Electron dose rate and oxygen depletion protect zebrafish embryo from radiation damage

Elke Beyreuther

Very High Energy Electron Radiotherapy Workshop (VHEE'2020)
Oct 5th – 7th 2020
CERN/Switzerland
Adding up the benefits to broaden the therapeutic window

- Tumor conform irradiation
- Differential tissue radiosensitivity

Sparring the normal tissue to broaden the window

**Response probability (%)**

- **Tumor control**
- **Normal tissue complication**
- **Therapeutic window**

Holthusen 1928
Adding up the benefits to broaden the therapeutic window

**Tumor conform irradiation**
Beneficial depth-dose distribution of protons spares normal tissue

**Differential tissue radiosensitivity**
High dose rate (Flash) beam delivery protects normal tissue, but treats tumours efficiently

---

Vozenin et al. Clin Cancer Res 2019
Proton Flash experiment in Dresden - Setup

- University Proton Therapy Dresden, $E_p = 70-230$ MeV
- Flash: 228 MeV, 95 nA $\rightarrow$ 100 Gy/s
- Continuous: 228 MeV, 0.3 nA $\rightarrow$ 0.08 Gy/s

Transmission proton beam @entrance plateau
Online dose control:
  Two Transmission ionisation chambers
Absolute dose:
  Markus chamber and radiochromic EBT3 films

Irradiation geometry

Zebrafish embryo, wildtype AB
- Doses $\leq 43$ Gy, treatment time $\sim 420$ ms (Flash)
- Survival and morphological malformations up to four days post irradiation

Dose homogeneity within beam spot

Zebrafish embryo, wildtype AB
- Doses $\leq 43$ Gy, treatment time $\sim 420$ ms (Flash)
- Survival and morphological malformations up to four days post irradiation
Proton Flash experiment in Dresden - results

No significant protecting proton Flash effect for zebrafish embryo

Why? What are the differences to other electron/proton Flash experiments?
Proton No-Flash: Biological model and endpoint?

- Majority of Flash experiments performed with mice or other higher vertebrates
- **Zebrafish embryo model** successfully applied, but
  - 4 hpf vs. 24 hpf → defines radiosensitivity (dose)

Morphological malformations in general vs. embryo length

Vozenin et al. Clin Oncol 2019
Proton No-Flash: Beam and pulse parameters?

Recipe for electron Flash-RT published in Wilson et al. (Frontiers in Oncology, 2020):

Proton Flash

Mean dose rate: 100 Gy/s ✓
Pulse dose rate: $10^3$ Gy/s ❌
Total delivery time: ~400 ms ✓
Pulse length: 2 ns

Relevance?
Proton No-Flash: No control of partial oxygen pressure

- Oxygen depletion theory of Flash effect:
  - Physoxia in tissue vs. uncontrolled partial oxygen pressure in zebrafish embryo (in medium)

- Zebrafish embryos irradiated in Eppendorf tube
  → Accidentally too much / less oxygen?

We need to control the oxygen partial pressure somehow!?

Wilson et al. Frontiers in Oncology, 2020; modified
Electron Flash experiment @ELBE

Research electron accelerator with highly variable pulse structure

Pulse structure @ELBE:
- Basic frequency: 13 MHz
- Bunch length: 5 ps
- Bunch interval: 77 ns

See Talk of U. Schramm@06.10.2020

Reference
- Quasi-continuous bunches at 13 MHz
- Mean dose rate: 7 Gy/min
- Pulse dose rate: $10^3$ Gy/s

Flash irradiation
- One pulse of 1441 bunches
- Mean dose rate: $2.6 \times 10^5$ Gy/s
- Pulse dose rate: $10^9$ Gy/s
Electron Flash experiment @ELBE – oxygen pressure

- Daily measurement under experiment conditions
- High \( pO_2 \): 25-100 mmHg
- Low \( pO_2 \): Radiobiological hypoxia (<5 mmHg)

**OxyLite™**
Oxford Optronix

**Zebradish embryo**
Electron Flash experiment @ELBE – results

• No significant influence on survival by electron dose rate

4 days post irradiation: radiation effects

Flash vs. cw:
- 20% less pericardial edema
- 25% less embryo with curved spine
+ 4 % longer embryos (p<0.001) with larger eyes
General outcome:
Embryos are significantly longer after Flash irradiation ($p<0.001$)

- **The lower the oxygen pressure, the more pronounced is the Flash effect**
- **The older the embryos the more pronounced is the Flash effect**
Radiation response of zebrafish embryo depend on electron dose rate

- Very high doses required for ≥ 24 hpf embryos
- Very high pulse dose rate of $10^9$ Gy/s
- Average dose rate of $10^5$ Gy/s
- Whole organism response, individual organs hard to investigate
- Are these dose rates required to induce the Flash effect in zebrafish embryo?

Control of partial oxygen pressure intensifies the Flash effect

- Protecting effect measured for oxygen levels below atmospheric levels, but most pronounced at radiobiological hypoxia
- Partial oxygen pressure inside embryo?
Repetition of proton Flash experiment under controlled oxygen conditions, @ University Proton Therapy Dresden, in preparation

Proton Flash experiment at very high pulse dose rates @ Laser proton accelerator Draco (cf. Talk U. Schramm)
- Controlled oxygen conditions
- Single, ns-long laser pulse of $10^9$ Gy/s
- In preparation
Acknowledgments

OncoRay/ Group of Laser-Radiooncology
Elisabeth Bodenstein, Leonhard Karsch, Elisabeth Lessmann, Michael Schürer, Jörg Pawelke

Other OncoRay groups
Experimental Radiation Therapy group: Kerstin Brüchner, Mechthild Krause, Liane Stolz-Kieslich, Katja Schumann, Anne Kluske, Elisabeth Jung, Dorothee Pfitzmann
Medical Radiation Physics: Wolfgang Enghardt, Steffen Löck
Universitäts Protonen Therapy Dresden & IBA crew

HZDR, Institute of Radiation Physics
ELBE operator crew: Peter Michel, Ulf Lehnert, Pavel Evtushenko, Rico Schurig
Group of Laser ion acceleration & Draco laser team

TU Dresden/CRTD
Group of Developmental Genetics: Michael Brand, Stefan Hans

ELI-Alps /Hungary: Rita Emilia Szabo, Katalin Hideghety