Clues for the differentiation between autolytic and bacterial products in decomposing brain

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Introduction: ^{1}H-MRS studies have shown that several otherwise unobservable metabolites become major constituents of decomposing sheep and human brain after about three days post mortem [1]. Further investigations demonstrated reproducible time courses of concentration changes for various metabolites under laboratory conditions. Parameterized models of these time courses hence allow for a back calculation of the postmortem interval PMI [2]; an information that is relevant for a majority of criminal investigations. However, mortal incidents do not happen under laboratory conditions and a thorough understanding of the decomposition mechanisms is a prerequisite to take variations of external conditions into account. One of the major steps towards this understanding is the differentiation of autolytic and bacterial products since it can be expected that the environmental temperature influences the kinetics of these two processes differently.

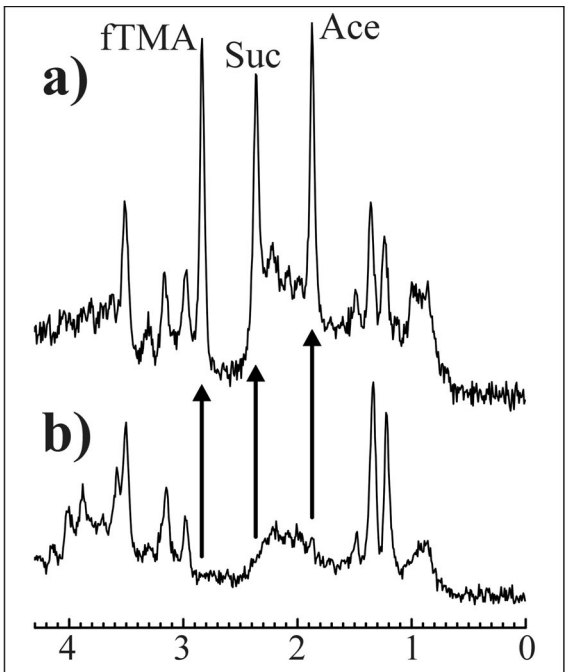

Fig 1: comparison of a spectrum from a body with a PMI of 520 hours a) with the baby spectrum b) (PMI=897 hours). The arrows indicate the absence of free trimethyl-ammonium, succinate and acetate.

Materials & Methods: In situ single voxel ^{1}H-MRS of brain tissue was performed on a total of 32 human bodies from the institute of forensic medicine (General Electric SIGNA, 1.5T, PRESS, TR=3s, TE=20ms) with PMIs between 11 and 920 hours. We report here on a specific case of a baby dying during birth who had to be exhumed after roughly five weeks for legal reasons (=> PMI = 897hours). Bacterial analysis was performed on brain tissue extract. The baby's ^{1}H-MR spectrum is compared with a spectrum from a body with a PMI of about 520 hours. Since changes at that post mortem period are very slow, the two PMI are considered sufficiently similar.

Results: Bacterial analysis revealed a completely sterile brain tissue sample. The in situ 1H-spectrum of the baby brain is shown in Fig. 1b) whereas Fig. 1a) gives an example of a spectrum from a body with comparable PMI. Most obvious difference is the absence of the singlets from free trimethlamonium (fTMA at 2.9ppm, not to be mixed up with TMA compounds at 3.22ppm), succinate (Suc) and acetate (Ace), which are labeled with arrows. Besides fTMA and Suc our interest was also focused on butyrate (But), isobutyrate (Ibut) and propionate (Prop), which are also supposed to be of bacterial origin. The evaluation of all 32 human bodies revealed that at least three of these metabolites are detected if the PMI is longer than 121 hours (13 cases). The spectrum of the baby represents a striking exception because fTMA, Suc and Prop do not appear at all in the LC-Model results and But (2.52 ± 0.54 mmol/kgww) and Ibut (0.33 ± 0.22 mmol/kgww) are elevated only slightly above in vivo levels after 897 hours postmortem.

Discussion: The ^{1}H-MR spectrum of this baby represents a remarkable exception among all examined 32 human cases, since several expected metabolites are either not detectable in significant amounts (Suc, fTMA, Prop) or not significantly above in vivo levels (But, Ibut) at this very long PMI. As the brain tissue in this exceptional case turned out to be sterile, it seems to be a safe assumption that no bacterial strain ever populated this brain. This assumption is also supported by the fact that only a tiny amount of acetic acid (1.55 ± 0.2 mmol/kgww) is detected in the spectrum (see Fig.1b), acetate being an accepted bacterial marker. Therefore, it can be speculated that the absent metabolites (Suc, fTMA, and Prop, and most likely also But and Ibut) are of bacterial origin if observed in postmortem brain tissue.

Conclusion: The comparison of a spectrum from a baby brain with a PMI of 897 hours with other postmortem brain spectra revealed significant differences. It is concluded that fTMA, Suc, Prop, But, and Ibut can be considered as marker for bacterial growth in postmortem brain tissue. This distinction will help separating autolytic and bacterial metabolic products, which are expected to respond differently to variations of ambient temperature.

References: [1] Ith M. et al., *Magn. Reson. Med.*, 48: 915-920, 2002. [2] Scheurer E. et al., Proc ISMRM 11: 2002.