



The  
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# **Elemental imaging via LA-ICP-MS**

## **New opportunities in the life sciences**

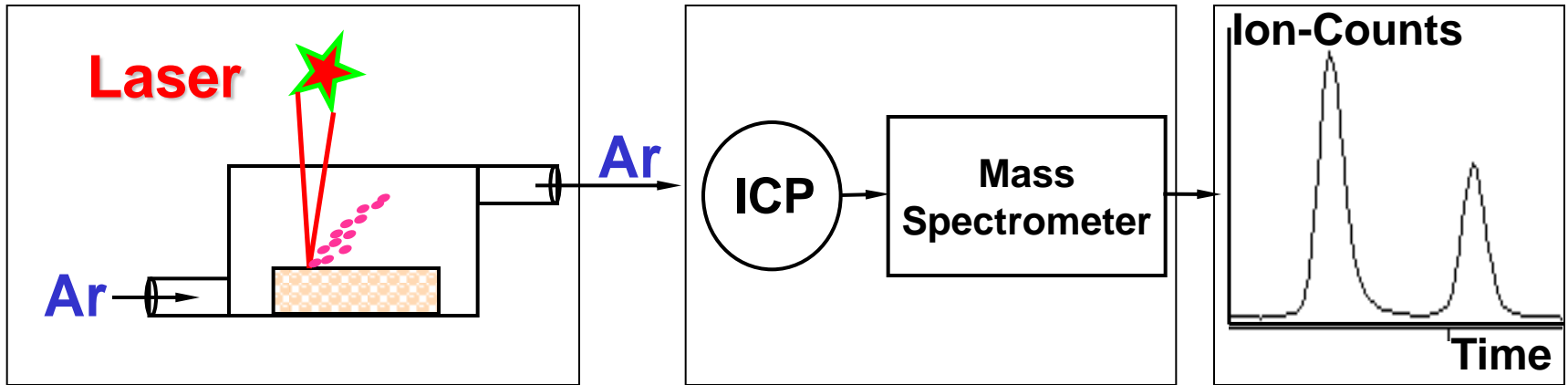
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# Contents

- LA-ICP-MS imaging overview
- MRI contrast agents
- Signal Quantitation
- Gd-tagged vascular disrupting agent
- Comparison with MRI
- Conclusions

# LA-ICP-MS Overview



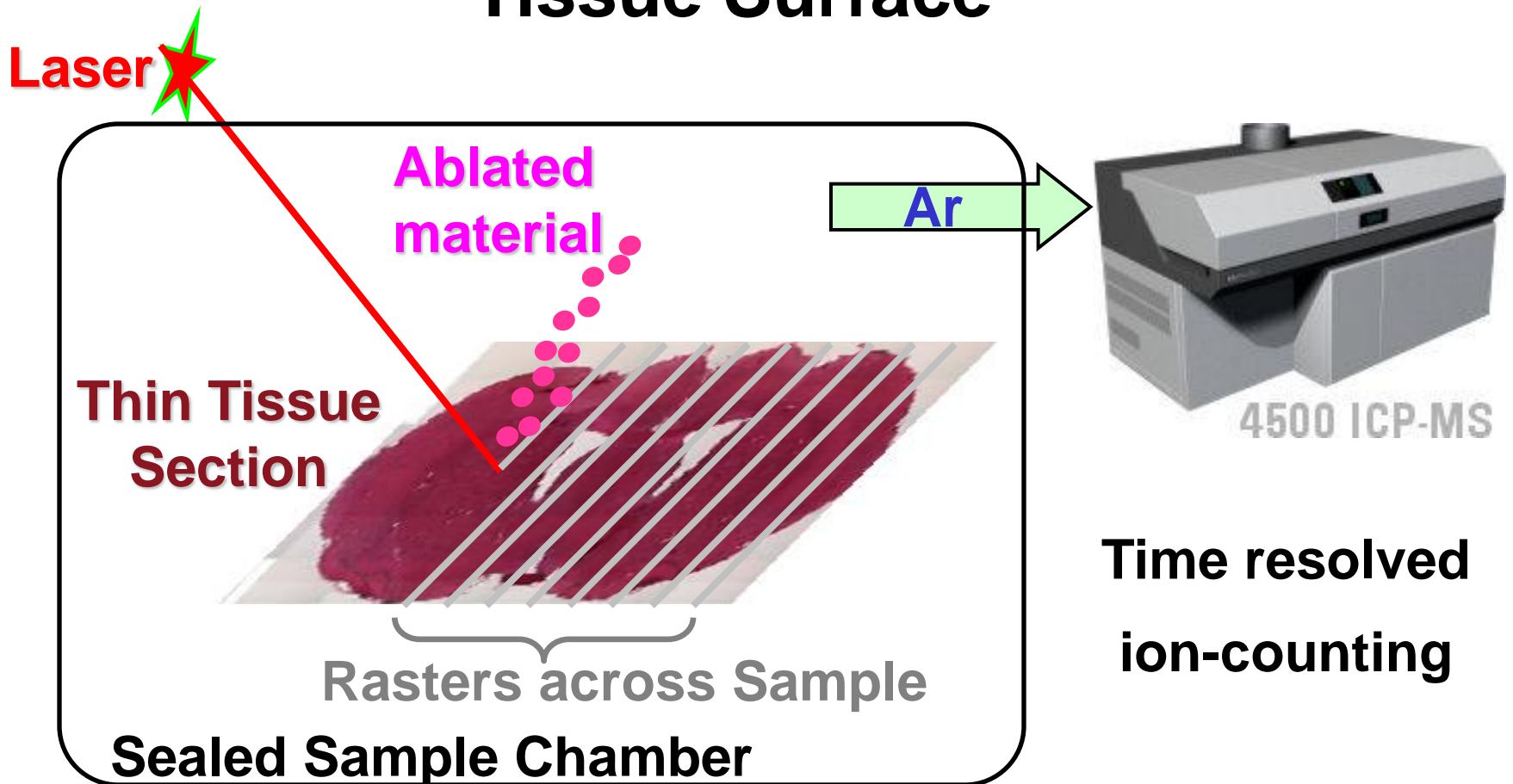
**Sample interrogation**

**Detection**

**Data**

- Elemental surface analysis technique.
- Laser used to vaporise sample.
- Ablated material is transferred to ICP-MS.
- Selected elements are detected/ measured in a time resolved mode.

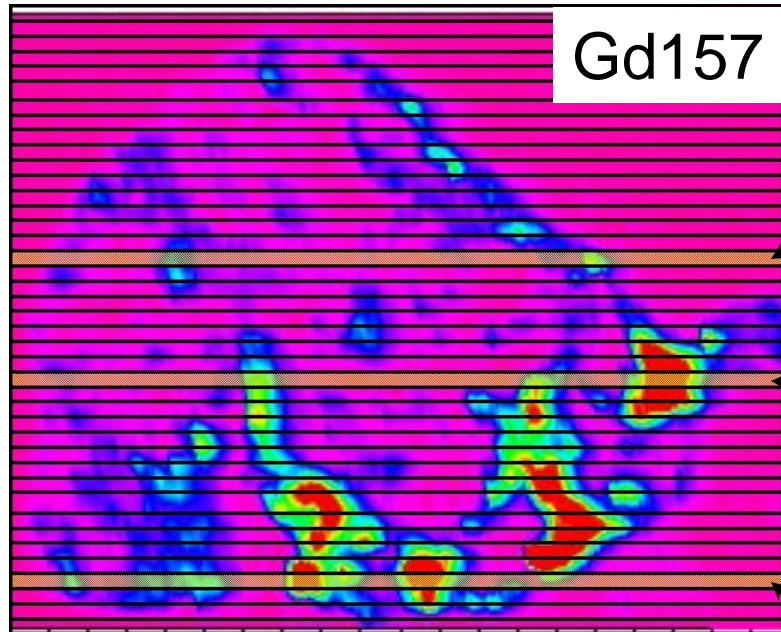
# Multiple Line Rastering Across Tissue Surface



- Specific time points will relate to laser position (x,y coordinate) on sample.

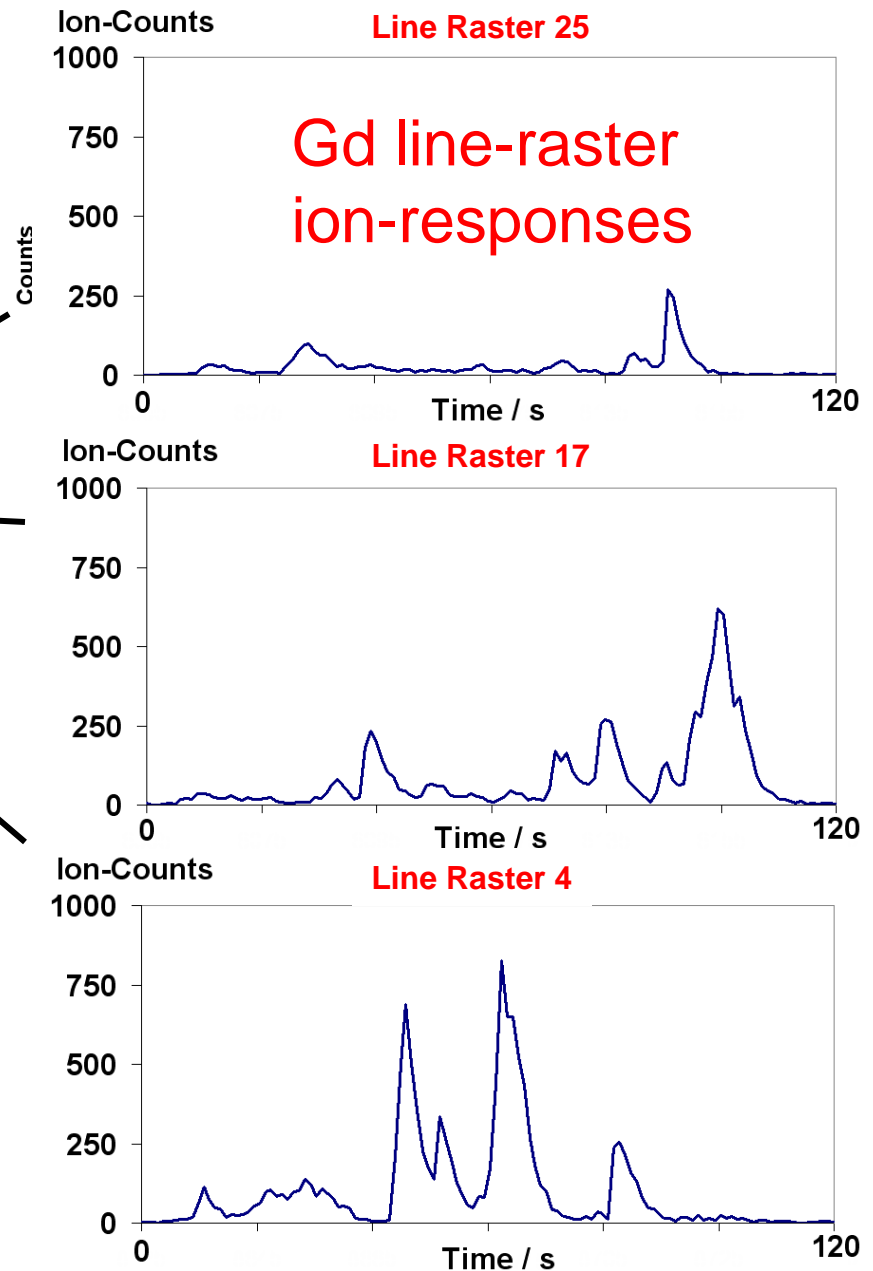
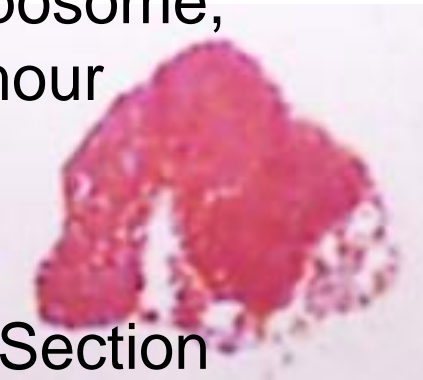
# Distribution Map Construction

LA-ICP-MS Distribution Map



GdDOTA-Liposome,  
Treated Tumour

Histological Section



# MRI Contrast Agents

- Used in MRI to enhance **image contrast**
- MRI measures relaxation of nuclei in a magnetic field following RF pulse
- **Paramagnetic** atoms (Fe, Gd, Mn) affect proton relaxivity
- Contrast imaging

**Tumours**

**Neural pathways and brain activity**

**Liver and kidney**

Gastrointestinal tract

Heart ischemia and blood vessel constrictions

# MRI Contrast Agents

- Three main elements : Mn(II), Gd(III), Fe(III)

## Gd(III) chelates

- Most commonly available contrast agents  
Magnevist, MultiHance, Omniscan, Gadovist, etc.
- Main applications as extracellular (e.g. tumour detection) and blood pool (vascular structure) agents.
- Novel targeted treatments currently under research.

# The Need for Quantitative Information on Contrast Agents in Tissue

- To enable better understanding of cellular chemistry of CA and how this correlates with MRI response.
- Better understanding of effective agent dose for *in vivo* experiments.
- ICP may be a valuable tool in understanding CA tissue concentration and distribution



# Quantitative LA-ICP-MS

Calibration standards for LA tissue imaging must demonstrate:

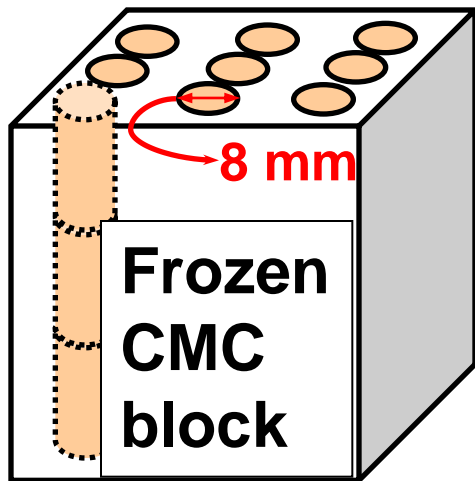
- Similarity with biological matrix (matrix-matching)
- Comparable section thickness (amount of material sampled)
- Homogenous elemental distribution

The following strategies were explored:

- a) Homogenised spiked tissue
- b) Spiked blood standards in drilled teflon
- c) Sectioning spiked blood standards in tygon tubing
- d) Sectioning lyophilised spiked serum in CMC blocks

# Quantitation – Gd Spiked Serum

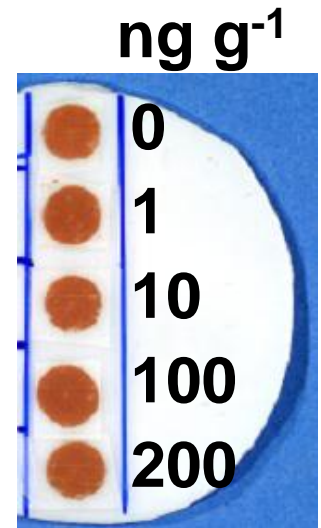
- Adapted from quantitative whole body autoradiography
- 2% carboxymethylcellulose (CMC) solution is frozen (-20°C) into a block
- Block machined with series of holes
- Serum/blood, spiked with Gd (ICP standard) solutions
- Solutions added to holes and frozen



• Section onto tape

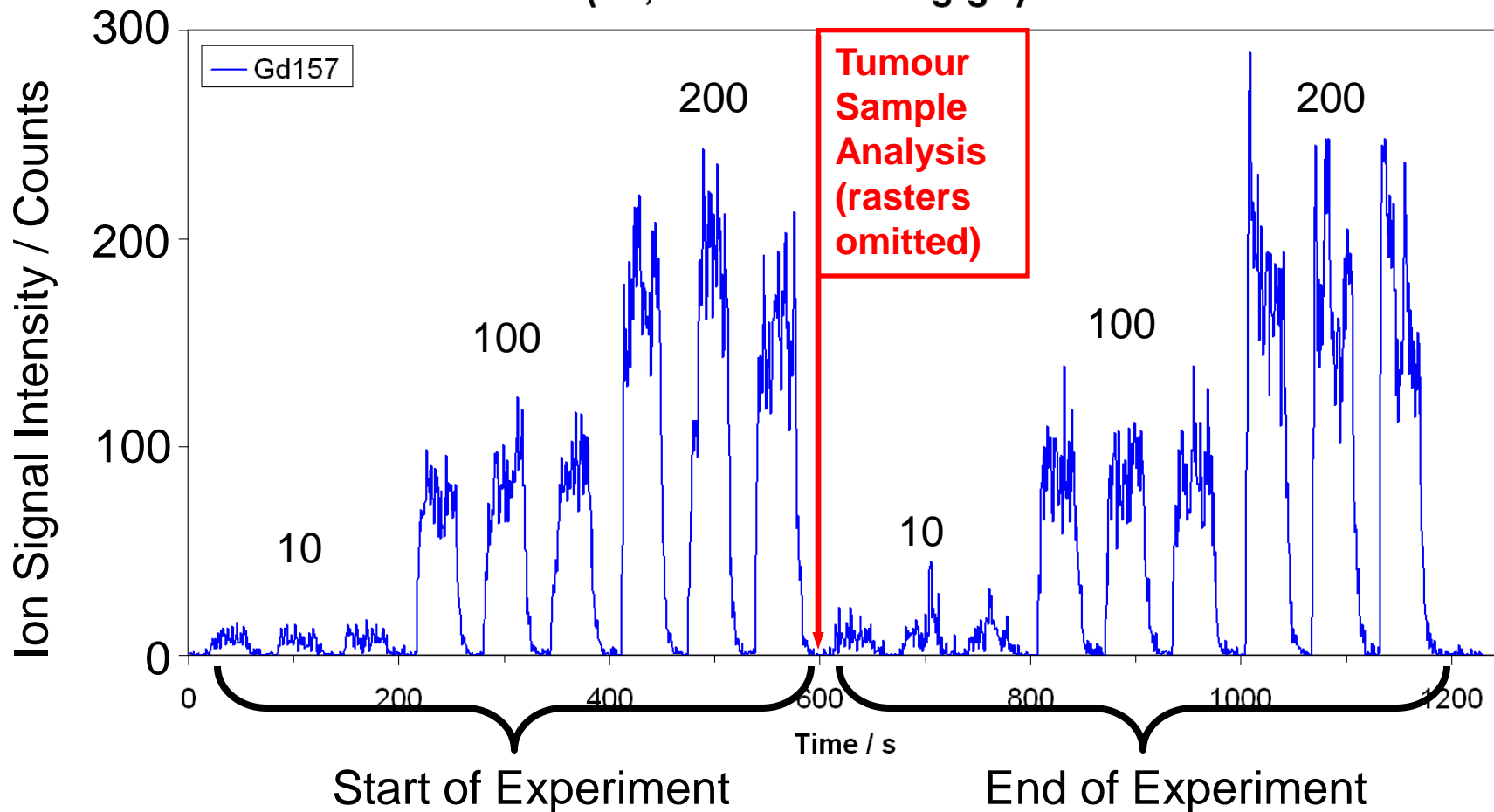


• Place on Teflon disc



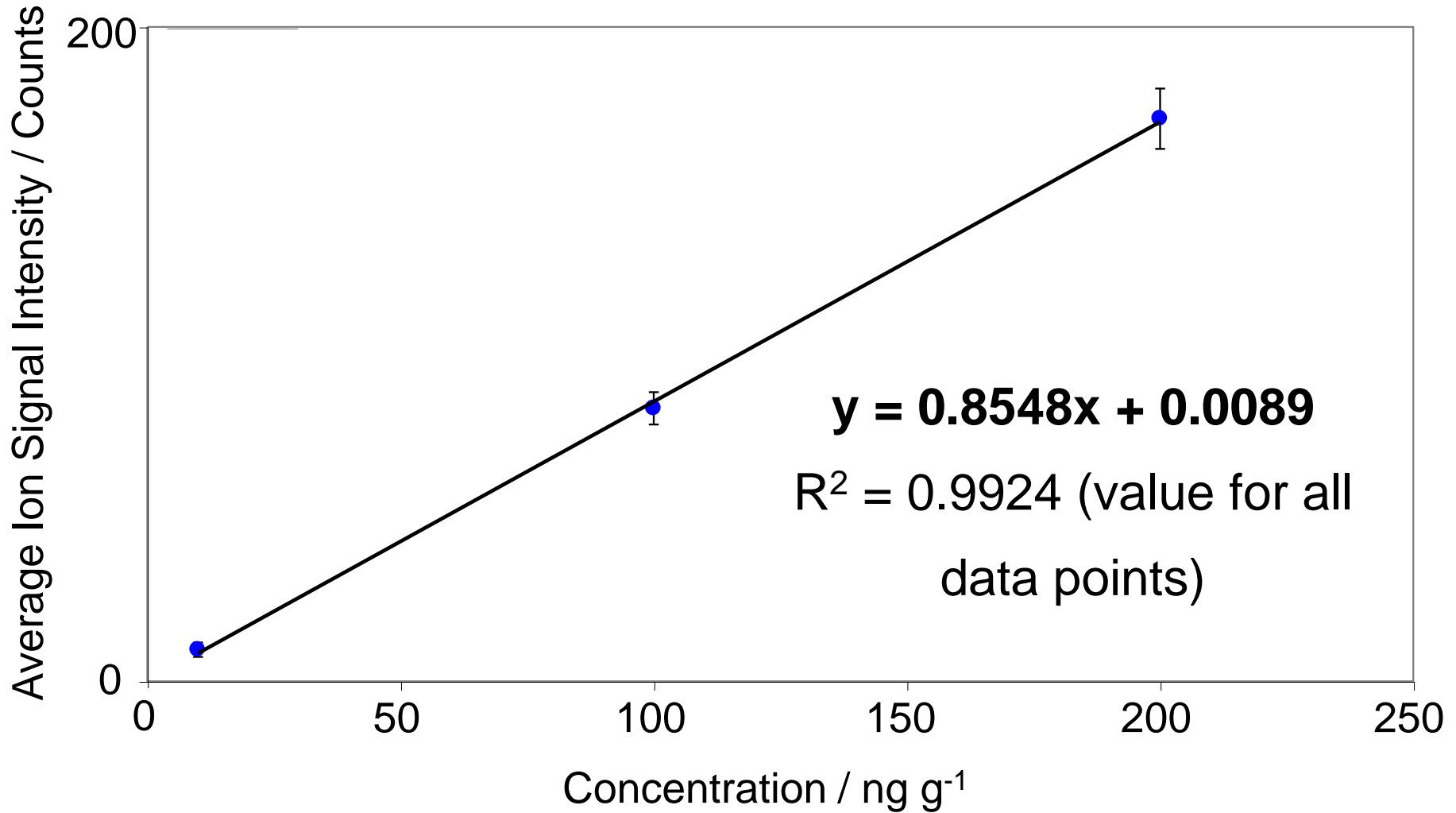
# Ion-time Response For Gd Standards

(10, 100 and 200 ng g<sup>-1</sup>)



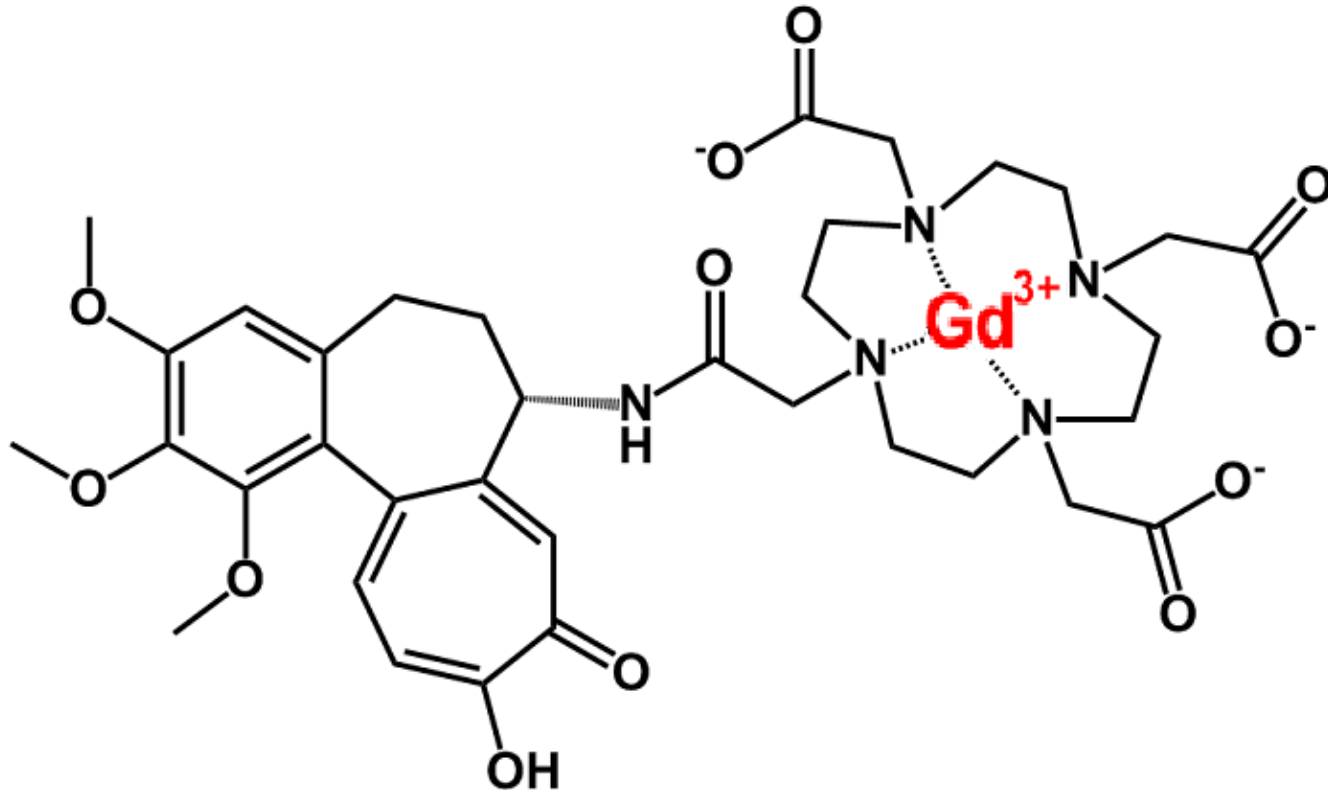
Average peak heights plotted against elemental concentration...

# Quantitation – Gd Calibration Graph



# Gd-tagged VDA - Tumour Therapy

- Novel therapeutic molecule with MRI contrast

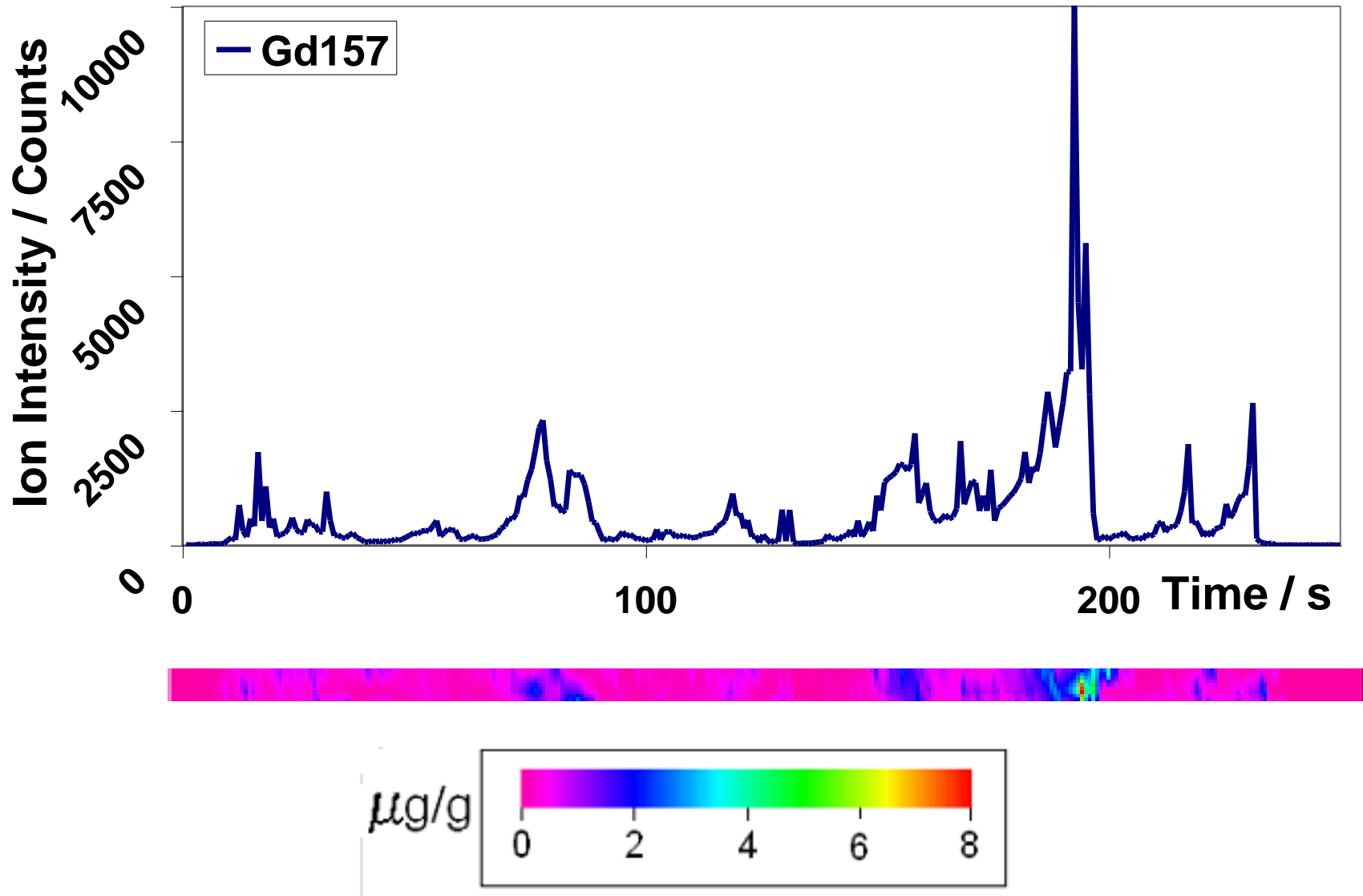


- Experiments performed in collaboration with MRI research group (ICL)

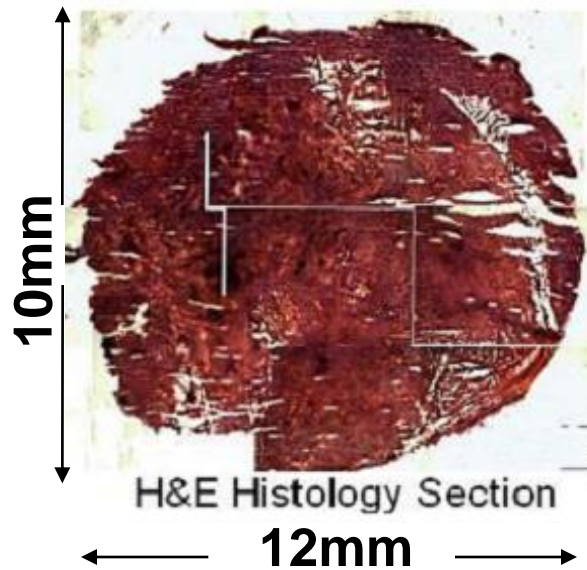
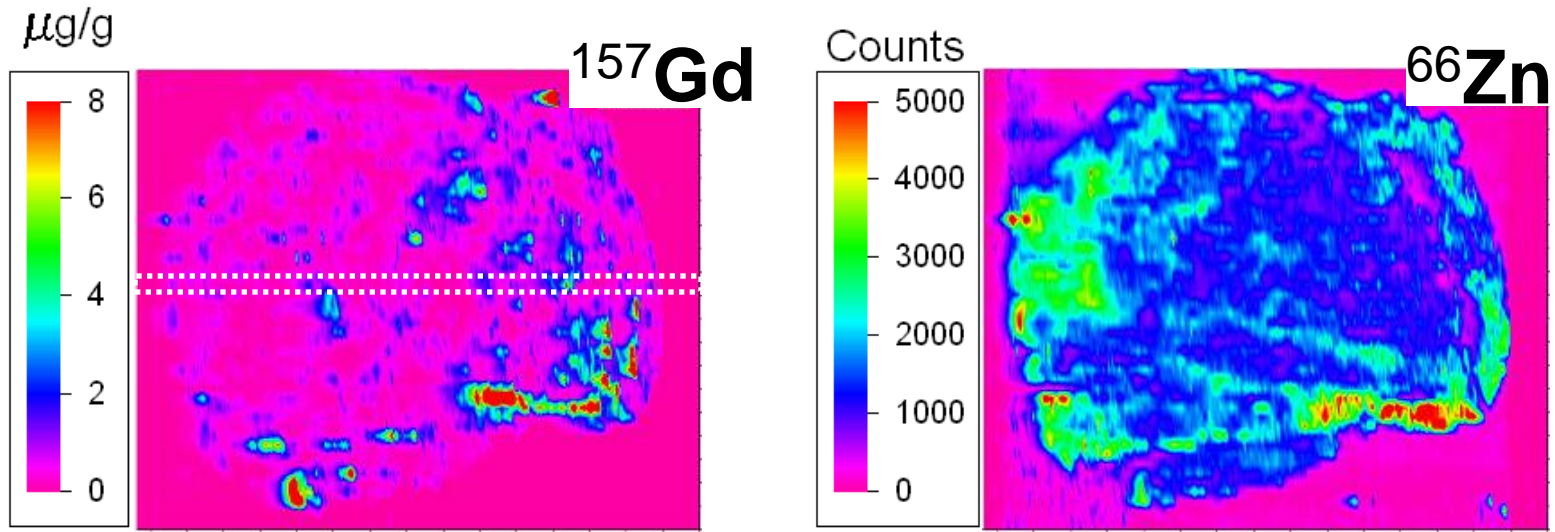
# Husbandry and Dosing Regimen

- $5 \times 10^6 / 0.1$  ml OVCAR-3 cells inoculated into the flank of 6-8 weeks old Balb/c nude mice
- When tumors reached  $\sim 7-10$  mm<sup>2</sup>, mice tail vein cannulated for the administration of either:
  - **200 mg/kg (22.64 mmol) Gd.DOTA.Colchicinic acid**
  - **200  $\mu$ l saline (control)**
- MRI scanning at 2, 8 and 24 hours post injection.
- After the final scan mice were sacrificed, the tumor excised and snap-frozen

# Gd Tumour Therapy Raw Raster Data



# Quantitative distribution Gd Contrast Agent

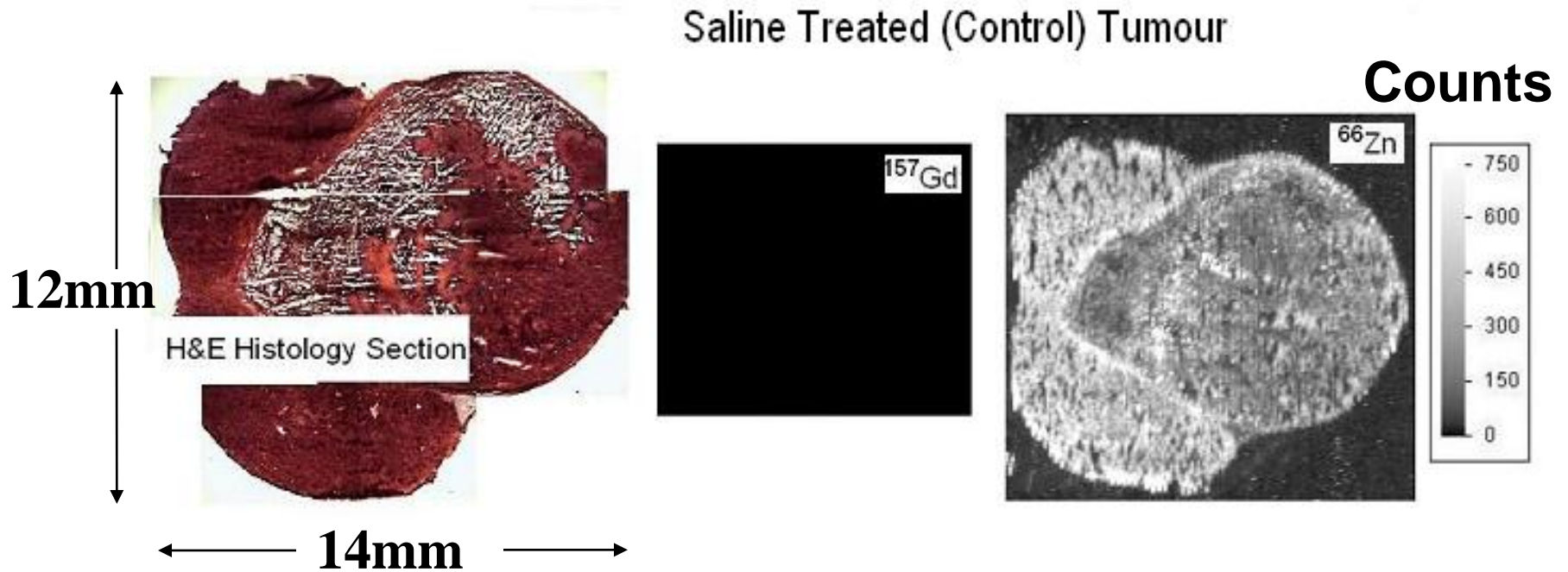




# LA-ICP-MS Parameters For Analysis of Tumour and Standards

<b>Laser Ablation Unit: New Wave MACRO (266nm) Nd:YAG</b>	
<b>Beam diameter</b>	<b>100µm</b>
<b>Raster spacing</b>	<b>200µm</b>
<b>Scan rate</b>	<b>50µm/s</b>
<b>Energy,</b>	<b>0.7mJ</b>
<b>Freq</b>	<b>10Hz</b>
<b>Runtime</b>	<b>~ 4.5 hours</b>

# Control Sample



**Cetac LSX-200 (266nm) Nd:YAG**

**Beam diameter 50 $\mu\text{m}$ , Raster spacing 100 $\mu\text{m}$**

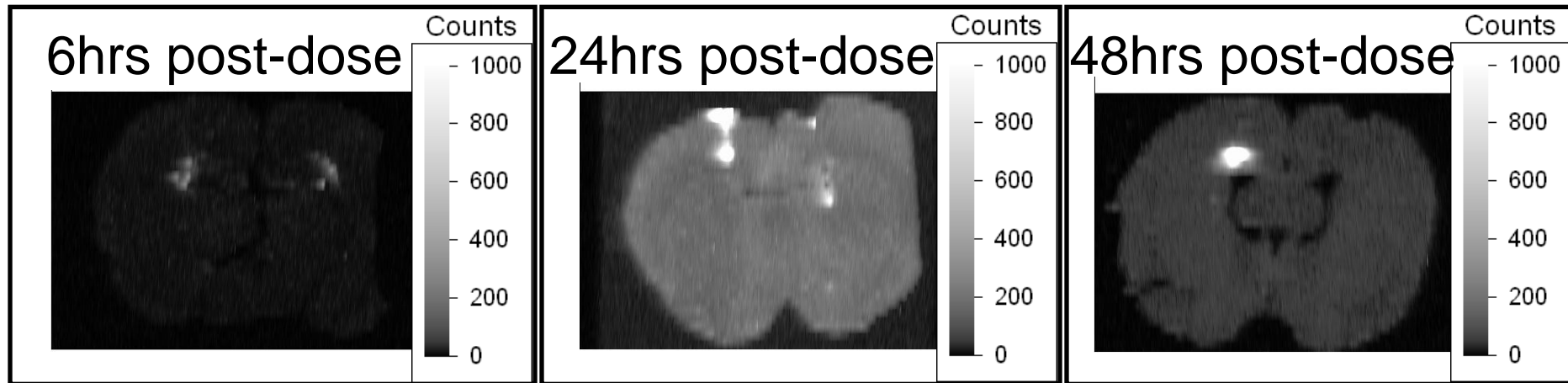
**Scan rate 50 $\mu\text{m}/\text{s}$ ,**

**Energy 0.6mJ, Freq 10Hz,**

# Comparative imaging – MRI and LA-ICP-MS

Study involving Gd-tagged nanoparticle delivery in mouse brain (developing treatment for dementia), collaborating with UCL

## LA-ICP-MS



## MRI

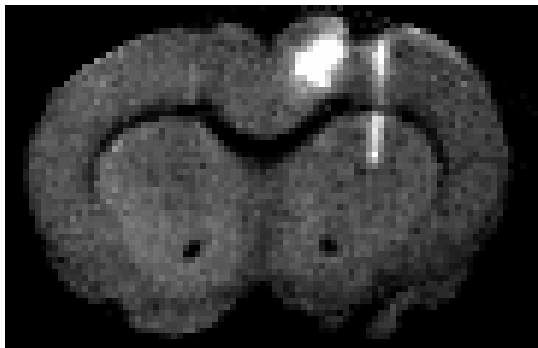


# Benefits of combining LA-ICP-MS with MRI

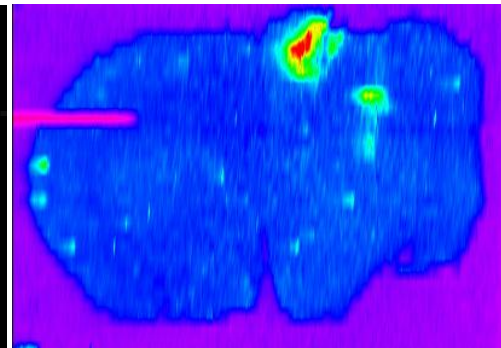
- Signal quantitation
- Affords superior spatial resolution ( $\sim 5\mu\text{m}$ ) and sensitivity
- Also affords detection of multiple elements

Particularly useful in determining sources of MRI contrast  
e.g. MRI image below demonstrates a 'false positive' result in a control sample

MRI TR=200



Fe<sup>57</sup>



Gd<sup>157</sup>



# Conclusions

- Studies show ultimate fate of Gd in selected tissues
- Signal quantitation achieved
- Benchmarking to MRI implemented
- Unique benefits of LA-ICP-MS utilised to complement MRI studies

## Further Work

- Ongoing studies will involve 2<sup>nd</sup> generation Gd nanoparticle formulations (UCL collaboration)
- Advance pharmacokinetic-like studies, determining elemental (Gd, Pt) quantitation and distribution over serialized time points

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