Postmortal 31P magnetic resonance spectroscopy of the skeletal muscle – PCr/ATP ratio as a forensic tool?

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Introduction

In recent years, the diagnostic radiology has been playing an important role in the forensic medicine. Magnetic resonance imaging (MRI) was introduced in the forensic medicine as a second line tool, especially for identifying soft tissue injuries. Since the numbers of autopsies decreased in recent times, radiological imaging could play a significant role in the forensic medicine by adding important information. Phosphor magnetic resonance spectroscopy (31P MRS) is an established method for metabolic examinations of resting and exercising skeletal muscle. So far, there are no MRS investigations of corpses. The aim of this study was to investigate the temporal pattern of Adenosine triphosphate and Phosphocreatine (ATP, PCr) and the PCr/ATP ratio in the Adductor magnus muscle post mortem and to check the value of MRS as a forensic tool especially for the determination of the time of death.

Material and Methods

The study protocol was approved by the local Ethics Committee, and informed consent was obtained from all relatives of the dead. Twenty-one corpses, died of natural cause (myocardial infarction, pneumonia, sepsis, malignant tumours), were examined over a period of 2 to 23 h post mortem (13 male, 8 female; mean age: 70.5 ± 8.7 y, range: 51 – 85 y, mean body weight: 74 ± 18 kg). The corpses were not matched concerning the gender and they were stored at room temperature until the scanning. The core temperature was rectally measured throughout the MRI examination. MRI scans were started not later than 6 h post mortem and performed with scan intervals of one hour over a period from 2 to 23 h post mortem. To check the comparability of the values a control group of 3 male subjects (mean age: 38.7 ± 24.5 y, range: 24 – 67 y, mean body weight: 81 ± 17 kg) was examined at a single time point as well. The MRS was performed on a 1.5 T whole body MR scanner (Magnetom Symphony; Siemens Medical Solutions, Erlangen, Germany) by using a standard 31P surface coil which was placed under the thigh of the corpses and that permits the receipt of 31P resonances at 35 Hz. For the localization of the Adductor magnus muscle transverse, sagittal and coronary localizers were performed. A free induction decay sequence with a repetition time of 700 ms, an echo time of 0.35 ms, a flip angle of 90° and 256 averages was used. The acquired spectra were interpreted using the Siemens based software (Syngo MR A30A, Siemens Medical Solutions, Erlangen, Germany). The area under the curve for each peak, as a parameter for the metabolite concentration, was determined using integration and was given as nondimensional value. Furthermore relative metabolite concentrations (PCr/ATP ratio) were calculated. In this study γ-ATP was used for the quantification of ATP because the β-ATP peak could not be interpreted. Parametric statistics (arithmetic mean value ± standard deviation (SD), standard error of the mean (SEM)) was used throughout this work.

Results

The comparison of ex vivo and in vivo spectra of the Adductor magnus muscle showed characteristic differences. The ex vivo peaks showed different chemical shifts (PCr 0 ppm, γ-ATP -7.5 ppm, α-ATP -12.5 ppm, β-ATP out of the obtained spectra) compared to the in vivo spectra (PCr 0 ppm, γ-ATP -2.5 ppm, α-ATP -7.5 ppm, β-ATP -16.5 ppm). Obviously, there was a right-hand shift of the peaks of 5 ppm which was caused by the pH decrease of the corpses after death. That is why the β-ATP resonance could not be quantified. We obtained an in vivo PCr/β-ATP ratio of 4.0 ± 1.4 and an in vivo PCr/γ-ATP ratio of 6.2 ± 2.1. At the earliest ex vivo measurement (2 h p.m.), the PCr/γ-ATP ratio (6.3) was nearly the same but it increased continuously during the following hours. The mean core temperature decreased throughout the MRI examination from 31.6 ± 3.3 °C to 27.9 ± 2.2 °C. The mean ATP concentration (averaged over 21 corpses) decreased continuously from 81.1 (2 h p.m.) to 36.2 (8 h p.m.), see Figure 1. After a meanwhile increase of the concentration to 52.0 (16 h p.m.), ATP decreased and reached the minimum of 14.3 (23 h p.m.). During the examination period the ATP concentration decreased 82.4 %.

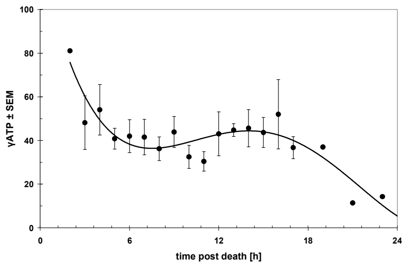

Figure 1. Postmortal course of the ATP concentration (mean ± SEM) in the Adductor magnus muscle with fitted trend line.

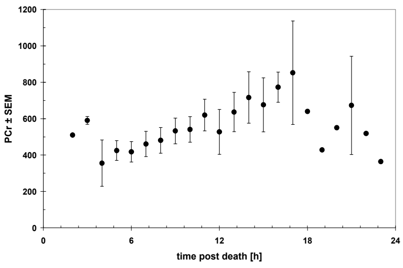

Figure 2. Postmortal course of the PCr concentration (mean ± SEM) in the Adductor magnus muscle.

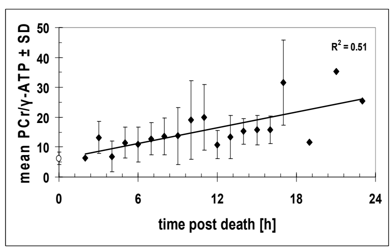

Figure 3. Postmortal course of the PCr/ATP ratio in the thigh muscle with fitted trend line and in vivo value (circle).

The PCr concentration showed a different time pattern, see Figure 2. After an initial decrease from 510.0 (2 h p.m.) to 355.3 (4 h p.m.), PCr increased and reached its maximum of 852.3 (17 h p.m.). After that maximum the PCr concentration decreased to 364 (23 h p.m.). The mean PCr/γ-ATP ratio increased nearly arithmetically from 6.3 (2 h p.m.) to 25.5 (23 h p.m.), see Figure 3. The ratio correlated with the time of death (R^2 = 0.51).

Conclusion

Especially, the acquired PCr/γ-ATP ratio showed a characteristic postmortal pattern. The ratio correlates with the time post death (R^2 = 0.51). 31P MRS could be an efficient technique that can be added to the methods of virtual autopsy (virtopsy) and the established forensic methods for the determination of the time of death, respectively.

Key words: Forensic Medicine, MRI, DWI