Research Group
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Quantifying the Impact of Formalin-Fixation on R_2^* Orientation Dependency in Human Brain White Matter

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Background and Objective

- R_2^* in brain white matter depends on fiber orientation relative to the magnetic field B_0 [1-4]
- Many studies use formalin-fixed tissue → **fixation alters tissue properties** [5-7]
- It remains unclear how fixation affects orientation dependency of R_2^*

- **Objective:** Determine how formalin fixation influences R_2^* orientation dependency

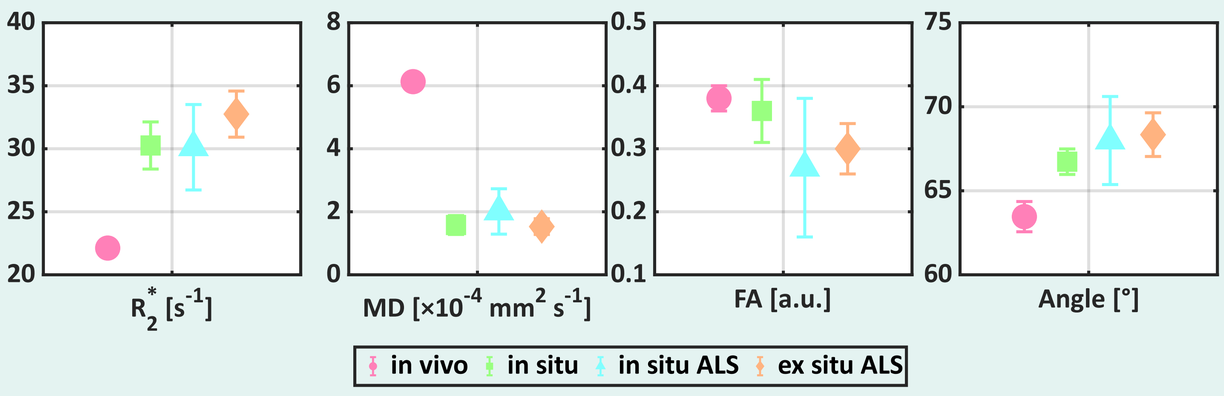
Methods

- Scans: 4 healthy in-vivo, healthy in-situ, ALS in-situ and ex-situ

- Sequences included: MP2RAGE or TSE (tissue segmentation), DTI (calculation of nerve fiber angle) and Multi-Echo GRE (R_2^* calculation)
- Fiber angles binned into 18 intervals à 5° (0° - 90°)
- R_2^* calculated voxel-wise, data normalised and registered

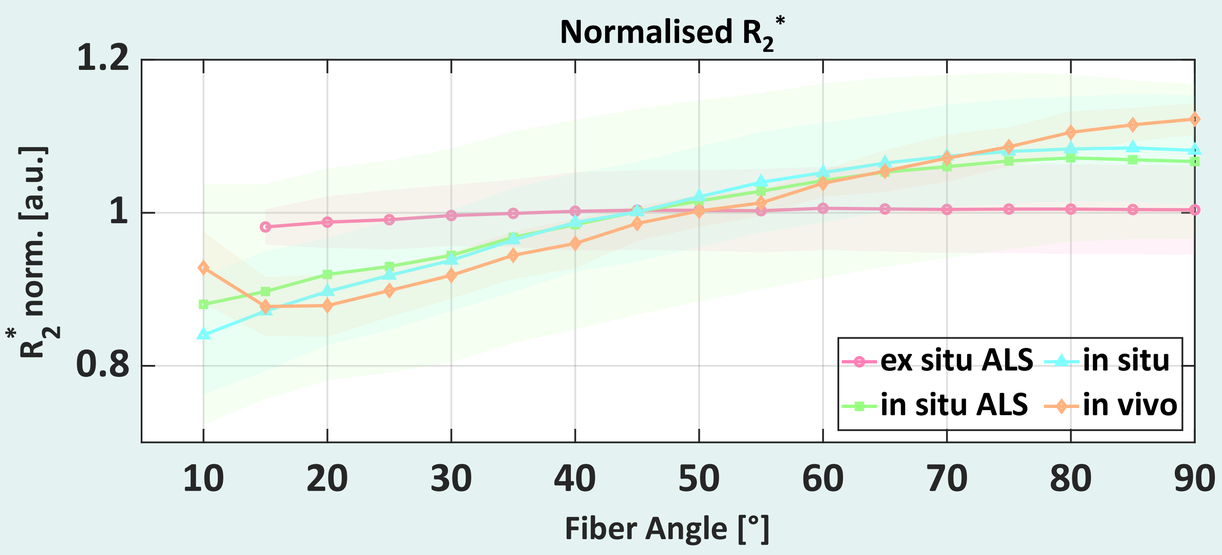
Results and Discussion

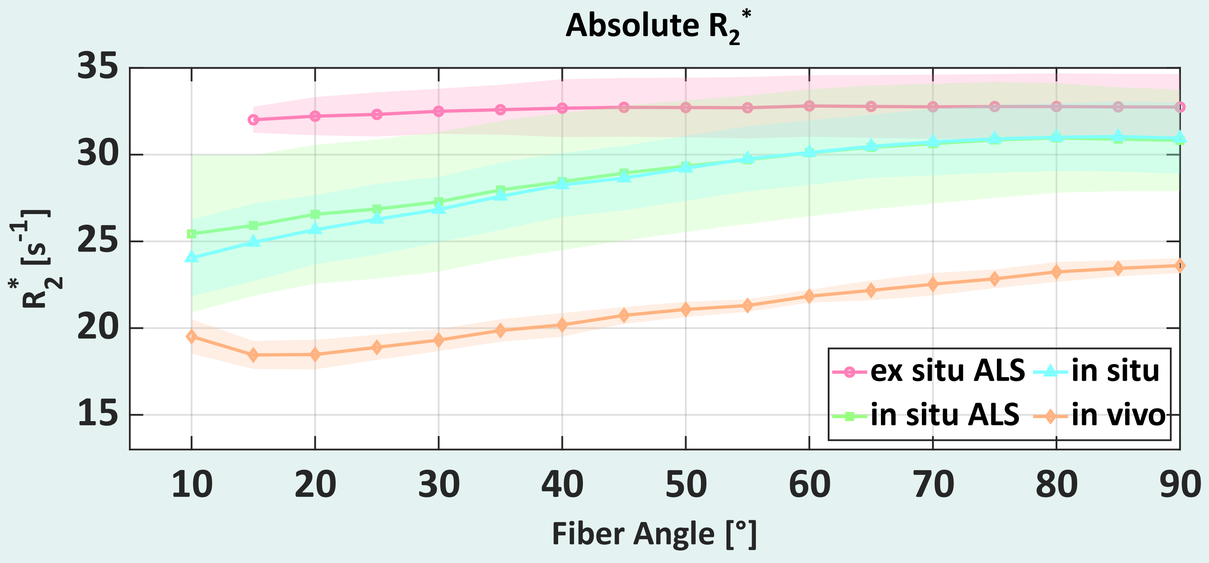
- Orientation dependency seen in all groups
- Strongest effect in-vivo (> ± 10 %), weaker in-situ, weakest ex-situ
- Absolute: R_2^* highest ex-situ, lowest in-vivo



- Formalin-fixation attenuates R_2^* orientation dependency
- Likely due to tissue microstructure changes (cross-linking, dehydration)
- Effect not fully explained by temperature or ALS-related changes

	in vivo	in situ	in situ ALS	ex situ ALS
R_2^* [s^{-1}]	22.10 ± 0.5	30.30 ± 1.9	30.10 ± 3.4	32.80 ± 1.8
MD [$x10^{-4}$ mm^2 s^{-1}]	6.10 ± 0.2	1.60 ± 0.3	1.70 ± 0.6	1.50 ± 0.3
FA [a.u.]	0.38 ± 0.02	0.36 ± 0.05	0.37 ± 0.03	0.30 ± 0.04
Angle [°]	63.50 ± 0.9	66.70 ± 0.8	66.00 ± 1.3	68.30 ± 1.3





Conclusion

✓ Formalin fixation attenuates R_2^* orientation dependency, likely due to protein cross-linking
✓ Fixed-tissue R_2^* values have to be interpreted carefully in translational and comparative studies

[1]: Bender et al.; *NMR Biomed*; 2020
[2]: Lenz et al.; *Magn Reson Med.*; 2021
[3]: Oh et al.; *NeuroImage*; 2013
[4]: Wharton et al.; *NeuroImage*; 2013
[5]: Leprince et al.; *ISMRM*; 2015
[6]: Pfefferbaum et al.; *NeuroImage*; 2004
[7]: Neuhaus et al.; *MRM*; 2024