Molecular dynamics and information on possible sites of interaction of intramyocellular metabolites *in vivo* from resolved dipolar couplings in localized ^{1}H MR spectra

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INTRODUCTION:

Residual dipolar couplings affecting ^{1}H MR resonances of endogeneous metabolites in human muscle tissue have been discovered 10 years ago [1]. A detailed analysis of this direct proton-proton interaction provides information on the mobility of molecular groups. The purpose of this study was to explore molecular dynamics of (phospho)creatine ((P)Cr), taurine (Tau), and carnosine (Cs) in human muscle tisssue *in vivo* by analyzing localized high-resolution ^{1}H MR spectra .

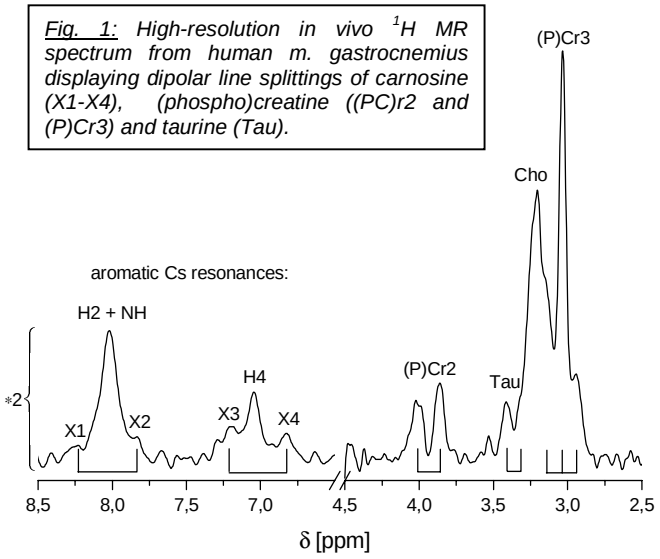
MATERIALS AND METHODS:

Experiments were performed on a clinical whole-body MR scanner at 1.5 T (Magnetom Vision with Helicon magnet, Siemens) with n = 5 male healthy volunteers (age 19-27 ys). Spectra of (2 cm)^3-VOIs from m. gastrocnemius were obtained using PRESS with TE = 36…146 ms and STEAM with TE = 20 ms (TR = 2 s each). The first technique was used to study phase modulation of the (P)Cr methylene doublet which depends linearly on TE, the latter to determine the resolved multiplet structures of the (P)Cr CH_3 triplet, Tau CH_2 doublet, and Cs multiplets (two doublets X1,X2 and X3,X4 assigned to the imidazole ring protons H2 and H4; Fig. 1). The Cs spin system exhibits second-order spectra [2] which allow to determine the dipolar coupling strength independently of the line splitting. All line splittings SD_0 were compared to the coupling constant D_0, (given by the internuclear distance and assuming completely freeze molecular libration) in order to get the order parameter S, which is a measure of molecular mobility. Effective motional averaging yields small values of S. Additionally, experiments on model solutions of histidine (His) and *N*-acetyl-*L*-aspartate (NAA) were performed to assign an unexpected signal of a NH group of Cs at 8 ppm.

RESULTS:

The values of S we obtained by analysis of highly resolved ^{1}H MR spectra indicate striking differences in reorientation of the spin systems and involve different magnitudes of line splittings of the multiplets (Fig. 1). While the aliphatic resonances reflect a high degree of mobility of the CH_3 and CH_2 groups of (P)Cr and Tau ($S \cong (1.4 - 3.0)\times10^{-4}$), the anisochronous spin system H2–H4 is characterized by $S \cong 125\times10^{-4}$. Additionally, comparison with spectra of model solutions indicates reduced chemical exchange of the NH proton at the peptide bond of Cs. This effect produces an unresolved doublet at 8 ppm *in vivo*.

Fig. 1: High-resolution in vivo ^{1}H MR spectrum from human m. gastrocnemius displaying dipolar line splittings of carnosine (X1-X4), (phospho)creatine ((PC)r2 and (P)Cr3) and taurine (Tau).



DISCUSSION:

The high value of S for Cs can be explained by the stiffness of the imidazole ring which enables a quite effective spin-spin interaction if the ring is hindered in motion (e.g. by interactions of the nucleophilic nitrogen N^1). In distinct contrast, the CH_2 and CH_3 subgroups are not embedded in a rigid structure. Additionally, the detection of the Cs NH group may be attributed to hydrogen-bond formation with a larger structure and underlines the highly reduced mobility of Cs.

CONCLUSION:

Detailed analysis of high-resolution ^{1}H MR spectra provides information on molecular mobility *in vivo*. Since the restriction in reorientation can be explained by interactions of the metabolites with larger structures (*e. g.*, cellular membranes or the actin/myosin filaments) MRS can contribute to studies of intermolecular processes. For example, Cs is suspected to react with phospholipids during cell aging [3] and the immobilized pool of this metabolite which is detectable by *in vivo* ^{1}H MRS could add an interesting aspect to this process.

REFERENCES:

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