CEST-MRI for sugar and amino acid imaging in plant specimen

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INTRODUCTION: Chemical Shift Imaging (CSI) enables the *in vivo* detection of metabolites in living plant tissues (1). However, it has some major disadvantages: especially magnetic field inhomogeneities, which often occur inside plants due to their heterogenous structures, have strong influence on the quality of the NMR spectra. Therefore, in this work we applied CEST for the first time to plant seeds (barley) to detect sugars and amino acids, as these are the most abundant metabolic components delivered to the largest storage organ (endosperm) of barley.

METHODS: MR measurements on young barley grains were performed on a 9.4 T MRI scanner (Bruker BioSpin). We conducted CEST experiments in the range from ±5 ppm to detect the exchanging protons of the hydroxyl group (at around 1 ppm) and the amino group (3 ppm). It was found that even small saturations, such as a single block pulse of 200 ms duration and 2 µT amplitude, provided sufficient CEST contrast. For read-out a 2D spin echo sequence (RARE) was used. Before calculating the asymmetry spectra MRT_{asy}, Z-spectra were B_0-corrected using the WASSR technique (2). For comparison, CSI experiments were performed on the same slices. The spatial resolution of the CEST experiments was 80 µm, whereas for CSI only lower resolutions (above 300 µm) were possible in a similar time. CEST sugar (amino acid) maps were generated by integrating over the 1 ppm (3 ppm) peak in the asymmetry spectrum.

RESULTS: Both, sugars and amino acids, could be detected in the endosperm of barley grains by CEST (see Fig.1b). CEST exhibited a much lower sensitivity to magnetic field inhomogeneities than CSI: The endosperm of barley is shaped in two symmetrical wings, and the CEST spectra in these wings matched perfectly. Whereas the CSI spectra varied due to differences in T_2^* within the endosperm making the comparison of CSI spectra unreliable (see Fig.1c). In the following, a growing barley grain (attached to an intact ear and supplied with a nutrient solution containing sucrose) was monitored via CEST over 60h (see Fig. 2). During the experiment, the endosperm size grew significantly. The CEST signal of the amino acids decreased continuously, whereas the accumulation of the sugars peaked at approximately 46 h after the start of the experiment.

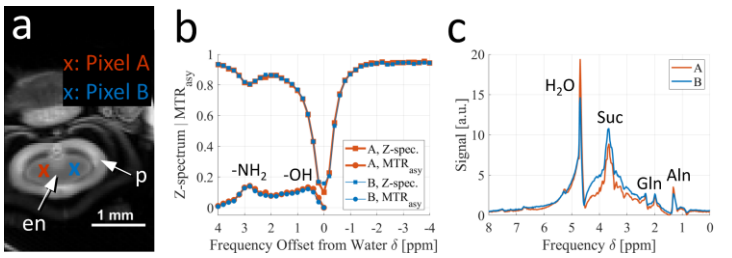

Figure 1. CEST and CSI measurement on barley. (a) Structural image of the examined slice. (b,c) Two exemplary CEST and CSI spectra from the endosperm, positions labeled in (a). Abb.: en, endosperm; p, pericarp.

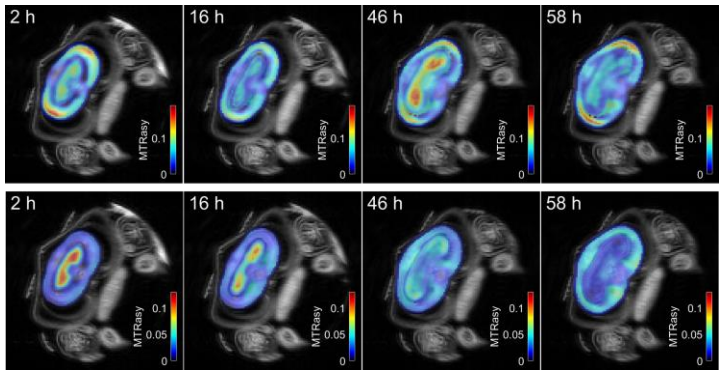

Figure 2. CEST maps during sucrose feeding of barley caryopsis. CEST MTR_{asy} maps for sugars (upper figures) and amino acids (lower figures) at different times.

DISCUSSION: It was demonstrated that CEST is a versatile tool for the detection of sugars and amino acids in plant seeds. Monitoring dynamic growing processes is an interesting application where CEST-MRI is not only feasible, but provides new insight into the inner workings of intact plant tissues. In contrast to CSI, CEST does not enable the differentiation between individual sugars or amino acids, but delivers higher spatial resolutions in similar acquisition times. Furthermore, the decreased sensitivity to magnetic field inhomogeneities, which usually occur in plant tissues, is possible the most important advantage of CEST compared to CSI. Intrinsic B_0-correction of the Z-spectra is of fundamental importance, especially for the calculation of MTR_{asy} for sugar signals.

CONCLUSION: In our opinion, CEST-MRI is a promising tool for examining plants and should be pursued in future research.

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REFERENCES:

1. Borisjuk L, Melkus G, In:In vivo imaging: new research. New York: Nova Science Publishers (2014).
2. Kim M, *et al.* Magn Reson Med 2009;61:1441-1450.