

# Histological growth patterns and genotype in oligodendroglial tumours: correlation with MRI features

Michael D. Jenkinson,<sup>1,3</sup> Daniel G. du Plessis,<sup>3,4</sup> Trevor S. Smith,<sup>2</sup> Kathy A. Joyce,<sup>5</sup> Peter C. Warnke<sup>1,3</sup> and Carol Walker<sup>5</sup>

Departments of <sup>1</sup>Neurosurgery and <sup>2</sup>Neuroradiology, The Walton Centre for Neurology and Neurosurgery, Division of Neuroscience, <sup>3</sup>University of Liverpool, Liverpool, <sup>4</sup>Department of Neuropathology, Hope Hospital, Salford and <sup>5</sup>Clatterbridge Cancer Research Trust, JK Douglas Laboratories, Clatterbridge Hospital, Bebington, Wirral, UK

Correspondence to: Mr Michael D. Jenkinson, Division of Neuroscience, Clinical Sciences Centre, Lower Lane, Liverpool, L9 7LJ, UK  
E-mail: michael.jenkinson@liv.ac.uk

**Oligodendroglial neoplasms with the  $-1p/-19q$  genotype are more indolent with longer survival and increased therapeutic responsiveness than those with intact  $1p/19q$ , but the biological basis for these clinical differences is unclear. Recent research suggests that oligodendrogliomas with and without the  $-1p/-19q$  genotype may be distinguished by their magnetic resonance imaging (MRI) appearance, suggesting possible differences in growth characteristics. This study examined the relationship between genotype and histological growth patterns of oligodendroglial neoplasms in association with MR imaging characteristics. Tumour imaging features assessed on MRI included sharp-versus-indistinct border, smooth-versus-irregular contour, homogeneous-versus-heterogeneous signal, contrast enhancement and paramagnetic susceptibility effect. Growth patterns (solid : mixed : infiltrative), tumour-margin transitions in cellularity and calcification were determined histopathologically. Allelic imbalance in chromosomes  $1p36$  and  $19q13$  was determined. Thirty-three oligodendrogliomas (25 with  $1p/19q$  loss) and 53 oligoastrocytomas (18 with  $1p/19q$  loss) were investigated. Solid, mixed or infiltrative growth patterns were seen in grade II and grade III tumours with or without  $1p/19q$  loss, but infiltrative growth was more common in tumours with intact  $1p/19q$  ( $\chi^2: P = 0.029$ ). Grade III tumours were more likely to have a solid growth pattern ( $\chi^2: P = 0.046$ ) associated with contrast enhancement ( $\chi^2: P = 0.011$ ). Transition in cellularity at the radiological margin did not differ according to genotype. All cases with  $T_1$  or  $T_2$  signal homogeneity had intact  $1p/19q$ . Tumours with sharp/smooth borders were more likely to have intact  $1p/19q$  than those with indistinct/irregular borders ( $\chi^2: P < 0.001$ ), but this was not related to histological growth characteristics. This study identified a group of oligodendroglial tumours with intact  $1p/19q$  displaying distinctive MR imaging features that were unrelated to the histopathology characteristics.**

**Keywords:** brain tumour; MRI; molecular genetics; histopathology

Received June 1, 2005. Revised September 19, 2005. Accepted March 30, 2006

## Introduction

Oligodendroglial neoplasms account for up to 33% of all adult gliomas (Perry, 2001; Engelhard *et al.*, 2002) and have a more indolent clinical history and prolonged survival (Engelhard *et al.*, 2002, 2003) compared with astrocytic tumours of the same grade. The majority of oligodendrogliomas and some oligoastrocytomas respond to chemotherapy or radiotherapy (Cairncross and Macdonald, 1988; Engelhard *et al.*, 2003), and research suggests that molecular genetic analyses may be the most appropriate way to identify

the chemosensitive subgroup (Cairncross *et al.*, 1998; Ino *et al.*, 2001; van den Bent, 2004). Combined loss of heterozygosity (LOH) on chromosome arms  $1p$  and  $19q$  is an early genetic event in the histogenesis of oligodendroglial neoplasms and is considered to be their genetic hallmark, being present in up to 90% of oligodendrogliomas and 50% of oligoastrocytomas (Reifenberger *et al.*, 1994; Kraus *et al.*, 1995; Bigner *et al.*, 1999). The  $-1p/-19q$  genotype is associated with longer time to first therapy, therapeutic

response to initial therapy, longer progression-free survival, response at recurrence and prolonged overall survival (Bauman *et al.*, 2000; Smith *et al.*, 2000; Ino *et al.*, 2001; Van Den Bent *et al.*, 2003; Fallon *et al.*, 2004; Felsberg *et al.*, 2004; Walker *et al.*, 2005). However, the biological basis for these clinical differences is not well understood. While much of the research effort has been addressed at a molecular level (Kitange *et al.*, 2003; Jeuken *et al.*, 2004), less is known of the comparative pathological and *in vivo* biology of these tumours in relation to genotype (Megyesi *et al.*, 2004; Walker *et al.*, 2004).

Whether oligodendroglial neoplasms with or without the  $-1p/-19q$  genotype differ in their 'invasiveness' or ability to diffusely infiltrate normal brain parenchyma is not known, and the literature evidence is limited, circumstantial and conflicting. Microarray analysis has demonstrated that oligodendrogliomas with loss of 1p and 19q have a similar expression profile to normal brain (Mukasa *et al.*, 2004), which has been hypothesized to occur if normal brain tissue is trapped within an invasive tumour (Megyesi *et al.*, 2004), although this was not assessed histologically. In contrast, using *ex vivo* rodent organotypic brain slice assays and implanted biopsy samples of gliomas, oligodendroglial tumours with 1p/19q loss were less invasive and grew as compact, hypercellular proliferating masses compared with astrocytic tumours of a similar grade (Palfi *et al.*, 2004). Clinically, untreated newly diagnosed and recurrent oligodendroglial and astrocytic gliomas can show extensive infiltration of brain parenchyma (Kelly *et al.*, 1987; Daumas-Duport *et al.*, 1997b), while multifocal tumours are occasionally found in both lineages (Batzdorf and Malamud, 1963; Barnard and Geddes, 1987). In gliomas, three types of growth pattern have been defined: type I (solid), type II (mixed) and type III (infiltrative) (Kelly *et al.*, 1987; Daumas-Duport *et al.*, 1997a, b), but these have not been studied in oligodendroglial neoplasms classified by genotype. A recent study of oligodendrogliomas has reported associations between genotype and MR imaging; an indistinct tumour border, heterogeneous signal intensity and paramagnetic susceptibility are associated with 1p/19q LOH, which suggests that MRI characteristics may be related to tumour invasiveness (Megyesi *et al.*, 2004).

The aim of this study was to investigate the histological growth characteristics of oligodendroglial neoplasms in relation to the 1p/19q genotype and to relate these findings to MR imaging characteristics.

## Material and methods

### Case selection

Cases for study were selected from a cohort of primary (previously untreated) and recurrent adult gliomas at the Walton Centre for Neurology and Neurosurgery between July 1999 and July 2005. Eligibility criteria included (i) histological diagnosis of oligodendroglioma or oligoastrocytoma based on the current WHO classification (Kleihues, 2000); (ii) pre-therapy MRI scan; (iii) known 1p36 and 19q13 status. The local research ethics committee

approved the study, and consent was obtained from all patients. Two otherwise eligible cases with post-resection, pre-therapy MRI but insufficient residual tumour were excluded from the study.

### Tumour tissue acquisition

Tissue for pathology diagnosis was obtained by serial stereotactic biopsy, image-guided biopsy or resection. Thirteen patients underwent frameless image-guided biopsy or resection using a neuro-navigation system (SNN, Toronto, Canada), and samples were taken for intra-operative smear analysis and routine histopathology. Seventy-three patients underwent frame-based, CT or T<sub>1</sub>-weighted MRI-guided serial stereotactic biopsy. Using the stereotactic planning station (Leibinger, Germany) the tumour volume was calculated with the target position (TP) in the centre of the tumour as determined by volumetry. The biopsy trajectory was calculated to encompass the most aggressive part of the tumour as defined by contrast enhancement or, in non-enhancing cases, to maximize representation of the tumour. Biopsy samples commenced 2–3 mm outside the radiological tumour margin and were taken at 1-mm intervals up to and in some cases beyond the target position. Brain shift and CSF loss were minimized by the use of a 6-mm stereotactic burr hole and a 1-mm internal diameter biopsy cannula. Alternating samples were taken for intra-operative smear diagnosis, formalin fixation and in some cases for snap freezing.

### Histopathology

The histopathology diagnosis for each case was made according to current WHO guidelines (Kleihues, 2000) based on examination of all routinely processed tissue taken throughout the biopsy trajectory, or from open surgery. Pathology slides were re-reviewed by a neuropathologist (D.duP.) to assess growth pattern [solid, mixed or infiltrative—respectively corresponding to structure type I, II and III tumours (Daumas-Duport *et al.*, 1997a, b)] and presence of calcification. In the 73 cases diagnosed with frame-based CT or MRI-guided serial stereotactic biopsy, each sample was recorded relative to its position along the biopsy trajectory, enabling assessment of changes in cellularity at the radiological tumour margin within a 5-mm interval. This was scored as 'marked', that is, a change in cellularity from normal or low cellularity to high cell density, or 'insidious', that is, little or no change in cellularity. The median number of biopsies taken for each case for routine histology was 19 (range: 4–32). Comparison of MRI appearance and histology at the tumour margin was made in the stereotactic group only.

### Molecular genetics

For each case, regions of tumour histology in biopsy specimens representative of the overall pathology diagnosis were selected for study. Laser capture microdissection was used to enrich the tumour component in the samples for analysis, and determination of allelic imbalance was carried out using paired normal and tumour samples and polymerase chain reaction (PCR) amplification of microsatellite markers at 1p36 (D1S2667, D1S508, D1S214) and 19q13 (D19S412, D19S112, D19S596), in a two-round PCR procedure followed by capillary electrophoresis as described previously (Walker *et al.*, 2001, 2003, 2004).

### Image analysis

MRI scans were assessed by a neuroradiologist (T.S.S.), neuropathologist (D.duP.) and neurosurgeon (M.D.J.) blinded to the

genotype of each case. T<sub>1</sub>-weighted, T<sub>1</sub> post-gadolinium and T<sub>2</sub>-weighted MR axial images were consistently available for analysis. According to previously described methods (Megyesi *et al.*, 2004) the following magnetic resonance imaging features were evaluated: (i) sharp-versus-indistinct tumour border on T<sub>1</sub> and T<sub>2</sub>; (ii) homogeneous-versus-heterogeneous signal intensity throughout the tumour on T<sub>1</sub> and T<sub>2</sub>; (iii) contrast enhancement, present versus absent; and (iv) paramagnetic susceptibility effect, present versus absent. Additionally, the tumour contour on T<sub>2</sub> was also evaluated as smooth versus irregular. Images were scored by consensus.

**Statistical analysis**

Data were analysed using SPSS for windows (SPSS, UK). Two-tailed Fisher’s exact and Pearson’s  $\chi^2$  test were used to determine the significance of associations. Probability (*P*) values < 0.05 were considered significant.

**Results**

Eighty-six cases were eligible for the study, and the clinico-pathological findings are shown in Table 1 (additional data

in Supplementary Table 1). Median age at operation was 44 years with 43 males and 43 females in the study. Of the 64 primary, previously untreated cases, 5 had an earlier pathology diagnosis and 59 were newly diagnosed at study. Twenty-two recurrent tumours had received radiotherapy before MRI (median: 69 months; range: 6–165). Complementary T<sub>1</sub> and T<sub>2</sub> images were not available in 12 cases. 1p/19q loss was present in 15/22 OII, 10/11 OIII, 11/31 OAI and 7/22 OAIII. One patient had loss of 1p without loss of 19q.

**Genotype and histopathology**

Solid, mixed or infiltrative growth patterns were seen in tumours with or without 1p/19q loss and in grade II or grade III tumours. Tumours with infiltrative growth were more likely to have intact 1p/19q, and those with mixed or solid growth patterns were more likely to have 1p/19q loss (Table 2). However, these associations were not significant if only primary tumours were analysed (data not shown:  $\chi^2$ : *P* = 0.113). Grade III tumours were more likely to have a solid

**Table 1** Radiological, histopathological and genetic characteristics of the study population (details for each case are in Supplementary Table 1)

	OII	OIII	OAI	OAIII	All cases
Number of cases	22	11	31	22	86
Primary : recurrent	16 : 6	7 : 4	28 : 3	13 : 9	64 : 22
Loss of 1p36 and 19q13	15/22	10/11	11/31	7/22	43/86
Sharp T <sub>1</sub> border	3/21	0/9	7/29	6/21	16/80
Sharp T <sub>2</sub> border	3/21	1/11	8/30	6/22	18/84
Smooth T <sub>2</sub> contour	2/21	1/11	7/30	6/22	16/84
Homogeneous on T <sub>1</sub>	0/21	0/9	5/28	2/21	7/79
Homogeneous on T <sub>2</sub>	1/21	0/11	5/30	2/22	8/84
Contrast enhancement	12/20	10/10	14/29	21/21	56/80
Paramagnetic susceptibility effect	8/15	5/7	4/12	4/14	21/48
Solid : mixed : infiltrative	2 : 11 : 9	2 : 6 : 3	2 : 13 : 16	6 : 10 : 6	12 : 40 : 34
Calcification	10/22	6/11	6/31	5/22	27/86
Marked transition in cellularity	0/19	2/8	2/26	4/20	8/73

Denominator less than the number of cases in each group indicates incomplete or non-assessable data.

**Table 2** Correlation between growth patterns, histopathology, calcification, transition in cellularity and genotype for the series

	1p/19q loss		<i>P</i> -value	WHO grade		<i>P</i> -value	Subtype		<i>P</i> -value
	No	Yes		II	III		O	OA	
Growth pattern									
Solid	5	7	0.029 <sup>+</sup>	4	8	0.046 <sup>+</sup>	4	8	0.758 <sup>+</sup>
Mixed	15	25		24	16		17	23	
Infiltrative	23	11		25	9		12	22	
Transition in cellularity									
Insidious	33	32	0.712*	43	22	0.048*	25	40	0.457 <sup>+</sup>
Marked	5	3		2	6		2	6	
Calcification									
No	34	25	0.037 <sup>+</sup>	37	22	0.760 <sup>+</sup>	17	42	0.007 <sup>+</sup>
Yes	9	18		16	11		16	11	

<sup>+</sup>Pearson  $\chi^2$  test; \*Fisher’s exact test.

growth pattern, and grade II tumours, mixed or infiltrative growth patterns in the series (Table 2) and in primary tumours (data not shown:  $\chi^2$ :  $P = 0.009$ ). There was no association between growth pattern and histopathology subtype. When changes in cellularity at the radiological margin were assessed in the 73 cases diagnosed by CT/MRI-guided serial stereotactic biopsy, no differences were observed in

**Table 3** MRI characteristics and growth pattern

	Growth pattern			P-value
	Solid	Mixed	Infiltrative	
Contrast enhancement				
No	1	7	15	0.011 <sup>+</sup>
Yes	11	29	17	
Combined features				
Sharp/smooth	1	5	8	0.391 <sup>+</sup>
Indistinct/irregular	10	27	23	

<sup>+</sup>Pearson's  $\chi^2$  test.

**Table 4** MRI characteristics and transition in cellularity

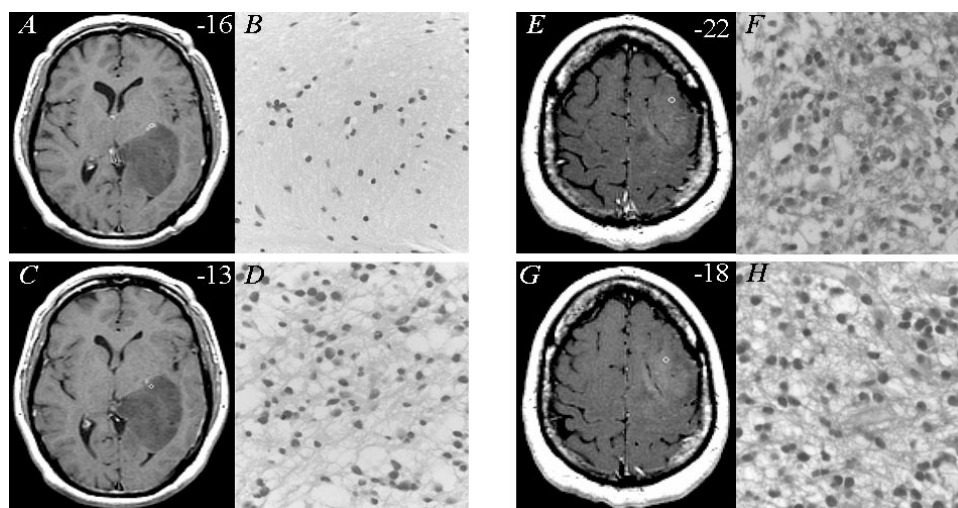
	Transition in cellularity		P-value
	Insidious	Marked	
T <sub>1</sub> border			
Sharp	10	3	0.127*
Indistinct	50	4	
Combined features			
Sharp/smooth	9	2	0.287*
Indistinct/irregular	47	4	

\*Fisher's exact test.

relation to genotype. Tumours in the series (Table 2) and primary tumours (data not shown) with and without the  $-1p/-19q$  genotype were as likely to show marked or insidious changes in cellularity. There was no correlation between transition in cellularity and histopathology subtypes; however, grade II tumours were weakly associated with insidious margins (Table 2). Calcification was significantly associated with genotype, being present in only 21% of tumours with intact 1p/19q, compared with 42% of  $-1p/-19q$  tumours. Calcification was also associated with tumour subtype (Table 2).

### Histopathology and MRI

Solid and mixed tumour growth patterns were associated with contrast enhancement, and infiltrative growth with no contrast enhancement in the series (Table 3) and primary tumours (data not shown:  $\chi^2$ :  $P = 0.011$ ). No other significant associations between imaging characteristics and growth pattern or transitions in cellularity were observed. Only 8 cases had a marked transition in cellularity; 3 out of 7 (43%) of these had a sharp T<sub>1</sub> tumour border, while 83% of cases with an insidious transition in cellularity had an indistinct T<sub>1</sub> tumour border (Table 4 and Fig. 1). There was no correlation between calcification and paramagnetic susceptibility effect (data not shown:  $\chi^2$ :  $P = 1.0$ ). Tumour border, signal intensity and paramagnetic susceptibility effect did not distinguish histopathological subtype or grade. However, contrast enhancement was present in all grade III tumours, and 53% of grade II tumours in the series ( $\chi^2$ :  $P < 0.001$ ) and 46% of primary grade II tumours ( $\chi^2$ :  $P < 0.001$ ).



**Fig. 1** (A–D) Serial-stereotactic biopsy trajectory for a grade II oligoastrocytoma. The circle identifies the position of the biopsy sample on the MRI and the number represents the distance (in millimetres) to the target position. (A) Sharp MRI tumour border; with (B) low cellularity at the edge of the tumour; (C) 3 mm inside the MRI tumour border; with (D) a marked transition to higher cellularity. (E–H) Serial-stereotactic biopsy trajectory for a grade II oligodendroglioma. (E) Indistinct MRI tumour border; with (F) moderate cellularity at the edge of the tumour; (G) 4 mm inside the MRI tumour border; with (H) continued moderate cellularity (haematoxylin- and eosin-stained sections; original magnifications  $\times 400$ ).

**Table 5** Association of MRI features and genotype

		1p and 19q loss		P-value
		No	Yes	
<b>T<sub>1</sub> border</b>				
All cases (n = 80)	Sharp	14	2	<0.001 <sup>+</sup>
	Indistinct	24	40	
Primary (n = 60)	Sharp	13	2	0.001 <sup>+</sup>
	Indistinct	17	28	
<b>T<sub>2</sub> border</b>				
All cases (n = 84)	Sharp	15	3	0.002 <sup>+</sup>
	Indistinct	28	38	
Primary (n = 62)	Sharp	15	3	0.004 <sup>+</sup>
	Indistinct	19	25	
<b>T<sub>2</sub> contour</b>				
All cases (n = 84)	Smooth	13	3	0.008 <sup>+</sup>
	Irregular	30	38	
Primary (n = 62)	Smooth	13	3	0.014 <sup>+</sup>
	Irregular	21	25	
<b>Combined features</b>				
All cases (n = 74)	Sharp/smooth	13	1	<0.001 <sup>+</sup>
	Indistinct/irregular	23	37	
Primary (n = 55)	Sharp/smooth	13	1	<0.001 <sup>+</sup>
	Indistinct/irregular	16	25	
<b>T<sub>1</sub> signal</b>				
All cases (n = 79)	Homogeneous	7	0	0.004*
	Heterogeneous	30	42	
Primary (n = 59)	Homogeneous	7	0	0.005*
	Heterogeneous	22	30	
<b>T<sub>2</sub> signal</b>				
All cases (n = 84)	Homogeneous	8	0	0.006*
	Heterogeneous	35	41	
Primary (n = 62)	Homogeneous	8	0	0.006*
	Heterogeneous	26	28	
<b>Contrast enhancing</b>				
All cases (n = 80)	No	15	8	0.061 <sup>+</sup>
	Yes	24	33	
Primary (n = 59)	No	15	8	0.078 <sup>+</sup>
	Yes	15	21	
<b>Paramagnetic susceptibility effect</b>				
All cases (n = 48)	Absent	11	16	0.883 <sup>+</sup>
	Present	9	12	
Primary (n = 35)	Absent	9	11	0.767 <sup>+</sup>
	Present	6	9	

<sup>+</sup>Pearson's  $\chi^2$  test; \*Fisher's exact test.

### MRI and genotype

Significant associations were seen when imaging characteristics were correlated with genotype; 14 out of 16 cases with a sharp T<sub>1</sub> border had intact 1p/19q (Table 5 and Fig. 2A–D). However, intact 1p/19q was also seen in 38% with an indistinct T<sub>1</sub> border, which suggests that individually the presence of a sharp border is not specific for intact alleles. Similar findings were also seen with the T<sub>2</sub> border and T<sub>2</sub> contour (Table 5). When an indistinct tumour border on both T<sub>1</sub> and T<sub>2</sub> was identified, it was always associated with an irregular T<sub>2</sub> contour (n = 60). Likewise, a sharp tumour border on both T<sub>1</sub> and T<sub>2</sub> was always associated with a smooth T<sub>2</sub> contour (n = 14) (Fisher's exact: P < 0.001). These combined

imaging features were significantly correlated with genotype in the series, tumours with sharp/smooth imaging being more likely to have intact 1p/19q (Table 5). However, 38% of tumours with an indistinct/irregular imaging appearance also had intact 1p/19q. There were no significant associations between genotype and contrast enhancement or paramagnetic susceptibility effect (Table 5). All tumours with homogeneous signal intensity on T<sub>1</sub> (n = 7) and T<sub>2</sub> (n = 8) imaging had intact 1p/19q.

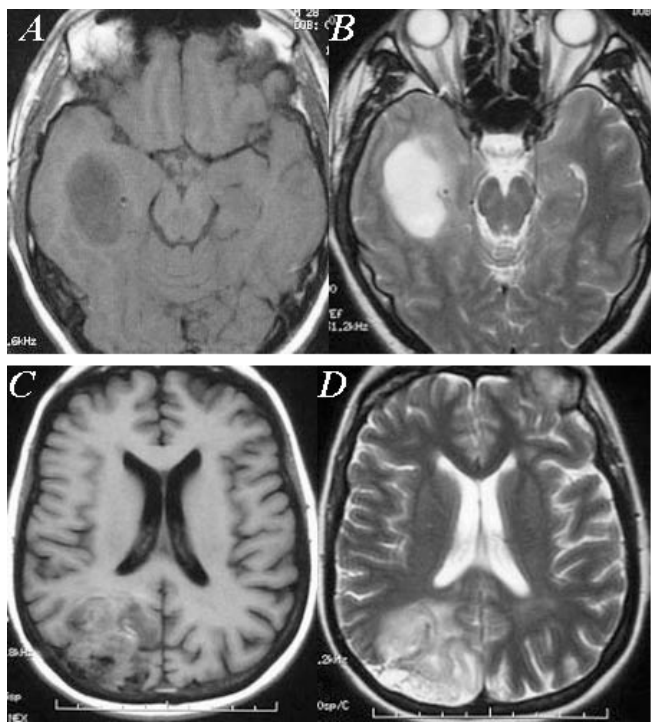
In the 14 cases with a sharp, smooth border, all were previously untreated; 12 were oligoastrocytomas; 13 had intact 1p/19q; 1 had solid, eight infiltrative and five mixed growth patterns; and 2 had marked transition in cellularity at the radiological margin. No unifying features were identified, and tumours with sharp borders were not influenced by their relationship to the grey/white matter interface or location (Table 6).

### Discussion

This is the first study correlating MRI features at the tumour border with actual histology from this imaging area in a cohort of oligodendroglial neoplasms classified according to genotype. The principal findings are that tumours with or without 1p/19q loss showed solid, mixed and infiltrative growth patterns, but infiltrative growth was more common in those with intact 1p/19q. Transition in cellularity at the margin was similar, irrespective of genotype; however, tumours with a sharp, smooth border and homogeneous signal intensity were more likely to have intact 1p/19q.

Gliomas have a propensity to be diffusely infiltrative, and it is now known that these migratory cells are refractory to conventional therapy, which may contribute to treatment failure (Giese *et al.*, 2003). Investigation of growth patterns is therefore fundamental to understanding the cellular interactions and tissue organization of these tumours. Diagnosis through frame-based serial stereotactic biopsy has proven diagnostic accuracy (Tilgner *et al.*, 2005) and was used in 85% of cases in this study. As in previous studies (Kelly *et al.*, 1987; Daumas-Duport *et al.*, 1997a, b), this permits the assessment of *in vivo* histopathological growth pattern and transition in cellularity in relation to co-registered clinical imaging characteristics. Oligodendroglial neoplasms irrespective of genotype displayed solid, mixed or infiltrative growth patterns, although in the series overall, infiltrative growth was associated with intact 1p/19q. Growth pattern correlated with WHO tumour grade, grade III tumours being more likely to exhibit solid tumour tissue and grade II tumours, infiltrative tissue. Consistent with previous observations, solid growth patterns were also associated with contrast enhancement, and infiltrative growth patterns with no contrast enhancement (Daumas-Duport *et al.*, 1997a). It has been suggested that the combined features of contrast enhancement and solid growth pattern reflect neovascularization enabling rapid growth of the developing tumour, with new blood vessels absent in regions of infiltration

(Daumas-Duport *et al.*, 1997*a, b*). Our data suggest that tumourigenesis in oligodendroglial neoplasms of both genetic lineages is similar with respect to both growth pattern and contrast enhancement, although there may be a tendency for tumours with intact 1p/19q to more commonly show infiltrative growth patterns.



**Fig. 2** MRI characteristics of oligodendroglial tumours. **(A and B)** Sharp, smooth T<sub>1</sub> and T<sub>2</sub> border with homogeneous signal intensity in a grade II oligoastrocytoma with intact 1p/19q. **(C and D)** Indistinct, irregular T<sub>1</sub> and T<sub>2</sub> border with heterogeneous signal intensity in a grade III oligodendroglioma with the -1p/-19q genotype.

In our study, 12 cases showed only a solid growth pattern, which contrasts with previously published series of oligodendrogliomas (Daumas-Duport *et al.*, 1997*b*) that may have included some oligoastrocytomas if current WHO guidelines were applied (Kleihues, 2000). The apparent disparity in growth pattern may be due to differences in the image guidance used for stereotactic biopsy in the two studies. We used the radiological tumour margin visible on CT or T<sub>1</sub>-weighted MRI to calculate the initial sampling point along the stereotactic biopsy trajectory, whereas previous studies used the T<sub>2</sub> margin (Daumas-Duport *et al.*, 1997*b*). Although there is a good correlation between CT and T<sub>1</sub>-weighted MRI for assessing tumour margins, the area of T<sub>2</sub> hyperintensity, which usually contains infiltrating tumour cells, often extends beyond this visible margin (Kelly *et al.*, 1987), and may not have been sampled in all cases in our series. In this study, tumours with an indistinct border tended towards an insidious transition in cellularity; however, this did not reach statistical significance. Similar findings were not evident on the T<sub>2</sub>-weighted images, which highlights the discrepancy between tumour margin assessed using CT or T<sub>1</sub>-weighted MRI and T<sub>2</sub> sequences (Kelly *et al.*, 1987).

As in previous studies, calcification assessed histologically was associated with genotype (Megyesi *et al.*, 2004); 18 out of 27 calcified tumours had 1p/19q loss. However, not all tumours with the -1p/-19q genotype were calcified, and calcification was also present in nine tumours with intact 1p and 19q. It is therefore unlikely that calcification is a direct biological effect of 1p/19q loss; indeed, calcification is also seen in pure astrocytomas (Kleihues, 2000).

Previous studies have postulated that the MR imaging characteristics of indistinct T<sub>1</sub> border and mixed signal intensity reflect increased 'invasiveness' associated with the -1p/-19q genotype in newly diagnosed, untreated oligodendrogliomas (Megyesi *et al.*, 2004). The present series included examples with and without the -1p/-19q genotype

**Table 6** Characteristics of the 14 cases with sharp/smooth imaging features

1p/19q intact	T <sub>1</sub> signal <sup>a</sup>	T <sub>2</sub> signal <sup>a</sup>	Contrast enhancing <sup>b</sup>	Subtype	Growth pattern <sup>c</sup>	Location	Cortex involved <sup>b</sup>	Transition in cellularity <sup>d</sup>
Yes	-	+	+	OII	II	Frontal	-	-
Yes	-	-	-	OII	III	Parietal	+	-
Yes	+	+	-	OAI	II	Temporal	-	-
Yes	+	+	-	OAI	II	Frontal	-	+
Yes	-	-	+	OAI	II	Temporal	-	+
Yes	-	-	-	OAI	III	Parietal	+	-
Yes	+	-	-	OAI	III	Temporoparietal	+	-
Yes	+	+	+	OAI	III	Frontal	+	No data
Yes	-	-	-	OAI	III	Frontal	-	-
No	-	-	+	OAI	I	Frontal	+	No data
Yes	+	+	No data	OAI	II	Frontal	+	-
Yes	-	-	+	OAI	III	Temporal	-	-
Yes	+	+	+	OAI	III	Frontoparietal	+	-
Yes	-	-	+	OAI	III	Frontal	-	-

<sup>a</sup>+, homogeneous; -, heterogeneous; <sup>b</sup>+, yes; -, no; <sup>c</sup>I, solid; II, mixed; III, infiltrative; <sup>d</sup>+, marked; -, insidious.

with MRI characteristics considered by neuroradiologists as typical of diffusely infiltrative tumours. Significant associations between genotype and radiological tumour border were found in the present study, tumours with a sharp border being likely to have intact 1p and 19q. Indeed, when all tumour border parameters were assessable ( $T_1$  and  $T_2$  border and  $T_2$  contour), 93% of tumours with a sharp, smooth border had intact 1p and 19q alleles. No features of neuroanatomy were evident to account for these sharp tumour boundaries. However, these imaging characteristics were not diagnostic of genotype, and 38% of tumours with indistinct, irregular margins also had intact 1p and 19q. The previous study described the association in reverse, that is, an indistinct  $T_1$  border in 17 of 26 cases was associated with 1p and 19q loss (Megyesi *et al.*, 2004). Whilst we found this feature in 40 of 42 cases with 1p and 19q loss, it was also present in 24 of 38 cases with intact alleles. In keeping with previous studies, loss of 1p and 19q was rarely associated with a sharp border, and no association between contrast enhancement and genotype was observed (Megyesi *et al.*, 2004). In addition, all cases with homogeneous signal intensity on  $T_1$  and  $T_2$ -weighted imaging had intact 1p/19q, and whilst heterogeneous signal intensity was present in both genotypes, it was more likely in tumours with 1p/19q loss. We did not find any associations between paramagnetic susceptibility effect and genotype (Megyesi *et al.*, 2004). Paramagnetic susceptibility was only assessable in 56% of our series as magnetization transfer was applied to  $T_1$ -weighted images in the remainder. We used the images illustrated in the previous study (Megyesi *et al.*, 2004) as a benchmark for assessment of all imaging parameters, but, nevertheless, such judgements are subjective and may be influenced by differences in image processing between different neuroradiology departments. Direct comparison with the previous study (Megyesi *et al.*, 2004) would require analysis of the newly diagnosed, previously untreated oligodendroglioma subgroup. Although this was not possible owing to sample size and the small number of cases with intact 1p/19q (6 out of 23), the imaging characteristics were similar to those of the series (Supplementary Table 1). In our study, the imaging features of sharp borders and signal homogeneity associated with intact 1p/19q were more common in oligoastrocytomas, but represented only 23% (12 out of 53) of oligoastrocytomas, 30% (13 out of 43) of those with intact 1p/19q and 16% (14 out of 86) of the series overall. Furthermore, although we have confirmed these imaging characteristics in a larger series, combined analysis of these and other series is necessary to establish further the relationship between MRI features and genotype in oligodendroglial tumours.

## Conclusions

Tumours with and without the 1p/19q genotype show similar cellularity transition at the radiological margin, and showed solid, mixed and infiltrative growth patterns, but infiltrative growth was more common in those with intact

1p/19q. Tumours with sharp, smooth imaging features and homogeneous signal intensity were more likely to have intact 1p/19q, which may be clinically useful in the absence of genetic testing, but these imaging factors were not related to histological growth characteristics. The lack of strong associations between genotype, imaging and growth suggests that these features are unlikely to account for the differences in clinical behaviour of these genetic subtypes of oligodendroglial neoplasms.

## Acknowledgements

We acknowledge the support of the Neuroradiology and Neuropathology departments at The Walton Centre and funding from Clatterbridge Cancer Research Trust, The Walton Centre for Neurology and Neurosurgery, and The Royal Colleges of Surgeons of Edinburgh and Ireland.

## References

- Barnard RO, Geddes JF. The incidence of multifocal cerebral gliomas. A histologic study of large hemisphere sections. *Cancer* 1987; 60: 1519–31.
- Batzdorf U, Malamud N. The problem of multicentric gliomas. *J Neurosurg* 1963; 20: 122–36.
- Bauman GS, Ino Y, Ueki K, Zlatescu MC, Fisher BJ, Macdonald DR, et al. Allelic loss of chromosome 1p and radiotherapy plus chemotherapy in patients with oligodendrogliomas. *Int J Radiat Oncol Biol Phys* 2000; 48: 825–30.
- Bigner SH, Rasheed BK, Wiltshire R, McLendon RE. Morphologic and molecular genetic aspects of oligodendroglial neoplasms. *Neuro-oncology* 1999; 1: 52–60.
- Cairncross JG, Macdonald DR. Successful chemotherapy for recurrent malignant oligodendroglioma. *Ann Neurol* 1988; 23: 360–4.
- Cairncross JG, Ueki K, Zlatescu MC, Lisle DK, Finkelstein DM, Hammond RR, et al. Specific genetic predictors of chemotherapeutic response and survival in patients with anaplastic oligodendrogliomas. *J Natl Cancer Inst* 1998; 90: 1473–9.
- Daumas-Duport C, Tucker ML, Kolles H, Cervera P, Beuvon F, Varlet P, et al. Oligodendrogliomas. Part II: A new grading system based on morphological and imaging criteria. *J Neurooncol* 1997a; 34: 61–78.
- Daumas-Duport C, Varlet P, Tucker ML, Beuvon F, Cervera P, Chodkiewicz JP. Oligodendrogliomas. Part I: Patterns of growth, histological diagnosis, clinical and imaging correlations: a study of 153 cases. *J Neurooncol* 1997b; 34: 37–59.
- Engelhard HH, Stelea A, Cochran EJ. Oligodendroglioma: pathology and molecular biology. *Surg Neurol* 2002; 58: 111–7; discussion 117.
- Engelhard HH, Stelea A, Mundt A. Oligodendroglioma and anaplastic oligodendroglioma: clinical features, treatment, and prognosis. *Surg Neurol* 2003; 60: 443–56.
- Fallon KB, Palmer CA, Roth KA, Nabors LB, Wang W, Carpenter M, et al. Prognostic value of 1p, 19q, 9p, 10q, and EGFR-FISH analyses in recurrent oligodendrogliomas. *J Neuropathol Exp Neurol* 2004; 63: 314–22.
- Felsberg J, Erkwow A, Sabel MC, Kirsch L, Fimmers R, Blaschke B, et al. Oligodendroglial tumors: refinement of candidate regions on chromosome arm 1p and correlation of 1p/19q status with survival. *Brain Pathol* 2004; 14: 121–30.
- Giese A, Bjerkvig R, Berens ME, Westphal M. Cost of migration: invasion of malignant gliomas and implications for treatment. *J Clin Oncol* 2003; 21: 1624–36.
- Ino Y, Betensky RA, Zlatescu MC, Sasaki H, Macdonald DR, Stemmer-Rachamimov AO, et al. Molecular subtypes of anaplastic oligodendroglioma: implications for patient management at diagnosis. *Clin Cancer Res* 2001; 7: 839–45.

- Jeuken JW, von Deimling A, Wesseling P. Molecular pathogenesis of oligodendroglial tumors. *J Neurooncol* 2004; 70: 161–81.
- Kelly PJ, Dumas-Duport C, Scheithauer BW, Kall BA, Kispert DB. Stereotactic histologic correlations of computed tomography- and magnetic resonance imaging-defined abnormalities in patients with glial neoplasms. *Mayo Clin Proc* 1987; 62: 450–9.
- Kitange GJ, Templeton KL, Jenkins RB. Recent advances in the molecular genetics of primary gliomas. *Curr Opin Oncol* 2003; 15: 197–203.
- Kleihues PC, Cavenee WK. Pathology and genetics of tumours of the nervous system. Lyon, France: IARC Press; 2000.
- Kraus JA, Koopmann J, Kaskel P, Maintz D, Brandner S, Schramm J, et al. Shared allelic losses on chromosomes 1p and 19q suggest a common origin of oligodendroglioma and oligoastrocytoma. *J Neuropathol Exp Neurol* 1995; 54: 91–5.
- Megyesi JF, Kachur E, Lee DH, Zlatescu MC, Betensky RA, Forsyth PA, et al. Imaging correlates of molecular signatures in oligodendrogliomas. *Clin Cancer Res* 2004; 10: 4303–6.
- Mukasa A, Ueki K, Ge X, Ishikawa S, Ide T, Fujimaki T, et al. Selective expression of a subset of neuronal genes in oligodendroglioma with chromosome 1p loss. *Brain Pathol* 2004; 14: 34–42.
- Palfi S, Swanson KR, De Bouard S, Chretien F, Oliveira R, Gherardi RK, et al. Correlation of in vitro infiltration with glioma histological type in organotypic brain slices. *Br J Cancer* 2004; 91: 745–52.
- Perry A. Oligodendroglial neoplasms: current concepts, misconceptions, and folklore. *Adv Anat Pathol* 2001; 8: 183–99.
- Reifenberger J, Reifenberger G, Liu L, James CD, Wechsler W, Collins VP. Molecular genetic analysis of oligodendroglial tumors shows preferential allelic deletions on 19q and 1p. *Am J Pathol* 1994; 145: 1175–90.
- Smith JS, Perry A, Borell TJ, Lee HK, O'Fallon J, Hosek SM, et al. Alterations of chromosome arms 1p and 19q as predictors of survival in oligodendrogliomas, astrocytomas, and mixed oligoastrocytomas. *J Clin Oncol* 2000; 18: 636–45.
- Tilgner J, Herr M, Ostertag C, Volk B. Validation of intraoperative diagnoses using smear preparations from stereotactic brain biopsies: intraoperative versus final diagnosis—influence of clinical factors. *Neurosurgery* 2005; 56: 257–65.
- van den Bent MJ. Diagnosis and management of oligodendroglioma. *Semin Oncol* 2004; 31: 645–52.
- van den Bent MJ, Looijenga LH, Langenberg K, Dinjens W, Graveland W, Uytendewilligen L, et al. Chromosomal anomalies in oligodendroglial tumors are correlated with clinical features. *Cancer* 2003; 97: 1276–84.
- Walker C, Joyce KA, Thompson-Hehir J, Davies MP, Gibbs FE, Halliwell N, et al. Characterisation of molecular alterations in microdissected archival gliomas. *Acta Neuropathol (Berl)* 2001; 101: 321–33.
- Walker C, du Plessis DG, Joyce KA, Machell Y, Thomson-Hehir J, Al Haddad SA, et al. Phenotype versus genotype in gliomas displaying inter- or intratumoral histological heterogeneity. *Clin Cancer Res* 2003; 9: 4841–51.
- Walker C, du Plessis DG, Fildes D, Haylock B, Husband D, Jenkinson MD, et al. Correlation of molecular genetics with molecular and morphological imaging in gliomas with an oligodendroglial component. *Clin Cancer Res* 2004; 10: 7182–91.
- Walker C, du Plessis DG, Joyce KA, Fildes D, Gee A, Haylock B, et al. Molecular pathology and clinical characteristics of oligodendroglial neoplasms. *Ann Neurol* 2005; 57: 855–65.