Available online at www.sciencedirect.com
ScienceDirect
Methods 43 (2007) 12–20
METHODS
www.elsevier.com/locate/ymeth

Diffusion weighted imaging in small rodents using clinical MRI scanners

Feng Chen [a,b], Frederik De Keyzer [a], Huaijun Wang [a], Vincent Vandecaveye [a], Willy Landuyt [c], Hilde Bosmans [a], Robert Hermans [a], Guy Marchal [a], Yicheng Ni [a,*]

[a] *Biomedical Imaging, Interventional Therapy and Contrast Media Research, Department of Radiology, University Hospitals Leuven, Herestraat 49, B-3000 Leuven, Belgium*
[b] *Department of Radiology, Zhong Da Hospital, Southeast University, 87 Ding Jia Qiao Road, Nanjing, China*
[c] *Department of Experimental Radiobiology and Oncology, University Hospitals Leuven, Herestraat 49, B-3000 Leuven, Belgium*

Accepted 27 March 2007

Abstract

Diffusion weighted imaging (DWI) has emerged as a unique and powerful non-invasive magnetic resonance imaging (MRI) technique with a major potential impact on imaging-based diagnosis in a variety of clinical applications including oncology and tissue viability assessment. In light of increasing demand for applying this technique in preclinical investigations using small animals, we have explored the potentials of a clinical magnet for acquiring the DWI in rats and mice with either cerebral ischemia or solid tumors. Through technical adaptation and optimization, we have been able to perform a series of clinically relevant animal studies with conclusions based on DWI quantification. Focusing more on practical aspects and cross-referencing with the current literature, this paper is aimed to summarize our ongoing DWI studies on small rodents with stroke and tumors, and to provide protocols for researchers to replicate similar techniques in their own preclinical and clinical studies.

Keywords: Magnetic resonance imaging (MRI); Diffusion weighted imaging (DWI); 1.5 T; Clinical MRI scanner; Small rodents; Rats; Mice; Stroke; Cerebral ischemia; Neoplasm; Subcutaneous; Liver tumor; Imaging diagnosis; Tissue characterization

1. Introduction

The advent of *in vivo* magnetic resonance imaging (MRI) has enabled non-invasive examinations of morphological and functional changes in the normal and diseased biological systems. After two decades of intense research, diffusion weighted imaging (DWI) has progressed from the experimental to the clinical field and may have a major potential impact on imaging diagnosis [1]. DWI has been extensively studied in both neurological and extraneurological applications including cerebral ischemic stroke [2,3], tumor grading [4,5], differential diagnosis of benign and malignant tumors [6–8], characterization of viable and necrotic tissues [9–11], assessment of organ functions [12], prediction of pathologic outcomes [13,14], and evaluation of tumor response to therapies [15–17].

DWI is an MRI technique that provides *in vivo* information on the random Brownian motion of hydrogen protons in tissues. To make this possible, a fast MRI sequence is supplemented by two equal-sized but opposite gradients. The combination of these two gradients induces a signal loss in all moving hydrogen protons but has no effect on stationary hydrogen protons. In other words, this signal loss is more pronounced in tissues where a lot of movement is present, while the signal is less affected in tissues with restricted proton movement. Thus, the contrast in the resulting images is provided by the different tissue microstructures which facilitate or hamper hydrogen proton movement. If we make a few separate acquisitions with different b values, which indicate the motion sensitivity induced in the images, we can quantify this signal loss using the apparent diffusion coefficient (ADC).

* Corresponding author. Fax: +32 16 34 37 65.
E-mail address: Yicheng.Ni@med.kuleuven.be (Y. Ni).
URL: http://www.kuleuven.be/cv/u0014498e.htm (Y. Ni).

1046-2023/$ - see front matter © 2007 Elsevier Inc. All rights reserved.
doi:10.1016/j.ymeth.2007.03.007

Lower b value (e.g. $\leqslant 300$ s/mm^2) images are especially sensitive to the very fast moving protons, such as those in the blood vessels or microvasculature, while the higher b value (e.g. >300 s/mm^2) images are predominantly affected by the less movable protons in the extracellular and intracellular spaces [18].

Small rodent (mouse and rat) models are extensively used in biomedical research. Dedicated small-bore high field MR magnets have been developed for the purpose of imaging these animals. However, such special equipment is not always available or accessible particularly to many of the clinically based researchers. Alternatively, lower field ($\leqslant 3$ T) clinical MR scanners can also become practical tools for rodent MR studies as long as the MRI acquisition techniques are adequately adapted and optimized. Besides, clinical magnets could be advantageous over small animal MRI scanners due to easier translation of research outcomes from small rodents to patients.

Although technically challenging, DWI in rats [19–27] and mice [11] has been attempted and reported by a number of study groups using clinical MRI scanners.

To meet the needs in this particular field, focusing more on practical aspects, this paper summarizes our ongoing experiences in DWI studies on small rodents with cerebral ischemia or tumors at a clinical MR scanner in correlation with the current literature [11,19–22,24–26]. Detailed protocols are provided for researchers to replicate similar techniques in their own preclinical and clinical studies.

2. Methodological descriptions

2.1. MRI equipment and accessories

Rodent MR imaging is typically performed in our center with a clinical 1.5 T whole-body system (Sonata, Siemens, Erlangen, Germany) with a maximum gradient capability of 40 mT/m. To allow parallel imaging, phased array coils such as the commercially available 4-channel phased array wrist coil (MRI Devices Corporation, Waukesha, WI, USA) are used for shortened imaging acquisition time and improved signal homogeneity. However, small single-element surface loop coils may show benefits for a high local signal-to-noise ratio (SNR) and favourable cost-effectiveness. Imaging acquisition parameters given in Table 1 are applicable for both coils except the parallel imaging that can only be acquired with phase array coil.

2.2. Preparations of animals for DWI

Study subjects consisted of rat models of stroke with photochemically induced thrombosis (PIT) of proximal middle cerebral artery (MCA) as well as subcutaneously and intrahepatically implanted rhabdomyosarcoma (R1) as described in details elsewhere [20,22,25]. In addition, female C3H mice (weight range 21–25 g, purchased from Charles River Laboratories, Les Oncins, France) with liver implantation of radiation induced fibrosarcoma (RIF-1) have recently been included in our ongoing research.

2.2.1. Brain imaging

In order to obtain sufficient SNR as well as spatial resolution with DWI, two important measures proved effective. First, dental drills made of carbide (BRASSELER GmbH, Lemgo, Germany) instead of iron-based stainless steel are recommended for opening the skull window when preparing brain models. This way the microscopic metal pieces in the operative field and therefore the artifacts resulting from the proton relaxation susceptibility can be avoided. Second, any cavities or interspaces caused by surgical procedures should be filled with gelatin sponge, which not only effectively controls hemorrhage but also eliminates the air–tissue interfaces [19,20]. Both of these measures increase the SNR and minimize local distortion artifacts.

2.2.2. Body imaging

Especially in extracranial applications, susceptibility artifacts and geometric distortion are two of the major problems encountered in performing DWI that is particularly vulnerable to air–tissue boundaries. These effects can be minimized by using a mold application procedure, as previously validated and recommended [28]. The detailed mold application procedure is described in the aforementioned reference. Briefly, a fast-setting alginate powder Xantalgin® Select (Bayer Dental, Leverkusen, Germany) is commercially available for dental industry. The mold is made by mixing and stirring 100 g or 20 g of powder with 300 ml or 60 ml of 40 °C tap water to form a viscous mass that stiffens gently in less than two minutes for use in rats or mice, respectively. Before stiffening, the mass is poured into the plastic holder where the rodent is placed supine and is applied securely covering the tumors and the major part of the body up to the thoracic base. The hair around the area of interest has to be shaved to ensure direct contact of the mold material and skin. The formed mold should be about 1.5 cm thick for improved image quality. The mold remains somewhat flexible, and does not interfere with the breathing of the animals and can afterwards be removed manually from the body without any difficulty.

This mold technique has been successfully introduced to evaluate modulation of tumor oxygenation [29]. This study demonstrated the application of this rapid and easy procedure to reduce magnet susceptibility artifact associated with gradient-echo based ultrafast sequences such as blood oxygen level-dependent (BOLD) MRI. Also with DWI, improved image quality and better delineation of the anatomical structures results from the use of the mold (Fig. 1).

For rodent brain, the value of mold application is limited due to the unavoidable air within the external and internal acoustic meatus and air cells in the petrous bone. For a rat liver tumor model, the mold application is also of less value since the main imaging artifacts are caused by respiratory movement, bowel contents and peristalsis.