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Unique Patterns of Diffusion Directionality in Rat Brain Tumors Revealed by High-Resolution Diffusion Tensor MRI

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Abstract

Diffusion tensor imaging (DTI) is a new technique that uses the microscopic motion of water molecules to probe tissue three-dimensional (3D) microstructures. In this study, high-resolution DTI was performed on rats bearing intracranial 9L gliosarcoma, F98 glioma, and human glioblastoma. It was found that the tumors consisted of central zones with low diffusion anisotropy and peripheral structures (rim) with high diffusion anisotropy. In the rims, water diffusion directionality formed a circular pattern for the 9L and F98 tumors, and a radial pattern for the human glioblastoma xenografts. These well-organized diffusion patterns appeared at an early stage post-implantation and continued to exist with tumor growth in all three models. High-resolution ex vivo imaging and histology confirmed the in vivo findings. These distinct patterns, undetectable with conventional MRI, may reflect tumor organization and growth patterns at the cellular level.

Keywords

diffusion tensor imaging; DTI; high-resolution; glioma; diffusion pattern; microstructure; growth pattern

Malignant gliomas are the most common primary brain tumors. They are highly infiltrative, with cores of solid tumor (often mixed with necrosis) surrounded by regions of edematous brain infiltrated by tumor cells (1–5). Malignant solid tumors may destroy the brain parenchyma and are often accompanied by endothelial proliferation and the formation of new microvessels. The diagnostic, histologic features of glioblastoma distinctly include

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tissue necrosis and angiogenesis. Isolated tumor cells can spread very far from the solid tumor mass. Previous studies (1,3,4) have shown that infiltrating brain tumors migrate preferentially along blood vessel and white matter fiber tract pathways. Isolated tumor cells do not destroy the brain parenchyma and are usually not associated with angiogenesis. It is widely recognized that residual tumor cells remain after resection and are the source of tumor recurrence, contributing to the dismal prognosis for primary brain cancers. The local invasion of malignant gliomas ultimately decides the survival time for these patients. Therefore, it is critical to accurately map the growth patterns of malignant gliomas in the brain to aid in the development of therapies that can effectively arrest recurrence and progression.

It is known that the motion of water molecules is anisotropic in several biological tissues, such as brain white matter (6) and muscles (7). The diffusion tensor imaging (DTI) technique (8,9) can characterize the three dimensional (3D) properties of water diffusion in tissue by providing several types of information, including the extent and anisotropy of diffusion, and its predominant orientation. This capability of DTI can be used to detect the existence of well-ordered tissue at the microscopic level. For example, in axonal tracts, where neuronal fibers are coherently arranged and traveling in the same direction, the measured diffusion anisotropy is high and the predominant diffusion orientation is along the direction of fiber trajectories. Because of this, DTI has been used to map white matter tracts in the living brain (10,11) and to detect possible changes within diseased brain (12–18). In recent years, several groups (12–18) have used DTI to study the influence of infiltrating brain tumors on surrounding axonal tracts and to characterize the possible migration of tumor cells along neuronal tracts. Rat brain tumor models have been widely used in experimental neuro-oncology studies. In this study, we demonstrate that the DTI technique, when used under conditions of high spatial resolution, can probe microstructures at the cellular level inside experimental brain tumors, which are not visible with conventional MRI.

MATERIALS AND METHODS

Brain Tumor Models

Three tumor cell lines were used in this study. The F98 and 9L cell lines, originally derived from nitrosourea induced malignancies of glial origin, were cultured as previously described (19). The human Mayo 22 glioblastoma cell line (kindly provided by C. David James, Mayo Clinic, randomly called #22) was established from resected tumor, passaged serially as tumor xenografts, and cultured briefly prior to implantation to rat brain as previously described (20).

All surgical procedures were performed using proper aseptic techniques in a sterile field using sterile equipment. Eleven Fisher 344 rats and six nude rats (male, 200–250 g) were anesthetized by an intraperitoneal (i.p.) injection of 0.2 ml/100 g body weight of 2.5 mg/ml of xylazine and 25 mg/ml of ketamine hydrochloride. Through a small burr hole, tumor cells (25,000 9L gliosarcoma cells or F98 glioma cells in 2 μ l per Fisher rat, or 100,000 human glioblastoma cells in 2 μ l per nude rat) were stereotactically implanted to right caudate-putamen (3 mm lateral to the bregma and 4.5 mm deep). The scalp was re-approximated and the wound was closed with wound clamps. After surgery, buprenorphine hydrochloride (0.04 mg/kg body weight, i.p.) was used for analgesia. Upon awakening from anesthesia, the animals were returned to the caging area for study at a later time.

Animal Preparation for Imaging

On each day of the MRI measurements, tumor-bearing animals were re-anesthetized with 5% isoflurane in a mixture of 75% air and 25% O₂ in a box for about 5 minutes for induction, followed by 1.5–2% isoflurane through a mask during MR imaging. Rats were placed in a specially designed animal holder. To restrain head motions, the animals' head and body were fixed to the cradle with tape and ear pins. Rats in the magnet were monitored visually online through a small-animal monitoring and gating system connected by fiber optics, and the breathing rate was maintained at 40 ± 5 breaths per minute by adjusting the concentration of isoflurane (1.5–2%). At the end of the MRI study, anesthetized rats were perfused with phosphate buffered saline (PBS) followed by 4% paraformaldehyde, and brains were removed from the skull and preserved in 4% paraformaldehyde at 4 °C for a week for proper tissue fixation. After ex vivo MRI was done, the brains were sectioned, and histological sections (20 microns thick) were stained with hematoxylin and eosin. All the experimental procedures were approved by the Animal Care and Use Committee of the Johns Hopkins University School of Medicine.

In Vivo MRI

Imaging experiments were performed on a 4.7T horizontal bore animal MR system (Bruker Biospin, Billerica, MA) with an actively decoupled cross-coil setup of a 70 mm body coil for radiofrequency (RF) transmission and a 25 mm surface coil for signal reception. In vivo multi-slice images of rat brains were acquired in the horizontal plane (5 slices, slice thickness 1.5 mm, field of view (FOV) = 42 mm × 32 mm). The central imaging slice was set at the center of the tumor using acquired T₂-weighted (T₂w) localizer images in the coronal plane, and the location of the imaging slices was kept the same (the same distance from the top of the brain) for the same animal in subsequent experiments. High-resolution T₂W imaging with fast spin echo (FSE) acquisition was acquired (echo train length = 4, repetition time (TR) = 3 s, echo time (TE) = 64 ms, matrix = 256 × 192, resolution = 0.16 mm × 0.17 mm, number of average (NA) = 2). Diffusion tensor images were acquired using a multiple-slice, multiple-spin echo diffusion-weighted (DW) sequence (TR = 2 s, four echoes with TEs of 26.3/36.7/47.1/57.5 ms, matrix = 128 × 96, resolution = 0.33 mm × 0.33 mm, NA = 4). Trapezoidal diffusion gradients were used (Δ = 14 ms, δ = 5 ms, gradient rise time = 0.4 ms, gradient strength = 14 Gauss/cm). No eddy current compensation was employed, and the acquired images showed no apparent eddy current artifact. Seven DW images with different b values were acquired (one image with b value = 50 s/mm² and the rest with b value = 1000 s/mm²). Diffusion sensitizing gradients were applied along six different orientations: [0.707, 0.707, 0], [0.707, 0, 0.707], [0, 0.707, 0.707], [−0.707, 0.707, 0], [0.707, 0, −0.707], [0, −0.707, 0.707]. The images with the different TE values were averaged to improve the signal-to-noise ratio. The total imaging time for each animal was approximately 2 hours. For each animal, the MRI study was performed 2–3 times on different post-implantation days.

Ex Vivo MRI

One week after in vivo MRI, the brain samples were transferred into phosphate-buffered saline. Ex vivo DW images were acquired on a 9.4T NMR spectrometer using a 3D multiple-spin echo DW sequence, with the following imaging parameters: TR = 700 ms; TE = 30 ms; echo train length = 6; FOV = 26 mm × 13 mm × 13 mm; matrix = 160 × 82 × 82; resolution = 0.16 mm × 0.16 mm × 0.16 mm; and NA = 2. Seven DW images were acquired with the same diffusion parameters as in vivo MRI. T₂W images were acquired using a FSE sequence with the following imaging parameters: TR = 900 ms; TE = 50 ms; excitation pulse flip angle of 40°; NA = 4; and the same FOV and imaging matrix as the DW images. The total imaging time for each animal was about 20 hours.

Image Processing

The DTI data was processed in DTI Studio (developed by Dr. Hangyi Jiang and Dr. Susumu Mori; for more information, see <http://mri.kennedykrieger.org>). The diffusion tensors ($\bar{D}$) were calculated using a log-linear fitting method. Three pairs of eigenvalues (λ_1 , λ_2 , and λ_3) and eigenvectors (V_1 , V_2 , and V_3) were calculated for each pixel (21). The eigenvector associated with the largest eigenvalue was referred to as the primary eigenvector (V_1). Isotropic apparent diffusion coefficient (ADC) and fractional anisotropy (FA) (9) were calculated according to:

$$ADC = \text{Trace}(\bar{D})/3 = (\lambda_1 + \lambda_2 + \lambda_3)/3 = \bar{\lambda} \quad [1]$$

$$FA = \sqrt{3/2[(\lambda_1 - \bar{\lambda})^2 + (\lambda_2 - \bar{\lambda})^2 + (\lambda_3 - \bar{\lambda})^2]} / \sqrt{\lambda_1^2 + \lambda_2^2 + \lambda_3^2} \quad [2]$$

The orientation information was visualized through directionally encoded color (DEC) images (22), which were generated by combining the images of the primary eigenvector (V_1) and FA into RGB images. In this study, red was assigned to the medial-lateral orientation, green to the rostral-caudal orientation, and blue to the superior-inferior orientation. In the DEC images, the ratios among the green, red, and blue components of each pixel were defined by the ratios of the x, y, and z components of the primary eigenvector, and the intensity was proportional to the FA. The orientation information was also visualized by vector images, in which the measured V_1 at each pixel was visualized by a small line segment overlaid on DEC images, with the length of the line segment proportional to the FA value.

RESULTS

Water Diffusion Patterns in Tumors in Vivo

Figure 1 shows typical in vivo MR images from the three different brain tumor models, 9L, F98, and Mayo 22. In the T_2W images and ADC maps, regions with tumors had higher signal intensities than the striatum on the contralateral side. These results are consistent with previous reports (23), which show increases in T_2 and ADC values in tumors. Contrary to these conventional MR images, which render the tumor regions relatively homogeneous, the FA maps show high contrasts and interesting patterns within the tumors for all three models, which have not been previously reported. Inside the tumor regions defined by conventional MR images, there are small, dark central areas with a low or intermediate diffusion anisotropy, surrounded by structures (rim) with a high degree of diffusion anisotropy. The presence of a high diffusion anisotropy in the rims shows that water diffusion in certain regions is coherently, not randomly distributed, which clearly demonstrates the existence of a high level of organization at the cellular level inside these tumors.

DTI can measure the predominant diffusion orientation (V_1) at each pixel. In prominent white matter tracts, such as the corpus callosum and the hippocampal commissure (Fig. 1), the measured V_1 s were tangential to the trajectories of these axonal tracts. In the 9L tumor, the orientations in the rostral and caudal parts of the rim were along the medial-lateral orientation (red), while the orientations in the lateral parts were along the rostral-caudal orientation (green). This indicates that, water molecules in the rim primarily move in a circular fashion (surrounding the center of the tumor), as seen in the DEC or vector image. A similar pattern can be observed in the F98 tumor. On the other hand, the human Mayo 22 had different patterns of diffusion orientation compared to the 9L and F98 tumors. In the DEC image, the measured V_1 s in the rostral and caudal rims were along the rostral-caudal orientation (green), while the orientations in the lateral parts were along the medial-lateral

orientation (red). In the vector image, the measured V_1 s in the rim region with high diffusion anisotropy formed a radial pattern, suggesting that water molecules in these regions mainly diffuse in radial orientations (moving between the tumor center and outside through the rim).

Quantitative comparisons of DTI parameters measured in the central regions, rim regions, and contralateral striatum from a single horizontal slice through the center of each tumor are shown in Fig. 2. The rim was manually defined as a ring-like region with an apparent circular or radial pattern in the DEC image, while the region surrounded by the rim was defined as the central region (Fig. 2, diagram). Consistent with the above qualitative observations, the rims of the tumors generally had higher FA values than the centers, which were significant in the F98 and Mayo 22 models ($p < 0.001$), but not in the 9L model ($P > 0.05$). In the F98 and Mayo 22 models, the FA values in the centers were significantly lower than those in the contralateral striatum ($p < 0.001$), while the rims had significantly higher FA values than the contralateral striatum ($p < 0.001$). The differences were not significant for the 9L tumor. In contrast, the differences in ADC between the centers and rims of the tumors were not significant. In the 9L and F98 tumors, as expected (23), the values of ADC in the centers and rims were significantly higher than those in the contralateral striatum ($p < 0.01$). In the Mayo 22 model, the differences were not significant ($P > 0.05$).

Ex Vivo DTI

With ex vivo MRI, acquired with high spatial resolution in all three dimensions ($\sim 0.16 \times 0.16 \times 0.16 \text{ mm}^3$), we were able to examine the brain tumors in the horizontal, coronal, sagittal, or arbitrary orientations. Figure 3 shows examples of horizontal and coronal MR images acquired from the three experimental brain tumor models. Contrary to the in vivo cases, the T_2W images no longer provided good contrast between normal brain tissue and tumors. The tumor regions even showed T_2 hypointensity in the Mayo 22 xenografts. Significantly, ex vivo DTI results with high signal-to-noise ratios and higher spatial resolution confirmed our in vivo findings about the uniqueness of the structural patterns of tumor water diffusion. The comparison between the horizontal and coronal images shows that the distinctive patterns of water diffusion observed in vivo are not limited to a particular horizontal slice.

Histology

Hematoxylin and eosin-stained histology was performed to examine tumors directly for the presence of distinct cellular organizational patterns within tumors. Figure 4 shows the results for all three tumor models used in this study. For the 9L or F98 tumor models, no patterns similar to those revealed by DTI were visible in low magnification histology images (not shown). Examination under high magnification ($20\times$) showed that the orientation of individual tumor cells and the adjacent interstitial spaces formed a circular pattern in the outer part of the tumors, corresponding to the rims observed by DTI. In the case of the Mayo 22 glioblastoma xenografts, high magnification histology clearly showed tumor cells elongated and seemingly “streaming” in a radial pattern with parallel interstitial spaces, most apparent in the rims of the tumors. It is interesting that in the case of Mayo 22 tumors, small collections of radially oriented tumor cells were readily seen invading into the surrounding brain. Thus, the diffusion patterns of water molecules observed by DTI may reflect the patterns of the interstitial fluid flow and cellular organization within these tumors.

Diffusion Patterns on Different Post-implantation Days

To investigate how these diffusion patterns develop with time, we performed in vivo MRI (both DTI and T_2W) longitudinally on rats bearing intracranial 9L, F98, and Mayo 22 tumor models. Based on their relatively rapid growth properties, the 9L and F98 tumors were

imaged at 7, 10 or 11 days post-implantation, while the Mayo 22 xenografts were scanned at 28, 34, and 42 days post-implantation. Figure 5 shows the *in vivo* experimental results from three typical rats. The size of all three types of tumors clearly increased with time, as shown in the T₂W and ADC images. It is obvious that each type of tumor had the same diffusion pattern of water as those shown in Fig. 1. Even when the tumors were small (seven days for 9L and F98, and 28 days for Mayo 22), the 9L and F98 tumors had circular patterns of tissue diffusion orientation similar to those at a later stage (10 or 11 days), while the Mayo 22 tumors had radial patterns of tissue diffusion orientation similar to those at later stages (34 and 42 days). In the Mayo 22 model, even at a very late stage (42 days after implantation), when the tumor occupied the entire right striatum and deformed the surrounding tissue, the radial pattern was still well preserved in the tumors. These results show that the unique diffusion patterns (anisotropy and orientation) are formed at an early stage and are preserved throughout the tumor growth in all three models.

For all three tumor models used in this study, most of the tumors had one center with low diffusion anisotropy, as observed in the FA and DEC images. However, we observed multiple centers in two of the six rats bearing 9L tumors (not shown) and one of the six rats bearing Mayo 22 xenografts (Fig. 5). In the cases of multiple centers, the tumors appeared to be composed of several small confluent tumor foci, each of which contained a dark center with low FA, and a rim with high FA surrounding the center. The orientation of water diffusion in the rim was circular for the 9L tumors or radial for the Mayo 22 tumor, exactly the same as with a single center. Although the FA map with multiple centers shows a striking contrast, the entire solid tumor appears as a relatively homogeneous region in T₂W and ADC images.

DISCUSSION

Unique Contrasts from DTI

In this study, we have shown that the DTI technique, when used under conditions of high spatial resolution, provides useful information on tumor architecture for tumor detection and characterization. As shown in Fig. 1, all three tumors appear relatively homogeneous in conventional T₂W and ADC images, while the FA images generated from DTI measurements reveal high contrasts in the tumor regions. Particularly interesting is the presence of rim regions with a high diffusion anisotropy within the tumor areas. In the FA images, the central region of tumor has a low diffusion anisotropy, surrounded by a rim region of a high diffusion anisotropy. Both conventional MRI and histology show that the central zones with low FA are not necrotic. Further examinations show that the combined center and rim regions are generally smaller than the tumor areas defined by T₂W and ADC hyperintensity, which may be due to either the mass of solid tumor cells or to peritumoral vasogenic edema. Using diffusion anisotropy and orientation, DTI provides unique MRI contrasts for characterizing tumors.

Microstructures of Gliomas

It is commonly believed that the MRI contrasts generated by DTI reflect the arrangement of cells, membranes, and other structures inside a pixel. In axonal tracts, where neuronal fibers are coherently arranged and traveling in the same direction, the measured water diffusion anisotropy is high and the diffusion orientation is along the direction of fiber trajectories. However, the high diffusion anisotropy with the circular or radial pattern inside these solid tumors can not be explained by neuronal fibers that are distinctly absent from these tumor models that are mainly composed of tumor cells. Previous studies (24) have reported high diffusion anisotropy and coherently arranged diffusion orientations in the embryonic and neonatal cortex, probably due to the columnar structures of migrating neurons and radial

glia. The experimental results in this study show that the tumors consist of centers with low FA, surrounded by rims with high FA. In the rims of the 9L and F98 tumors, it is easier for water molecules to diffuse in a circular direction (i.e., within the rims) than in a radial direction (i.e., crossing the rims), while for the human Mayo 22 model, it is easier for water molecules to move in a radial direction than in a circular direction. The organized directional diffusion of water molecules in these solid tumors was found to reflect the directional organization of cells within the rims unique for different tumor models. The tumor rims displaying circular water diffusion contained cells that were elongated and organized in a circumferential direction. In contrast, tumor rims with radial water diffusion contained cells that were elongated and organized radially. In the latter cases, thin aggregates of tumor cells could be found invading as an extension of this radial pattern from the tumor margin into surrounding brain.

Malignant tumor cells both proliferate and migrate in the extracellular matrix of the brain. Gliomas are often described as consisting of a central proliferation zone and a surrounding invasive zone of lower cellular density. Rates of proliferation and infiltration are not strictly correlated. Many efforts have been applied to investigate the dynamics of malignant tumor growth in vitro (25–27) and in vivo (28,29). It has been suggested (25) that a malignant brain tumor is an opportunistic, self-organizing, and adaptive dynamic biosystem rather than an unorganized cell mass. Both theoretical and experimental results clearly show that the dynamics and resulting patterns of tumor growth are quite different for different tumor cell lines. This is supported by the findings in this study. The 9L and F98 brain tumors grew rapidly. Generally, the tumor reached a size detectable by MRI at one week post-implantation and most animals implanted with tumors died within 2–3 weeks post-implantation. In comparison, the Mayo 22 xenografts grew slowly and tumor-bearing animals usually lived for 2–3 months post-implantation. Whether rapidly growing tumors form circular patterns, or tumors with circular patterns grow faster is not addressed in this study. The distinct patterns of directional diffusion of water molecules inside these solid tumors were correlated with their microstructures and growth patterns. The cellular, biochemical, and molecular determinants of the distinct organization of these tumors as revealed by DTI remain to be determined. Possibilities include specific cell surface and extracellular matrix determinants of tumor cell adhesion and shape, cytoskeletal determinants of cell shape, and potentially tumor proliferation and invasiveness.

Comparison with Human DTI

Using DTI, both brain tumors and adjacent brain tissues, including white matter tracts and the cerebral cortex, have been studied intensively in recent years (12–18). It was found that growing brain tumors have an influence on the fiber tracts in the brain, and abnormalities may be categorized by displaced, infiltrated, disrupted, and edematous patterns (13,18). To the best of our knowledge, these DTI studies were all performed on human patients harboring brain tumors, except for a recent conference abstract (30), in which the 9L tumor model was scanned. Compared to our results, these human studies generally noticed decreased, low FA values inside the tumors and no specific diffusion patterns were reported. The fact that the unique diffusion patterns seen in our DTI studies on the experimental brain tumors have not been observed in DTI of patients with brain tumors may be attributed to two factors. The first and foremost factor is possible different characteristics between human brain tumors and tumors in animal brains. Brain tumor growth is very chaotic and includes areas of necrosis, micro- or macro-hemorrhage, and cystic components. These features are typically noted in a human glioma and form the basis of a diagnosis of a high-grade tumor. If these physiological features do exist, as seen in most clinical studies, one would expect the FA to go down as the tumor grows. The second factor is the high spatial resolution that was used in animal studies. Similar to conventional MRI, what DTI detects in a voxel is an

average of the entire voxel. In current, typical human DTI studies, the imaging resolution is about 2.5 mm in each dimension, much coarser than the resolution achieved in our animal studies ($0.33 \times 0.33 \times 1.5 \text{ mm}^3$). The voxel volume is about 100 times larger in human studies than in our animal studies. Increasing voxel size generally results in larger partial volume effects and may yield ambiguous DTI results (31).

It is possible that similar patterns of diffusion directionality may be associated with the location, size, time (e.g., at onset), and type of tumor in humans. The observation of these is currently hampered by spatial resolution limitations, but may become possible when spatial resolution for DTI improves to the sub-millimeter level in the future. It was reported recently (32) that the high signal-to-noise ratio achieved with the multi-channel array receivers at a 7T human MRI scanner enabled an unprecedented spatial resolution of up to $0.2 \times 0.2 \times 0.5 \text{ mm}^3$ in a simple gradient echo approach. DTI experiments require much longer TE and even though spin echoes are used, signal loss at 7T may be substantial in view of the very short T_2 of white matter tissue (on the order of 40 ms or less). Depending on available gradient strength, such fields may therefore require the use of stimulated echoes for diffusion imaging. For rapid imaging at high spatial resolution, the acquisition time and segmentation may be shortened by using a reduced FOV, for instance, through the use of a small surface coil and outer-volume suppression. Based on this zoomed imaging approach, a single-shot acquisition has been performed at sub-millimeter resolution in the human brain at 7T (33). We therefore expect that it is technically possible to achieve DTI experiments with sub-millimeter resolution in humans in clinically feasible times.

CONCLUSIONS

High-resolution DTI was applied to 9L, F98, and Mayo 22 tumor models. The FA and DEC maps clearly showed that the solid tumors consist of centers with low FA and peripheral structures with high FA, circular (9L and F98) or radial (Mayo 22) diffusion orientations. These unique water diffusion patterns within tumors, not observed on conventional T_2W and ADC images, are formed at an early stage post-implantation and continue to exist with tumor growth in all three models. The organized directional diffusion of water molecules within solid malignancies has never before been imaged. DTI at the sub-millimeter scale can identify brain tumors with patterns of cellular organization, which are distinct among different tumor models. The new findings may provide a new approach to investigate tumor microstructures at the cellular level and to monitor the growth of malignant gliomas in vivo.

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FIG. 1.

In vivo MR images of 9L gliosarcoma, F98 glioma, and Mayo 22 glioblastoma brain tumor models. Post-implantation days were 11 (9L), 10 (F98), and 27 (Mayo 22). cc stands for corpus callosum; hc for hippocampal commissure. The tumor regions are hyperintense on the T₂W and ADC images. The boundaries of T₂W hyperintense tumor regions were manually defined and overlaid on other images. In FA maps, the tumors show high contrasts and various degrees of high-level diffusion anisotropy. In DEC and vector images, the tumor areas exhibit a circular orientation for 9L and F98, or a radial orientation for Mayo 22 tumors, as illustrated by the insert diagrams.

FIG. 2.

Quantitative comparison of DTI parameters measured in different brain regions of 9L gliosarcoma, F98 glioma, and Mayo 22 glioblastoma brain tumor models ($n = 5$ for each group). Post-implantation days were 11 (9L), 10 or 11 (F98), and 26–28 (Mayo 22).

FIG. 3.

Ex vivo MR images from perfused rat brain samples with 9L, F98, and Mayo 22 glioblastoma xenografts. Post-implantation days were 11 (9L), 10 (F98), and 26 (Mayo 22). Transverse sections and coronal sections are shown. The tumor regions are hyperintense on the ADC images. The contrast between the tumor and surrounding brain tissues on the T₂W images is low. All three types of tumors show various degrees of high-level diffusion anisotropy and a unique arrangement of tissue diffusion orientation.

FIG. 4.

High magnification (20 $\times$) histology of 9L, F98, and Mayo 22 tumors. Post-implantation days were 11 (9L), 11 (F98), and 26 (Mayo 22). Bar was 0.1 mm. The morphology of individual tumor cells (blue) and the orientations of interstitial space (white) in the outer parts of the tumors (corresponding to the rims) form a circular pattern (9L and F98) or a radial pattern (Mayo 22). Note the collections of radially organized Mayo 22 tumor cells extending from tumor margins into surrounding brain.

FIG. 5.

In vivo MR images of 9L, F98, and Mayo 22 xenografts at different stages of tumor growth. A 9L tumor at 7 and 11 days post-implantation, an F98 tumor at 7 and 10 days post-implantation, and a mayo 22 tumor at 28, 34, and 42 days post-implantation. The boundaries of T₂W hyperintense tumor regions were manually defined and overlaid on FA images. In the FA and DEC images, tumor regions (inside the rectangular box) were enlarged and shown next to the DEC images. The tumors grow with post-implantation time, as observed within conventional T₂W and ADC images. The diffusion patterns appear at an early stage and enlarge with tumor growth in all three models. DTI reveals the presence of two centers for Mayo 22 xenografts, which are not visible with conventional MR images.