

Exploring the Biomechanical Properties of Brain Malignancies and their Pathological Determinants with Magnetic Resonance Elastography

Jin Li¹, Yann Jamin¹, Jessica K.R. Boult¹, Philippe Garteiser², Jose L. Ulloa³, Sergey Popov^{4,5}, Craig Cummings¹, Gary Box⁵, Suzanne A. Eccles⁵, Chris Jones^{4,5}, John C. Waterton³, Jeffrey C. Bamber¹, Ralph Sinkus^{2,6}, and Simon P. Robinson¹

¹Division of Radiotherapy & Imaging, The Institute of Cancer Research, Sutton, Surrey, United Kingdom, ²INSERM U773, CRB3, Centre de Recherches Biomédicales Bichat-Beaumon, France, ³Personalised Healthcare and Biomarkers, AstraZeneca, Macclesfield, Cheshire, United Kingdom, ⁴Division of Molecular Pathology, The Institute of Cancer Research, Sutton, Surrey, United Kingdom, ⁵Division of Cancer Therapeutics, The Institute of Cancer Research, Sutton, Surrey, United Kingdom, ⁶BHF Centre of Excellence, Division of Imaging Sciences and Biomedical Engineering, King's College London, King's Health Partners, St Thomas' Hospital, London, United Kingdom

Introduction

Cancer tissues are characterised by a loss of tensional homeostasis, often leading to marked differential stiffness between tumours and their host tissues. Magnetic resonance elastography (MRE) affords non-invasive and quantitative measurement of tissue viscoelastic properties *in vivo*, including the brain. Recently MRE revealed that tumours derived from intracranial injection of human triple negative breast adenocarcinoma MDA-MB-231, rat N-ethyl-N-nitrosourea-induced glioma RG2 or human adult glioblastoma U87-MG cells were softer (lower value for the real part, G_d , of the shear modulus) than healthy mouse brain tissue (Fig. 1) [1]. MDA-MB-231 tumours, presenting an infiltrative phenotype [2], were significantly softer and less viscous than the relatively well-circumscribed RG2 and U87-MG gliomas. In this study we explore the pathological determinants of the tumour viscoelastic properties in these models.

Materials and Methods

Studies were performed in compliance with licences issued under the Animals (Scientific Procedures) Act 1986 following local ethical committee review.

MRE was performed as previously described [1]. Following MRE, tumour-bearing brains were excised, formalin fixed, and paraffin-embedded. Using adjacent sections (5 μ m), we assessed:

- cellular density, by counting the number of nuclei on haematoxylin and eosin (H&E) stained sections.*
- microvessel density, by manual counting the number of vessels stained for the murine vascular endothelial marker CD31.*
(* $\times 200$ magnification, four 50 \times 50 μ m² ROIs per fields, >3 fields per section)
- myelin content using Luxol Fast Blue staining with nuclei counterstained with Gill's haematoxylin.‡
- collagen content using picrosirius red staining.‡
- (‡ reviewed and manually scored by a pathologist)

Results

MDA-MB-231 tumours presented regions of sparse cell density also characterised by the presence of oedema, which contrasted with the homogenous dense texture of RG2 and U87 tumours (Fig. 1). Quantitative analysis confirmed significantly lower cellular density in MDA-MB-231 tumours compared with RG2 or U87 tumours (Fig. 2). No areas of necrosis were detected in any of the tumour models. In the tumours, myelin, a main determinant of the elevated stiffness of corpus callosum, was only detected as sparse and very fine fibres within U87-MG tumours, indicating that no highly myelinated, and hence stiff structures, of the brain have been entrapped during tumour invasion. Collagen deposition, another determinant of tissue stiffness, was only observed along blood vessels in all tumour types, a feature that these tumours share with the normal brain, indicating a potential co-option of the brain extracellular matrix by the tumour cells. MDA-MB-231 tumours were also characterised by the presence of large distended vessels and a significantly lower microvessel density (Fig. 2). Positive correlations were found between the tumour cellular and microvessel density with MRE-derived elasticity G_d and viscosity G_l (Fig. 3).

Conclusion

In contrast to normal brain, the lack of anisotropic structure of intracranial tumours may underpin their relative softness, a feature that has been reported in human brain tumours using MRE [3]. Our data also suggest that both tissue cellularity and microvessel density are determinants of the viscoelastic properties of brain malignancies.

References:

[1] Jamin *et al.*, Proc. ISMRM 0448 (2013), [2] Boult *et al.*, Proc. ISMRM 1524 (2012), [3] Guo *et al.*, Proc. ISMRM 0973 (2013).

Acknowledgements:

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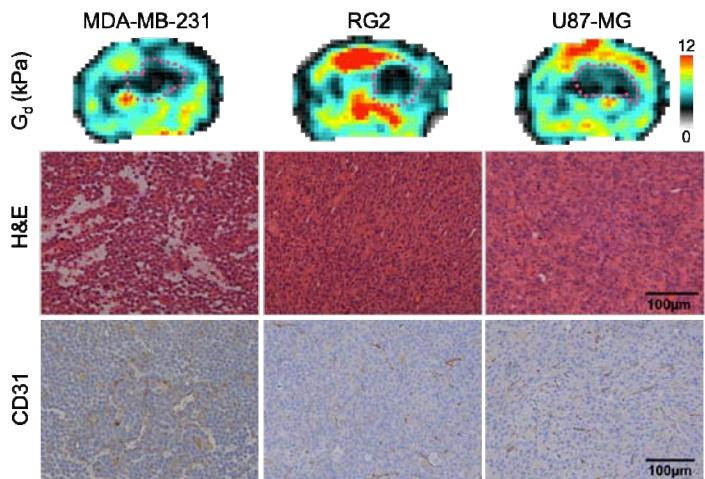


Figure 1. Representative calculated maps of G_d , H&E stained sections showing the cell density, and CD31 immunohistochemistry indicating microvessels, in orthotopically-implanted MDA-MB-231, RG2 and U87-MG brain tumours. Tumour burden was indicated (●) in maps of G_d .

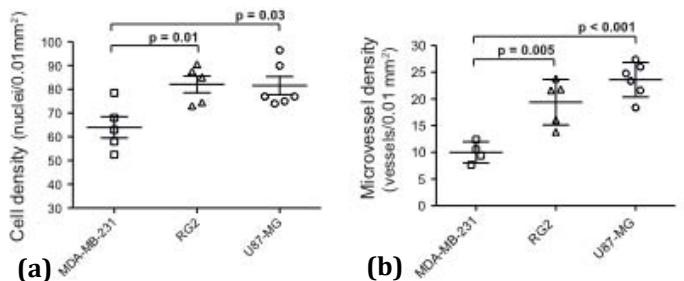


Figure 2. Quantitative assessment of (a) cell and (b) microvessel density in intracranially-implanted MDA-MB-231, RG2 and U87-MG tumours. p-values given for an unpaired Student's t-test with a 5% level of significance.

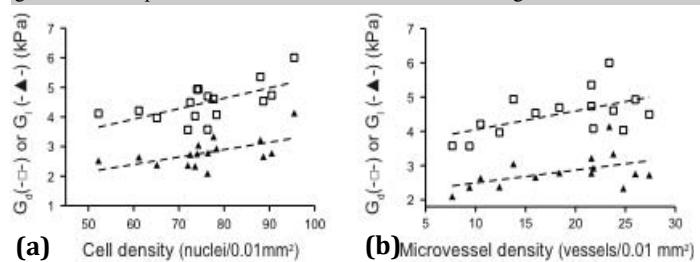


Figure 3. Correlation of elasticity G_d and viscosity G_l with (a) cell density ($r=0.61$, $p=0.01$ and $r=0.57$, $p=0.02$ respectively), and (b) microvessel density ($r=0.54$, $p=0.03$ and $r=0.48$, $p=0.07$ respectively) in intracranially-implanted tumours.