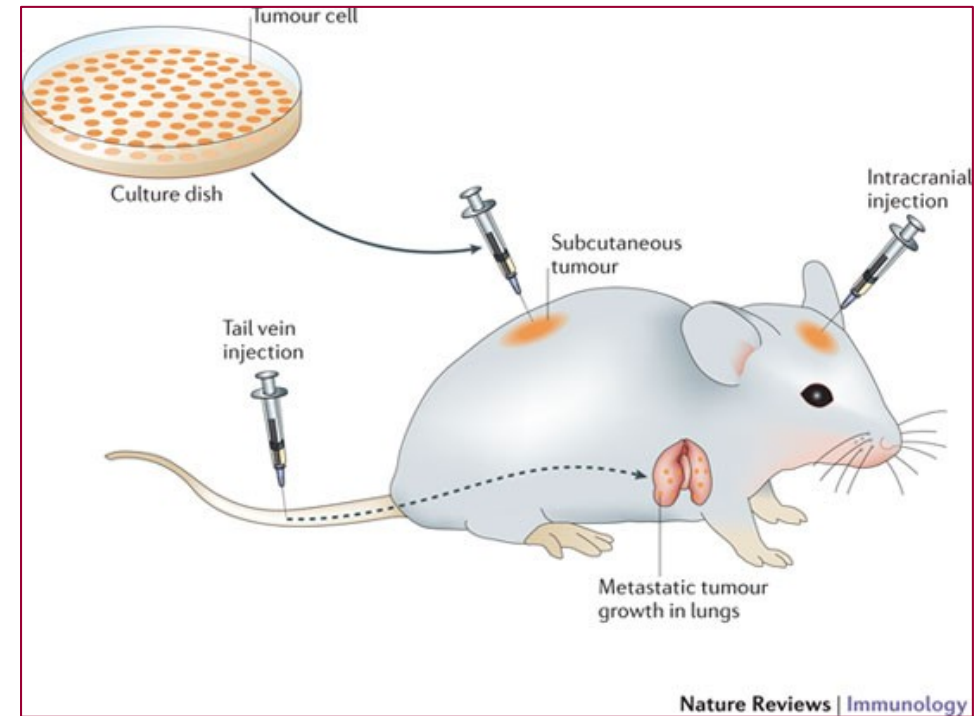
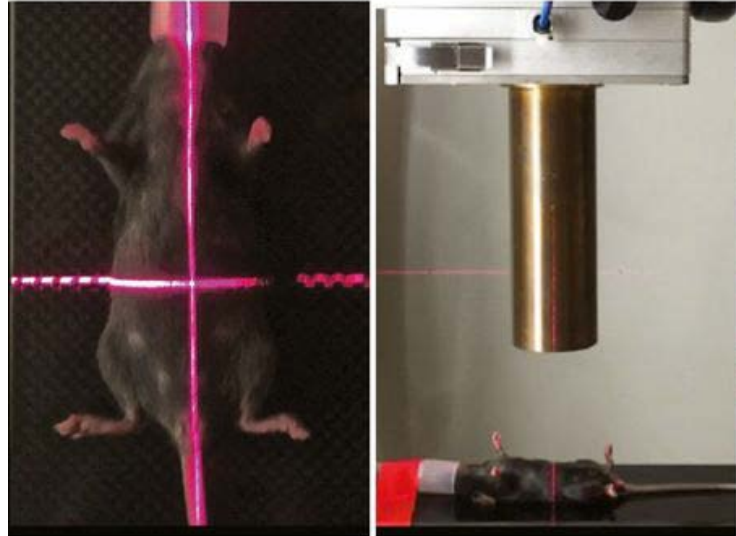
➔ Cellular alterations

- Compromise the viability of irradiated cells
- Effects on neighbouring non-irradiated cells: **bystander effect**
- Regulation of the immune system: (distant) **abscopal effect**



DONES facility: Biological response studies

In vivo studies: healthy and tumor mouse models



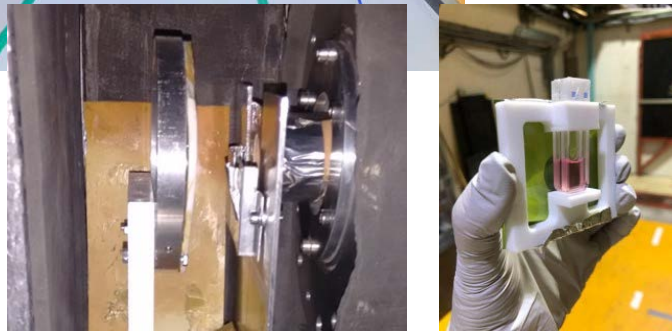
Design of biological response studies → DONES

An interdisciplinary group with previous experience in neutron radiobiology studies

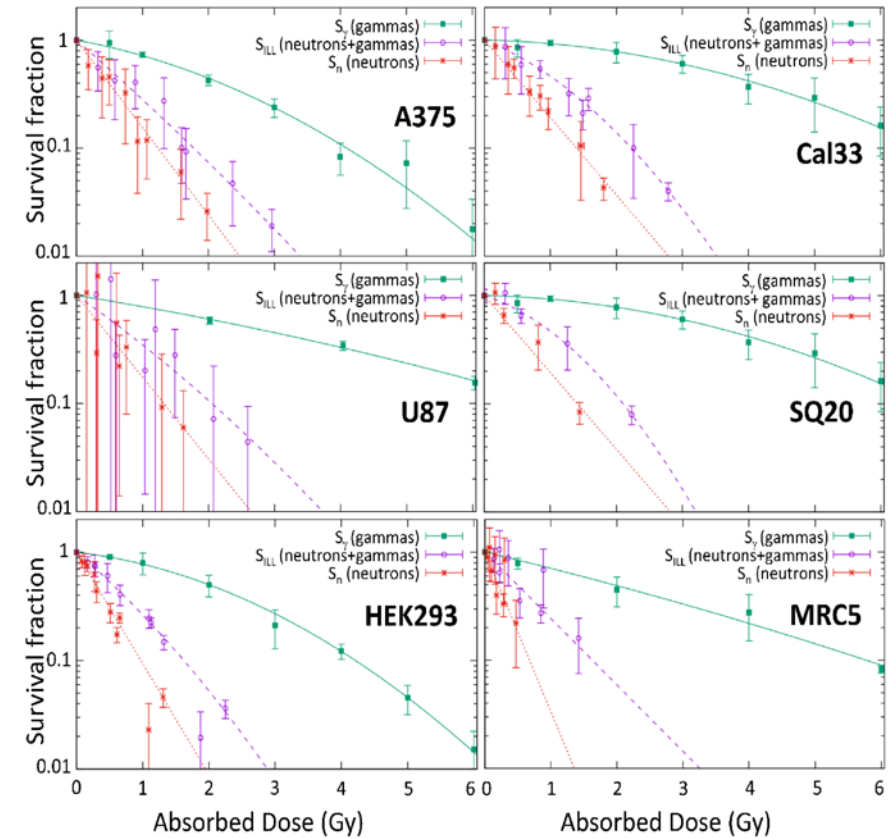
Institut Laue Langevin (ILL, Grenoble) - PF1b neutron line



Culture laboratory



Thermal neutrons

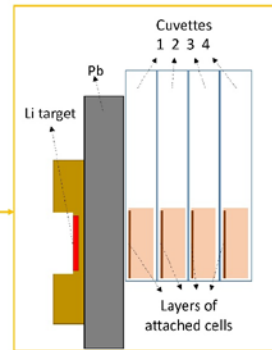
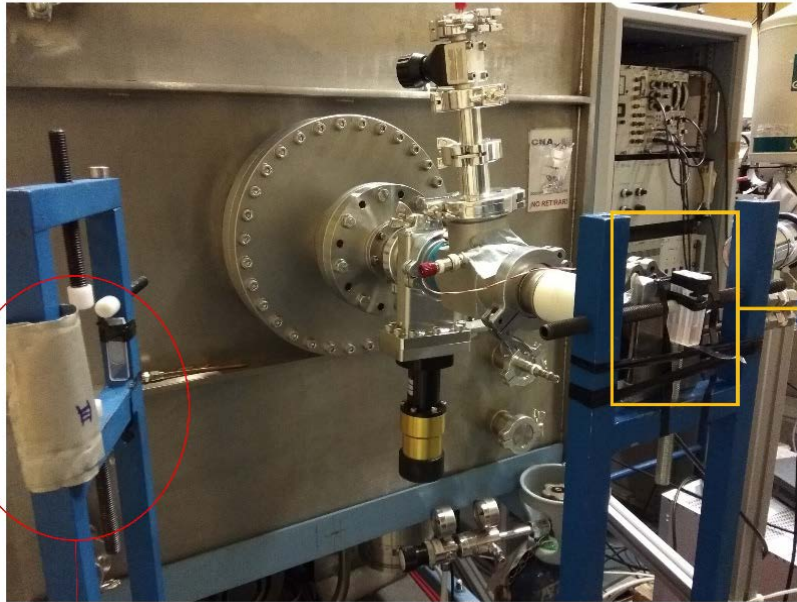


Cells 2020, 9(10), 2144; <https://doi.org/10.3390/cells9102144>

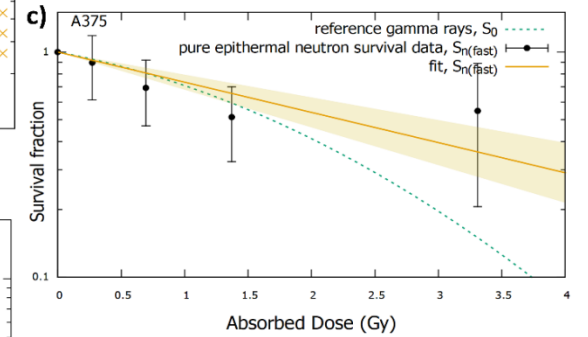
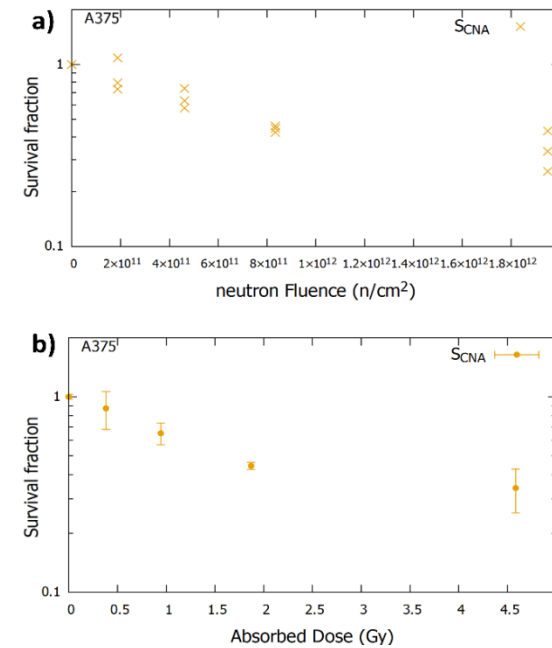
Design of biological response studies → DONES

An interdisciplinary group with previous experience in neutron radiobiology studies

National Accelerator Center (CNA , Sevilla)



Control cuvettes outside the beam



Near-epithermal fast neutron

Design of biological response studies ➔ DONES

An interdisciplinary group with previous experience in neutron radiobiology studies

Linear Accelerator

Hospital Universitario Virgen de las Nieves, Granada

J. Expósito-Hernández

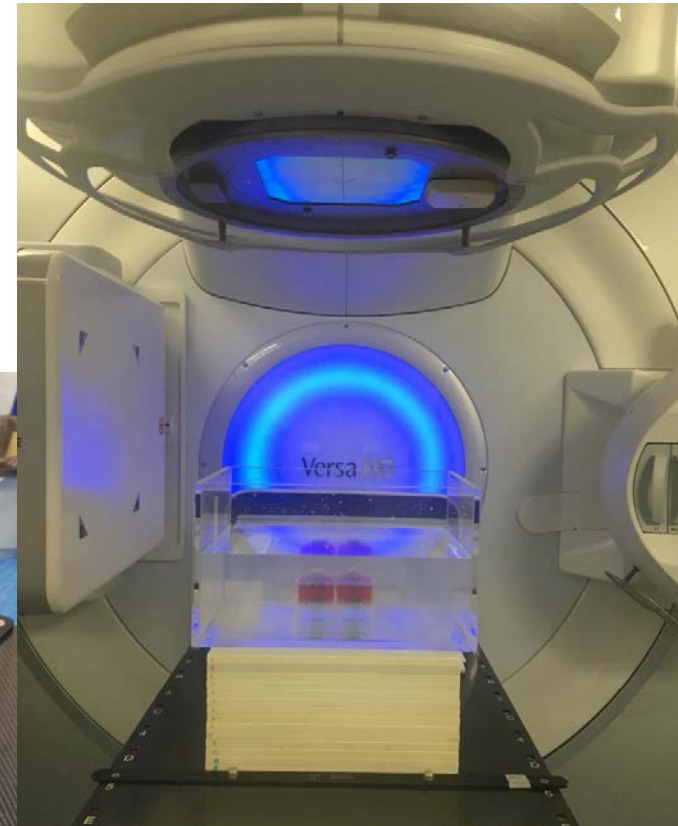
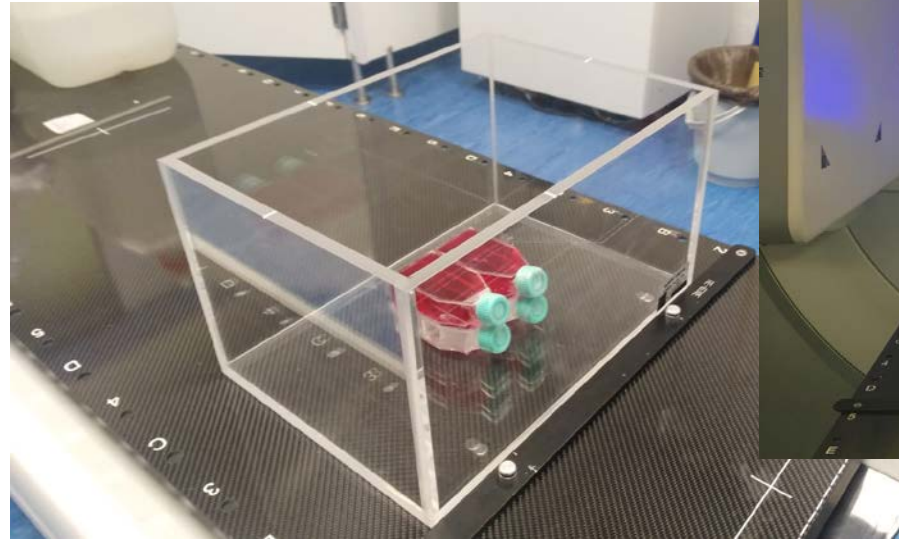
J.M. Llamas-Elvira

J.L. Osorio

R. Estrada

O. Liñan

Reference photon dose

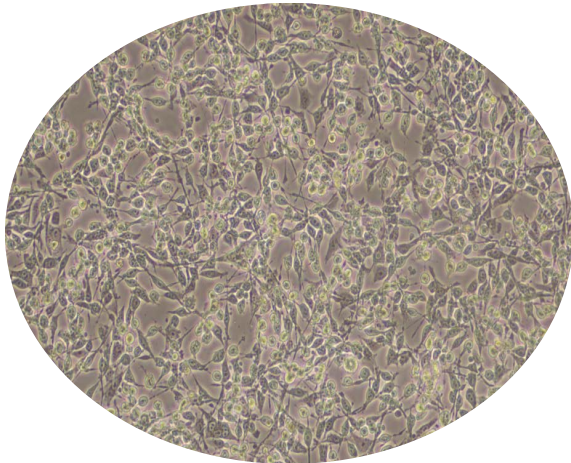


Design of biological response studies → DONES

CIEMAT (Madrid): Different fast neutron sources (^{252}Cf , $^{241}\text{Am/Be}$, others)

^{252}Cf , Neutron pattern laboratory (LRMI-3)

A375 cells (melanoma cell line)



Ciemat

Centro de Investigaciones
Energéticas, Medioambientales
y Tecnológicas

M. Oteo

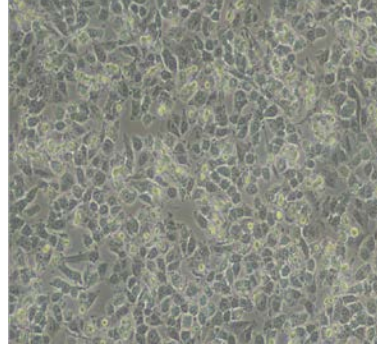
M.A. Morcillo

R. Méndez

M. Embid

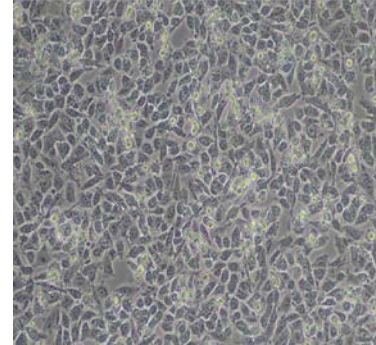
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Control cells

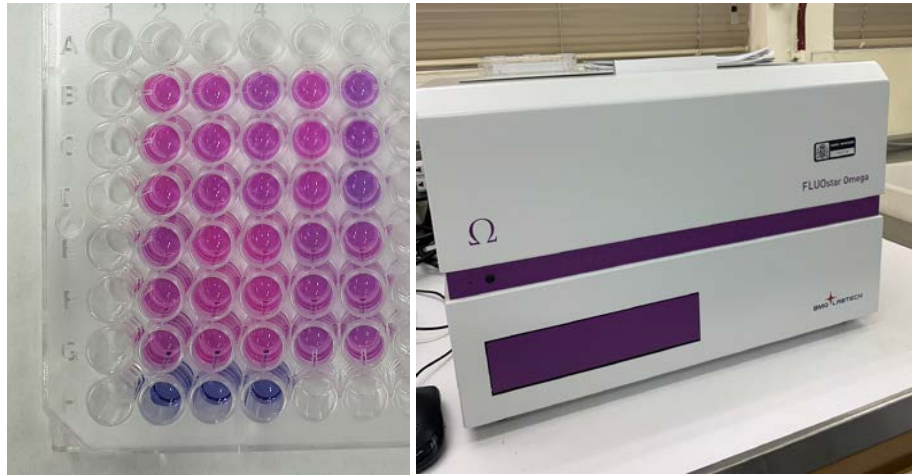


Irradiated cells

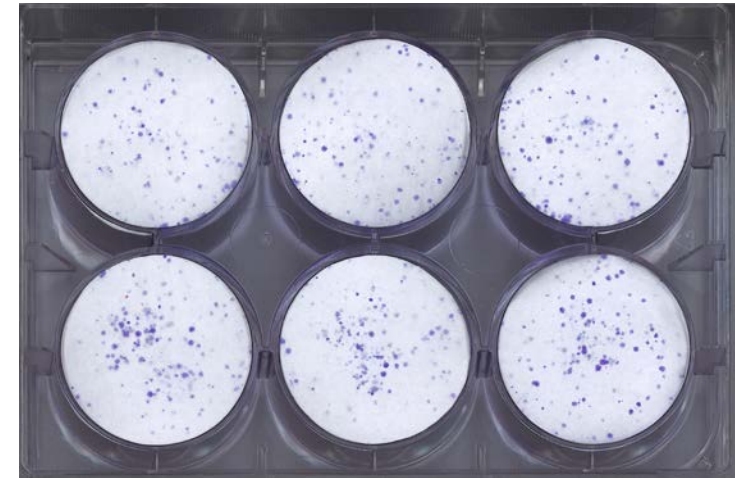
(0.4, 0.8 and 1.2 Gy)



Colorimetric viability assays (4 days post-irradiation)

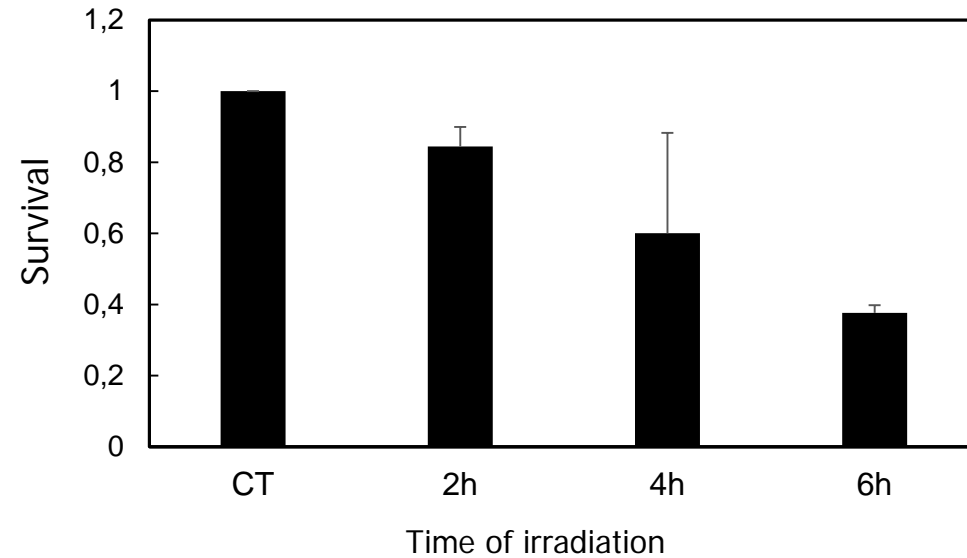


Clonogenic assays

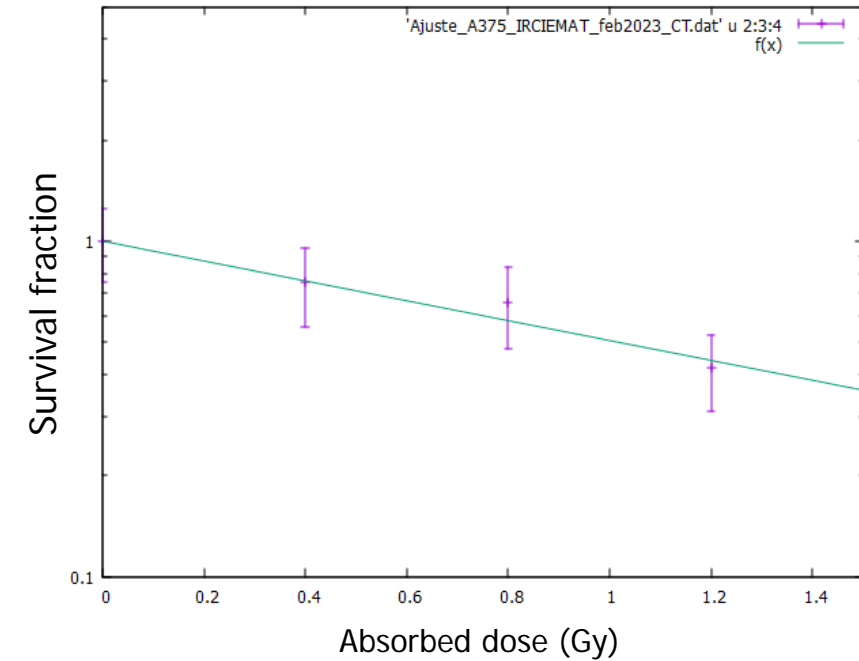


Design of biological response studies → DONES

Survival of A375 cells (clonogenic assay)



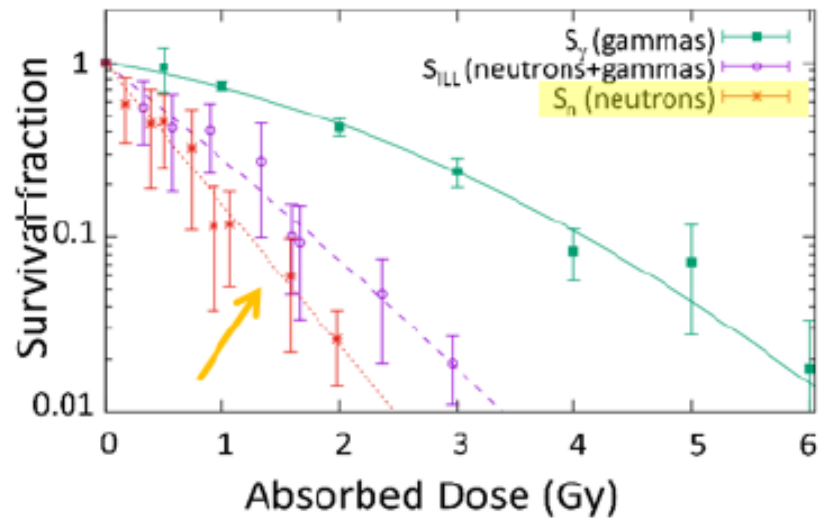
Survival fraction as a function of the absorbed dose



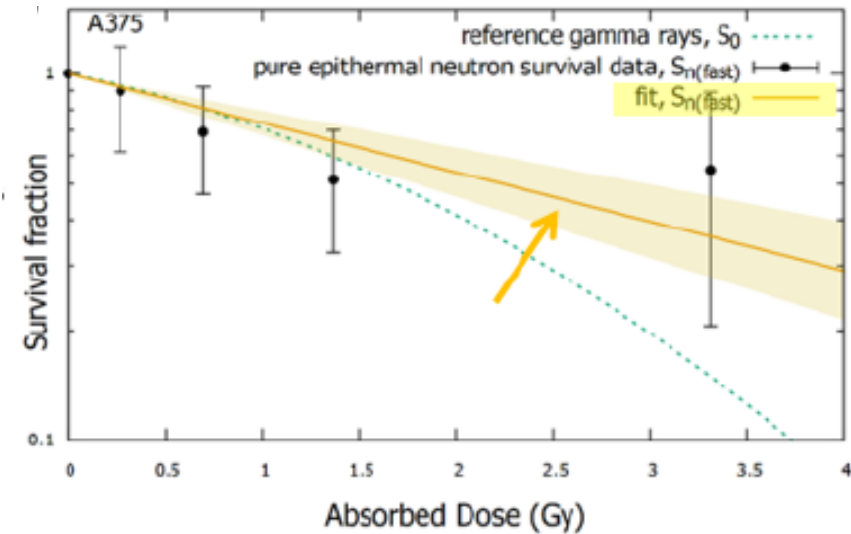
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Comparison of dose-survival curves

A375 cells irradiated with **thermal neutrons** (ILL)



A375 cells irradiated with **near-epithermal fast neutrons** (CNA)



Graphs show the reference data obtained by irradiations with **gamma** rays at LINAC (HUVN)

Design of biological response studies → DONES

Comparison of the α coefficients of dose-survival curves

| Irradiation of A375 cells | Energy | α Coefficient |
|---|------------------|--|
| Thermal neutrons (ILL, Grenoble) | 0,025eV | 0.84 +/- 0.05 * |
| Near-epithermal fast neutrons (CNA, Sevilla) | 10 KeV – 100 KeV | 0.31 +/- 0.08 * |
| Fast neutrons (CIEMAT, Madrid) | 2,2 MeV | 0.68 +/- 0.04 |

SUMMARY AND CONCLUSIONS

Studies of the biological response to fast neutrons are key for:

- ❑ Biomedical applications
- ❑ Radiation protection assessments for space missions

A Bio Lab in IFMIF-DONES will allow:

- ❑ To study the effects of high dose rate radiation in biological organisms
- ❑ To carry out studies of the energy dependence of the RBE factors

Radiobiology laboratory
at the DONES facility

UGR interdisciplinary group with experience in neutron radiobiology:

- ❑ Thermal neutron, LL, Grenoble (a Bio Lab at the experimental hall)
- ❑ Near-epithermal fast neutrons, CNA, Sevilla
- ❑ Fast neutrons, CIEMAT, Madrid