Interdependence of NAA and high energy phosphates in human brain

J. W. Pan[1], K. Takahashi[1]
[1]Neurology, Albert Einstein College of Medicine, Bronx, NY, United States

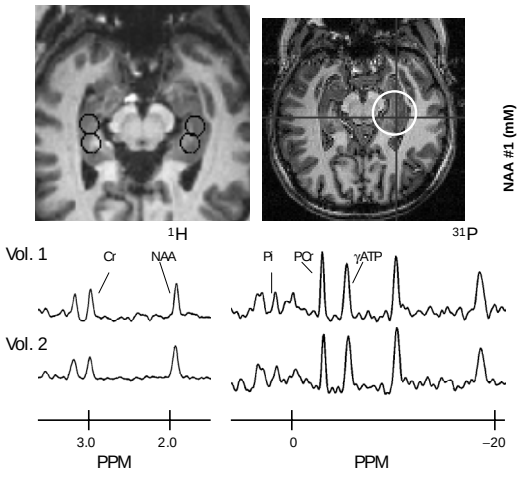
Introduction: Both NAA, known to be synthesized in neuronal mitochondria, and high energy phosphates are believed to be sensitive to energetic state. Using an *in vitro* model Patel (1) reported that NAA efflux in rat brain mitochondria was correlated to ADP. However, the relationship between NAA and high energy phosphates has not been evaluated *in vivo*.

Methods: N=15 healthy adult volunteers (27.6±7.4years) were studied. For each volunteer, two MR studies (^{1}H, ^{31}P) were performed within 2days of each other at approximately the same time of day, using a 4T Varian Inova system and volume TEM head coils. The ^{1}H spectroscopic imaging used an adiabatic refocused 3D localized sequence (2), TE/TR 72/2000, 24x24 spatial encoding over a FOV of 19.2x19.2cm and a 10mm slice (20min). ^{31}P spectroscopic imaging was performed using a pulse acquire acquisition and a three-dimensional spherical sampling scheme (13x13x13, FOV = 24x24x24cm). Including scout imaging and calibrations, the durations of the ^{1}H and ^{31}P studies were ~65 and 75min respectively.

The ^{1}H and ^{31}P data were analyzed using single voxel reconstructions (SVR) with voxel loci referenced to the midbrain aqueduct (Fig. 1). The reproducibility of this acquisition has been previously determined to have a CV of 9% (2) for NAA/Cr. Although the ^{31}P data has a large voxel size (12cc), with the SVR analysis the reproducibility of the ^{31}P acquisition in the hippocampal body was 10%. Quantification of the ^{1}H spectra was performed relative to CSF in the superior cistern measured in a high resolution proton density image, and corrected for tissue volume, coil loading and relaxation. Tissue volumes were determined from semi-automated image segmentation of T1 weighted images. The ^{31}P concentrations were determined via phantom replacement. Relaxation effects were corrected using T1 values measured at 4T. We calculated the concentration of ADP from the creatine kinase equilibrium (K_{cpk} of 1.66×10^{9} M^{-1}, assuming a pH of 7.0 and free [Mg+2] of 1mM, (3)), ratio of PCr/ATP and calculated free Cr concentration.

Results: Fig. 1 displays the loci for the ^{1}H and ^{31}P studies and spectra from two volunteers (age 26, 25). In locus #1, the raw ratio of NAA/Cr was 1.35±0.16. The fractional gray matter content in this locus was relatively consistent between volunteers, at 50±6.7%. Including relaxation corrections, the mean concentrations of NAA and creatine were 9.7±1.5mM, and 9.8±1.7mM respectively. The ratio of PCr/ATP in the hippocampal body was 1.2±0.1, with concentrations of PCr and ATP at 2.9±0.4mM, 2.4±0.4mM respectively. For the group, the calculated ADP concentration in the hippocampal body was 35±8uM. The linear regression analyses between NAA from locus #1 with ADP from the hippocampal body was highly significant, with R= +0.80, p<2x10-7, a slope of 145±20 (unit of slope is mM NAA/mM ADP) (Fig. 2).

Conclusions: It is not surprising that we have seen a relation between high energy phosphates and NAA. That this relationship is with ADP, rather than ATP or PCr provides in vivo evidence demonstrating ADP's role in regulating oxidative flux. It is of interest is that this regression with ADP is *positive*. We may have anticipated that a NAA increase (and increase in mitochondrial function) would result in a decrease in ADP (increased energy charge)—in this case, NAA should negatively correlate with ADP. However if an increased ADP provokes an increase in NAA (possibly in reaction to declining energetics), a positive correlation would result.



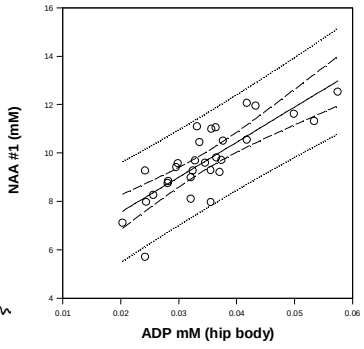


Fig. 1(left) Scout and spectra. Fig. 2(above): Regression between NAA and ADP.

These data strongly suggest a directionality in the bioenergetics-NAA relationship, where NAA and mitochondria function are responsive to energetic status and ADP, rather than ADP being responsive to mitochondrial function. This may be an in vivo manifestation of Patel and Clarke's observation that ADP levels positively relate to NAA efflux.

References: (1) Patel and Clark, Biochem J 184(3): 539-46, 1979. (2) Chu et al, ISMRM 2004; p105. (3) Veech et al J Biol Chem 254(14): 6538-47 1979. This work is in press with Annals of Neurology