Fluorine-19 NMR Imaging of the Biodistribution and Metabolization of the Antineoplastic Agent Gemcitabine in Tumor-Bearing Rats

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Introduction

The deoxycytidine analogue gemcitabine® (2',2'-difluoro-2'-deoxycytidine, dFdC) is a new potent anticancer drug with novel mechanisms of action [1]. The prodrug dFdC is activated intracellularly by nucleoside kinases to its triphosphate derivative dFdCTP. After dFdCTP incorporation into the DNA, one additional nucleotide is added to the growing DNA strands and proofreading enzymes are unable to remove dFdCTP from its 'locked' position. Thus, further DNA synthesis is inhibited ('masked chain termination') which leads to the induction of apoptosis [1].

Purpose

To develop and to apply a chemical-shift selective (CHESS) ^{19}F NMR imaging technique to map the biodistribution and metabolization of dFdC in tumor-bearing animals.

Methods

The dFdC uptake was mapped in six ACI rats with *s. c.* implanted Morris hepatoma (MH3924A) and in four Copenhagen rats with *s. c.* transplanted Dunning prostate adenocarcinoma (R3327-AT1). The experiments were performed on a 1.5-Tesla whole-body NMR system (MAGNETOM 63/84 SP®, Siemens AG) equipped with an additional frequency selective duplexer and preamplifier for ^{19}F NMR spectroscopy. A double-tuned linear-polarized animal resonator was employed for RF transmission and detection at ^{19}F and ^{1}H frequencies. For ^{19}F NMR imaging, a FLASH pulse sequence (TR = 100 ms, TE = 2.7 ms, TH = 15 mm, MS = 16×16, FOV = 160 mm) was used [2, 3]. A CHESS RF pulse was employed to presaturate the dFdC resonance or the dFdC-metabolite peaks before the desired ^{19}F NMR resonance was selected for image formation. The number of excitations was NEX = 500, resulting in a measurement time of 13 min for a selective ^{19}F NMR image. Immediately after *i. v.* administration of dFdC (dose, 200 mg/kg body-weight) into the tail vein of the animal, a dynamic series of ^{19}F NMR spectra was recorded, followed by the interleaved measurement of the CHESS dFdC and ^{19}F-metabolite images. Afterwards, the ^{19}F matrix size was expanded from 16×16 to 256×256 picture elements by zero-filling in the frequency domain.

Results

The dynamic series of slice-selective ^{19}F NMR spectra (1.7 min acquisition time) showed *in vivo* first the prominent dFdC resonance, resonating at $\delta = -42.3$ ppm with respect to an external standard of trifluoroacetic acid (TFA). Shortly after dFdC administration (3–5 min *p. i.*), a strong ^{19}F-metabolite resonance (called X_1, $\delta = -10.5$ ppm) appeared. At about 5–10 min *p. i.*, an additional, weak peak (called X_2, $\delta = -81.5$ ppm) could be detected in the fully-relaxed ^{19}F spectra. As an example of the dFdC imaging experiments, the ^{1}H spin-echo image of a tumor-bearing ACI rat is depicted in Fig. 1A. The corresponding CHESS ^{19}F dFdC map is shown in Fig. 1B (spatial resolution = 10×10×15 mm^3, sum of three interleaved 13-min ^{19}F images). Note the marked dFdC uptake in the kidneys, the liver, and the bladder while an heterogeneous dFdC signal distribution could be observed in the tumor. The ^{19}F signal from a reference vial filled with an aqueous 5 mM dFdC solution is clearly visible. The CHESS ^{19}F image generated from the X_1 resonance is shown in Fig. 1C. In this selective map, the ^{19}F signal from the dFdC vial is completely suppressed which demonstrates the editing efficiency of our CHESS RF pulse scheme.

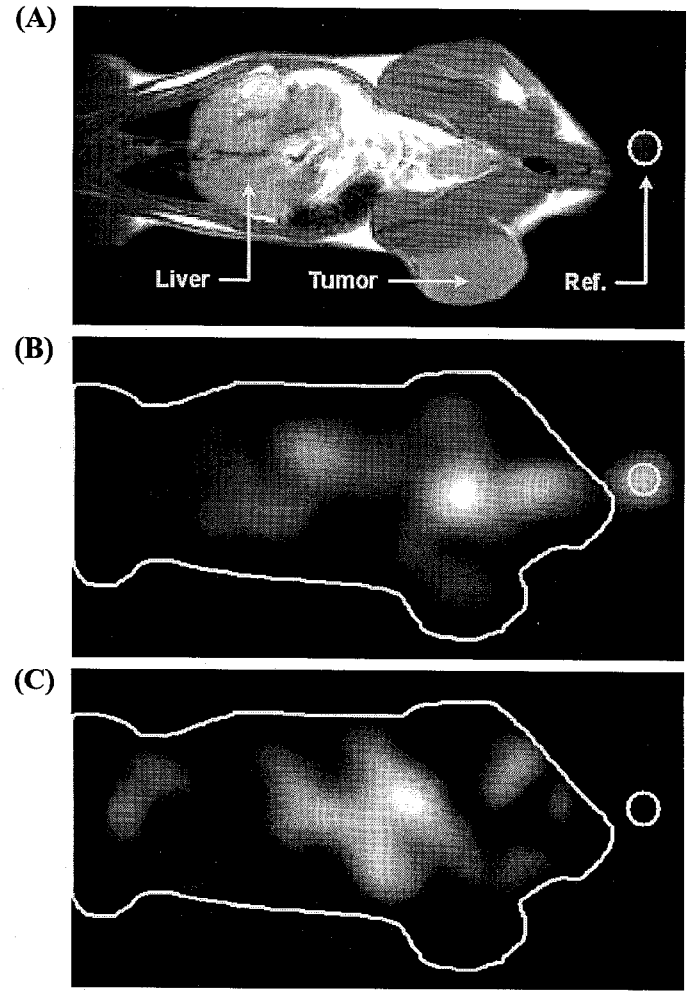

Figure 1. Coronal NMR images of an ACI rat with *s. c.* transplanted Morris hepatoma. **(A)** T_1-weighted ^{1}H spin-echo image. **(B)** Drug-specific ^{19}F dFdC FLASH image obtained 59 min post dFdC injection. **(C)** Selective ^{19}F-metabolite map from the X_1 resonance at −10.5 ppm measured 76 min after dFdC administration.

Discussion

CHESS ^{19}F NMR imaging provides an elegant means to measure the biodistribution and metabolization of ^{19}F-containing antineoplastic agents such as dFdC. For the *in vivo* quantification of drug concentrations with ^{19}F NMR, however, the exact knowledge of the T_1 and T_2* relaxation times in the various tissue compartments is mandatory. Theoretical considerations suggest that the chemical shift of the X_1 peak might be explained by hydrofluoride while the X_2 resonance is evidence for fluorine in an sp^2 orbital. For the correct assignment of the X_1 and X_2 peaks, the acquisition of high-field ^{19}F NMR spectra from *ex vivo* samples is under way to measure the small ^{19}F J-couplings which are not resolved at 1.5 Tesla.

References

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