MPI for Biological Cybernetics

Fortgeschrittene Anwendungen der in vivo MR Spektroskopie

6. Doktorandentraining der Deutschen Sektion der ISMRM

Bremen 2003

Josef Pfeuffer

MAX-PLANCK-GESELLSCHAFT

BIOLOGISCHE KYBERNETIK

Agenda

- 1H Spektroskopie im Rattenhirn bei 9.4T - Quantifizierung in der Frequenzdomäne (LCModel)
- Diffusionsgewichtete 1H Spektroskopie von Wasser und Metaboliten bei grossen b-Werten - technische Aspekte und Anwendungen
- Lokalisierte 1H-13C Spektroskopie in vivo - neurochemische Anwendungen

In Vivo Spectroscopy at 9.4 Tesla CMRR - Minneapolis

Rolf Gruetter
Ivan Tkac
Michael Garwood
Joseph Lin
Kamil Ugurbil

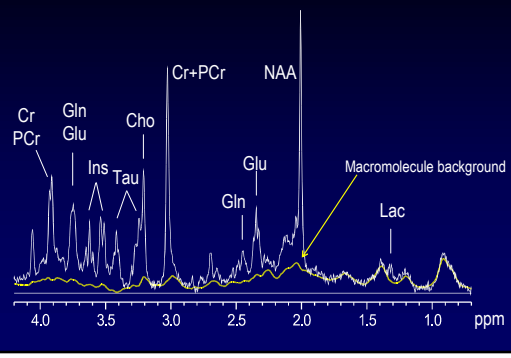
Towards an In Vivo Neurochemical Profile: Quantification of 18 Metabolites in 1H NMR Spectra of Rat Brain at TE=2 ms

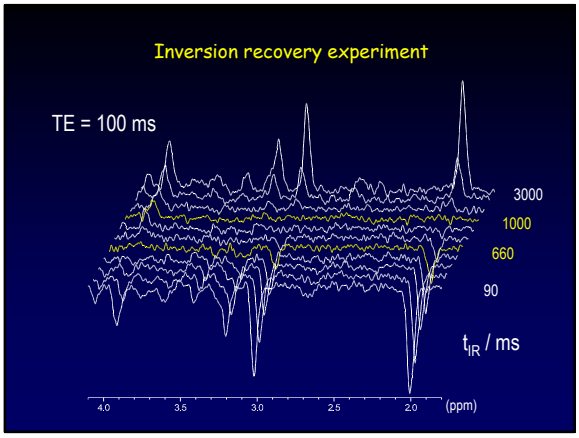
- improved sensitivity for in vivo ^{1}H MRS by the increased spectral resolution at high magnetic fields
  → detection of PCr, phosphoethanolamine (PE)
- quantitation of hitherto poorly resolved metabolites
  e.g. PCr, GABA, Gln, Tau, Glc, GSH, Ala, resting Lac
- determination of the macromolecule baseline in ultra-short echo time spectra based on T_1 differences

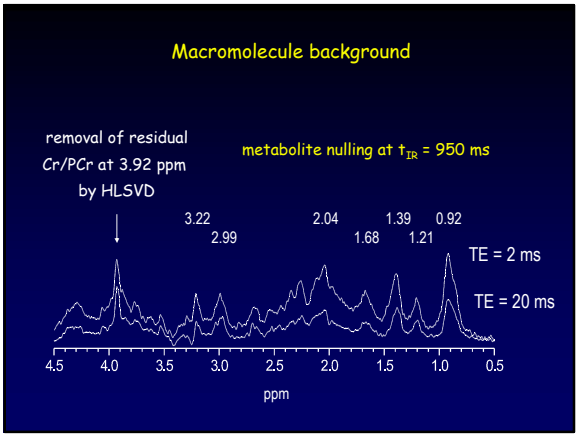
Methods

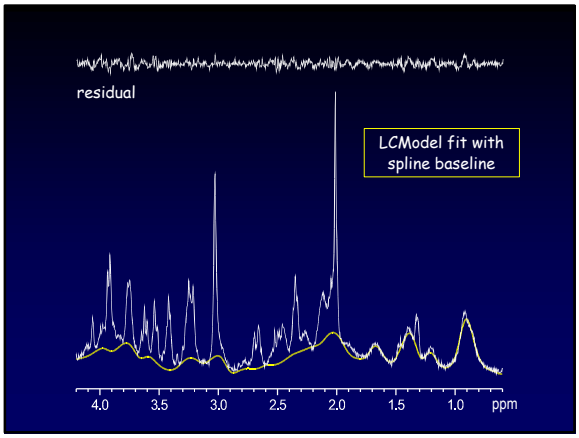
- male Sprague-Dawley rats (n > 20)
- 9.4 Tesla / 31 cm Magnex / Varian INOVA system
  quadrature ^{1}H surface coil
- STEAM (64 µL voxel) with VAPOR (7 optimized water suppression pulses) and OVS (Tkac et al., 1999)
- TE = 2 ms, TM = 20 ms, TR = 6 sec
- FASTMAP shimming (1st and 2nd order)
  → singlet linewidth of metabolites 0.02 ppm

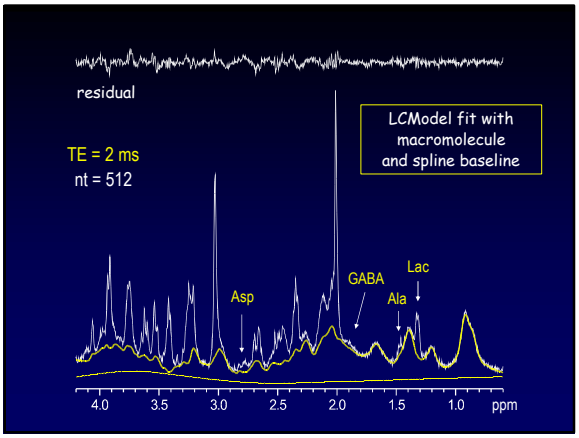
1H NMR spectroscopy in rat brain at 9.4 T

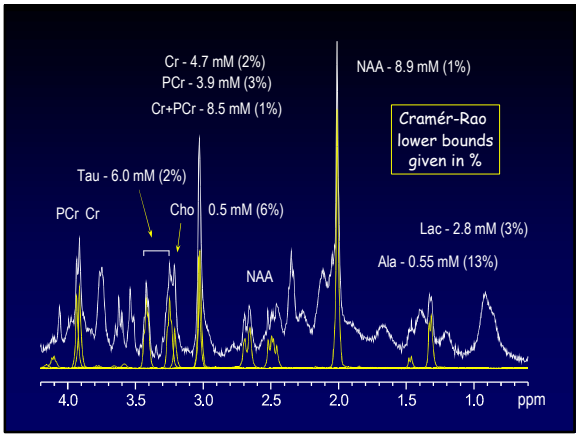


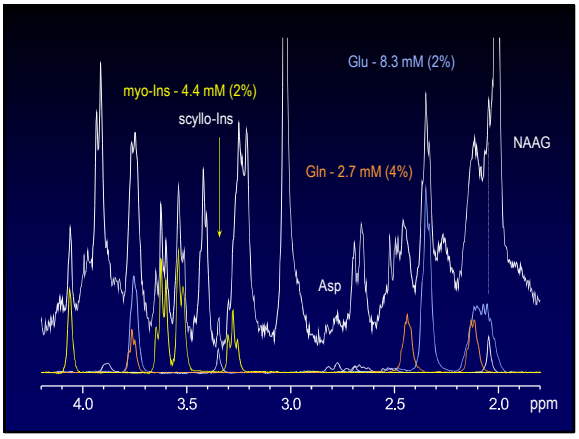

Inversion recovery experiment
TE = 100 ms
3000
1000
660
90
t_{IR} / ms
4.0
3.5
3.0
2.5
2.0
(ppm)

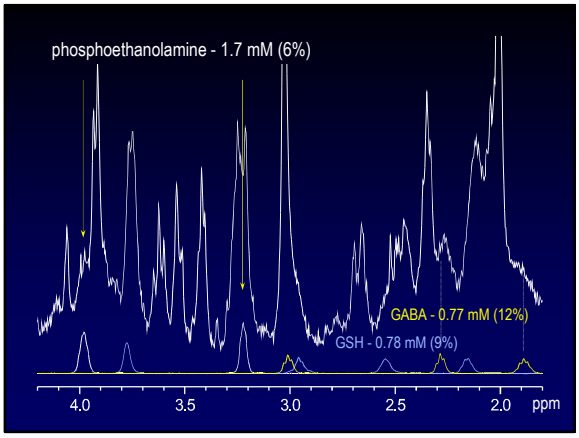

Macromolecule background
removal of residual
Cr/PCr at 3.92 ppm
by HLSVD
metabolite nulling at t_{IR} = 950 ms
3.22
2.99
2.04
1.68
1.39
1.21
0.92
TE = 2 ms
TE = 20 ms
4.5
4.0
3.5
3.0
2.5
2.0
1.5
1.0
0.5
ppm

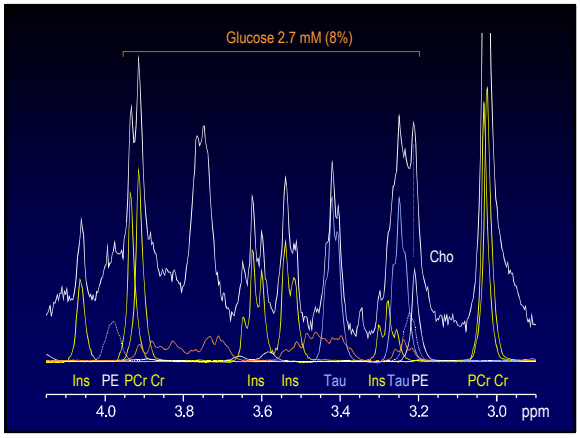

residual
LCModel fit with
spline baseline
4.0
3.5
3.0
2.5
2.0
1.5
1.0
ppm


residual
LCModel fit with
macromolecule
and spline baseline
TE = 2 ms
nt = 512
Asp
GABA
Ala
Lac
4.0
3.5
3.0
2.5
2.0
1.5
1.0
ppm


Cr - 4.7 mM (2%)
PCr - 3.9 mM (3%)
Cr+PCr - 8.5 mM (1%)
NAA - 8.9 mM (1%)
Cramér-Rao
lower bounds
given in %
Tau - 6.0 mM (2%)
PCr Cr
Cho
0.5 mM (6%)
NAA
Lac - 2.8 mM (3%)
Ala - 0.55 mM (13%)
4.0
3.5
3.0
2.5
2.0
1.5
1.0
ppm


myo-Ins - 4.4 mM (2%)
scyllo-Ins
Glu - 8.3 mM (2%)
NAAG
Gln - 2.7 mM (4%)
Asp
4.0
3.5
3.0
2.5
2.0
ppm





Summary

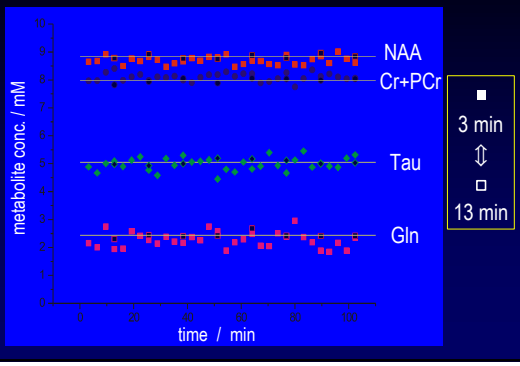
- Cramér-Rao lower bounds

CR < 4%	NAA, Cr, PCr, Tau, myo-Ins, Glu, Gln, Lac
CR < 13%	GPC+PC, PE, Glc, GSH, GABA, NAAG, Ala
CR < 22%	scyllo-Ins, Asp

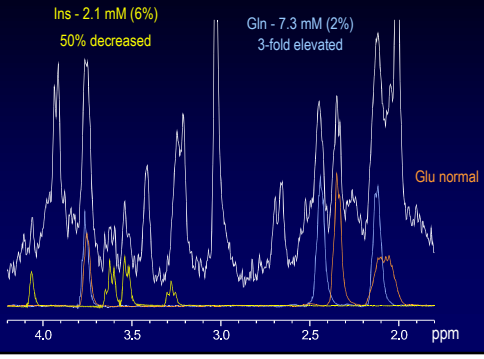
- NAA / (Cr+PCr) ratio = 1.05
- choline concentration very low 0.5 mM (6%)

(GPC+PC) / (Cr+PCr) ratio = 0.06

Reproducibility of 3-min spectra (64 mL)



Hepatic encephalopathy



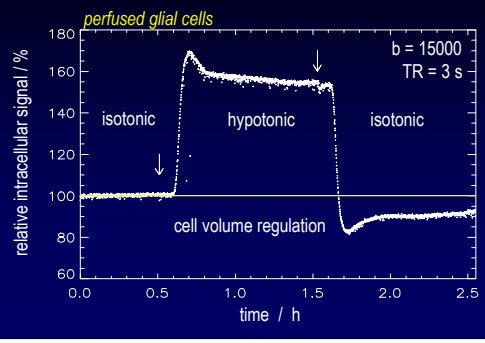
Conclusions

- neurochemical profile of 18 metabolites in rat brain, most with Cramér-Rao lower bounds < 10%
- concentrations of phosphocreatine, phosphoethanolamine, γ-aminobutyric acid, glutathione, and alanine from in vivo ^{1}H NMR spectra without any editing
- maximum of spectral information possible with ultra-short echo times due to high sensitivity and excellent shimming

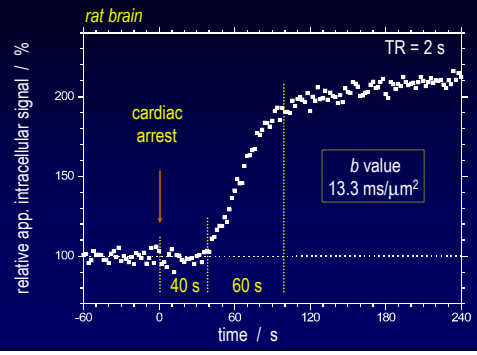
Diffusion-Weighted Spectroscopy of Water and Cerebral Metabolites at Very Large b Values – What Multiple Components Tell us

- Why do we need experiments at large diffusion weighting ?
- What about modeling a multi-exponential decay ?
  compartments vs. components
- Diffusion-weighted ^{1}H spectroscopy of cerebral metabolites:
  intracellular and extracellular contribution
- Diffusion-weighted ^{13}C spectroscopy:
  metabolically active tracer (glucose → lactate)

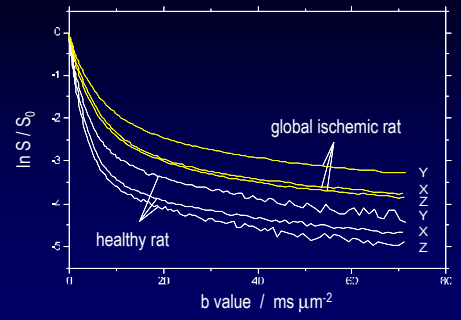
Monitoring of intracellular water at large b



Dynamic changes of the water signal at large b

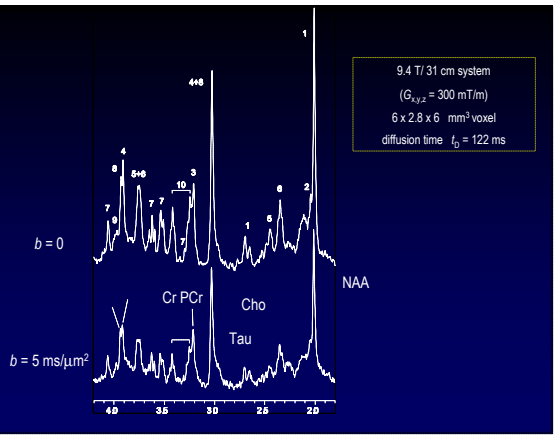


Multiexponential decay in rat brain tissue

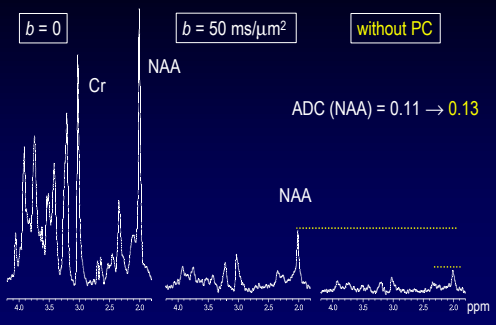


Diffusion-weighted 1H spectroscopy of cerebral metabolites

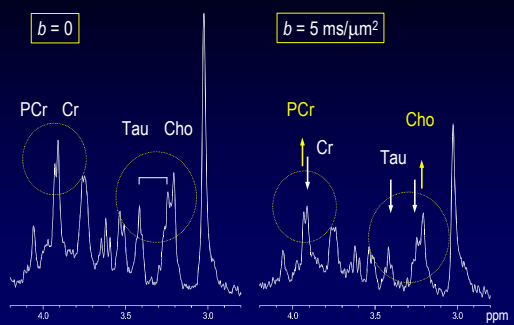
- metabolites in brain are mostly intracellular ↔ glucose and lactate have substantial extracellular contribution
- assumption that glucose in brain is equally distributed in the extra- and intracellular space (in vitro studies)
- potential concentration gradients at the extra- / intracellular interface in vivo ?
- diffusion-weighted ^{1}H NMR spectroscopy has provided methods to separate intracellular contributions → gain indirect knowledge of extracellular fraction



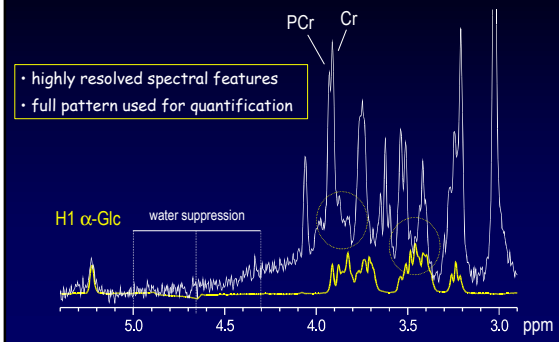
Effect of single trace phase correction



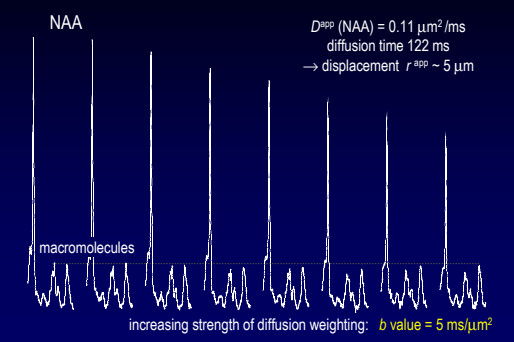
Fine details in diffusion attenuation



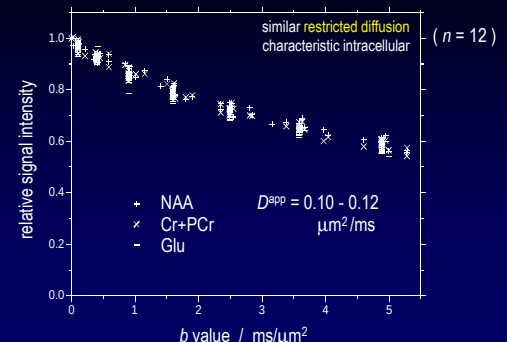
Quantification of glucose



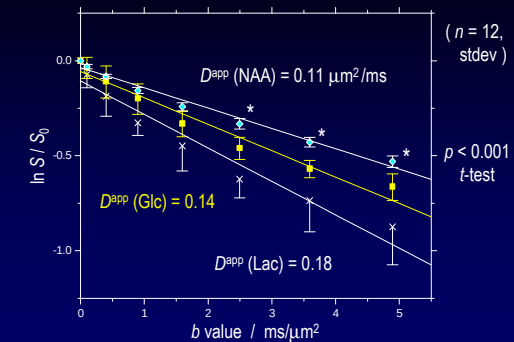
Diffusion weighting



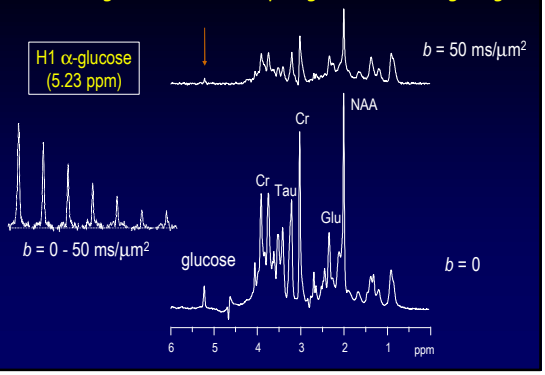
Signals of intracellular metabolites are NOT discernible



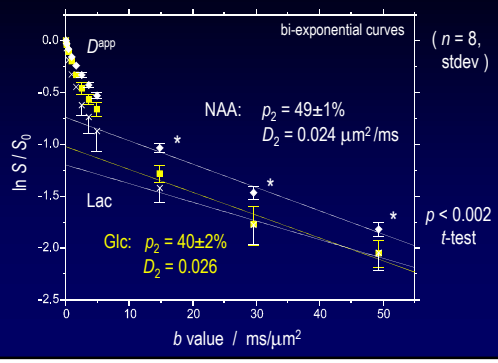
Apparent diffusion coefficient of glucose and lactate



1H NMR signals in vivo at very large diffusion weighting



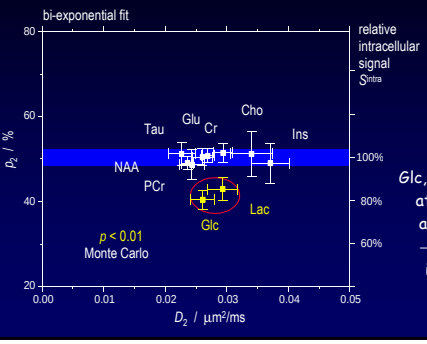
Glc, Lac: decreased fraction p2 at large b values



Estimation of the extracellular fraction

- bi-exponential model - two intracellular components p_1, p_2
  → separable component p_2 at large b values,
  e.g., NAA as internal standard: $p_1 : p_2 \sim 1 : 1$
- glucose is known to have extracellular contribution
  → calculation from p_2 and ratio $p_1 : p_2$,
  e.g., Glc: $p_2 = 40\% = p_1$ → $p^{extra} = 1 - p_1 - p_2$
- extracellular fraction p^{extra} (Glc) = 19±5% is consistent with equally distributed glucose in brain

Volume fraction p2 vs. diffusion coefficient D2



Glc, Lac detectable at very large b and similar D_2 → substantial intracellular fraction

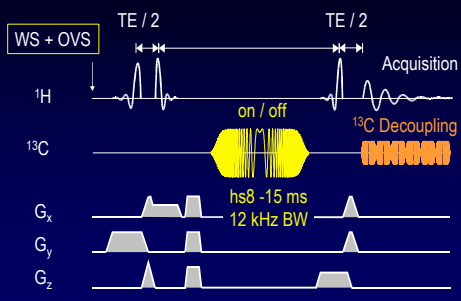
Localized in vivo 1H NMR detection of neurotransmitter labeling during infusion of [1-13C] D-glucose

- Indirect ^1H detection of ^{13}C label, e.g. POCE, HSQC
  → more sensitive, but spectral resolution reduced
  - is detection of ^{13}C enrichment in resonances like Gln, GABA, Lac, and Ala possible ?
  - resolve Gln from Glu resonances?
- broadband ^{13}C decoupling in vivo at 9.4 Tesla
- simultaneous ^1H and ^{13}C information:
  → concentrations and ^{13}C fractional enrichment

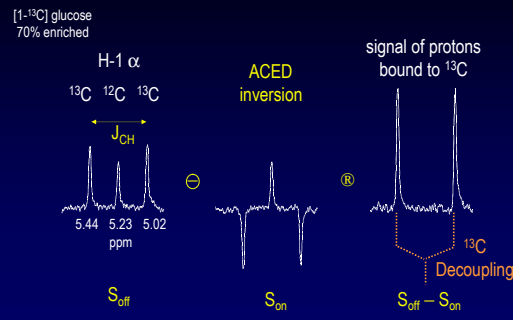
Methods

- ^1H quadrature / ^{13}C linear triple-turn surface coil
- male SD rats → [1-^{13}C] glucose infusion (16 mg/min)
- localization based on STEAM with VAPOR water suppression and OVS (Tkac et al., MRM 41, 649, 1999)
- localization 7 x 2.8 x 7 mm³ voxel (137 μL)
- TE = $1/J_{CH}$ = 7.9 and 5.9 ms, TR = 4 s

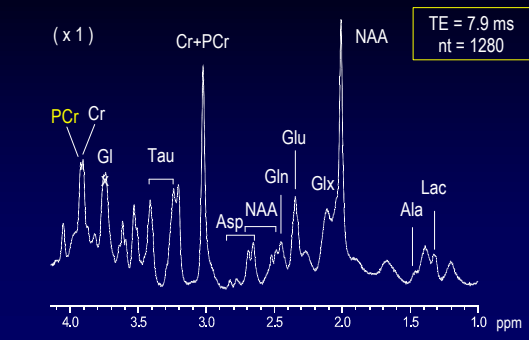
ACED - Adiabatic Carbon Editing and Decoupling



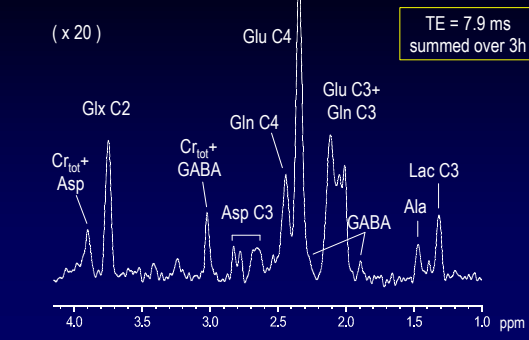
ACED - Editing process



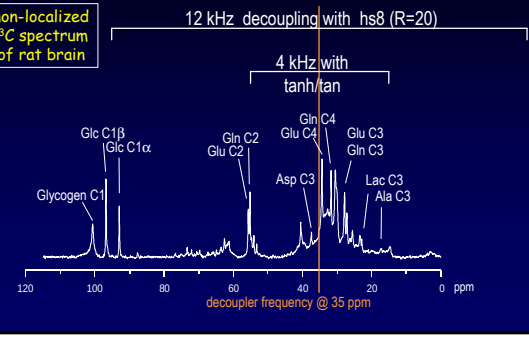
ACED subspectrum - S_{off}



ACED $S_{off} - S_{on}$

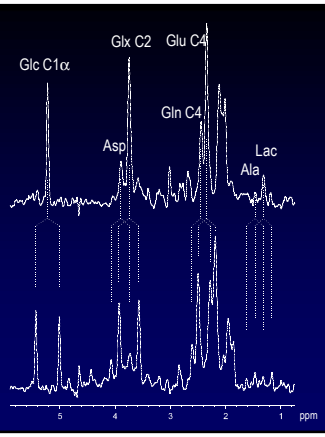


Direct ^{13}C detection



ACED - Broadband Decoupling

- decoupling applied during 400 ms acquisition
- adiabatic AFP pulses on carbon channel:
  [tanh / tan, R = 54] - 1.5 ms
  → 4 kHz decoupling width (40 ppm)
  [hs8, R = 20] - 1.8 ms
  → 12 kHz decoupling width (120 ppm)
- five-step phase cycle combined with MLEV-4
- γB_2 ~ 1 kHz, peak decoupler power below 2 W



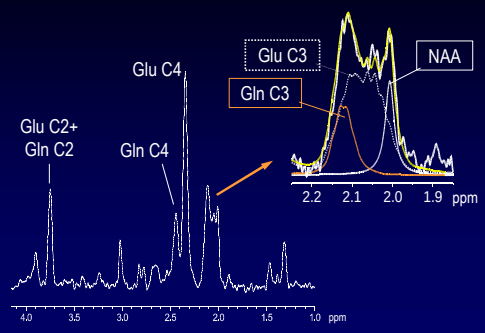
Adiabatic ^{13}C broadband decoupling in vivo

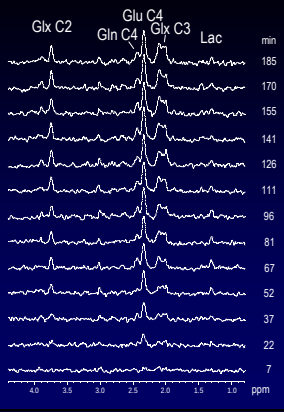
with/ without hs8 decoupling
doffset(^{13}C) @ 35 ppm
BW = 12 kHz / 120 ppm

TE = 6 ms / J_{CH}(Glc)
30 min interleaved acquisition
225 scans each

Ala, Lac doublet detectable despite low concentration
Glc and Lac decoupled

ACED detection of ^{13}C label in glutamate and glutamine C3





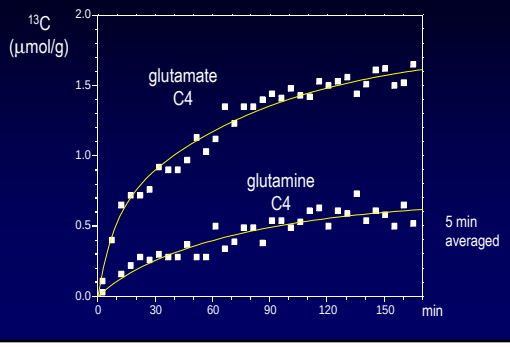
^{13}C turnover during infusion of [1-^{13}C] glucose

TE = 7.9 ms
15 min averaged
192 scans

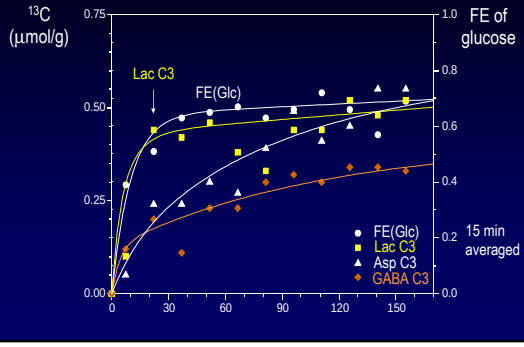
Summary

- ACED for editing with localized $^{1}H\{^{13}C\}$ detection
  → adiabatic inversion, adiabatic broadband decoupling
- very short TE possible for editing (T_2 effects, J_{HH})
- simultaneous ^{13}C label detection and total $^{1}H\{^{12}C+^{13}C\}$ metabolite concentration
- detection of separate Glu - Gln in positions C4
  → deconvolution of C3 peaks possible

Temporal evolution of ^{13}C enrichment



^{13}C labeling and fractional enrichment



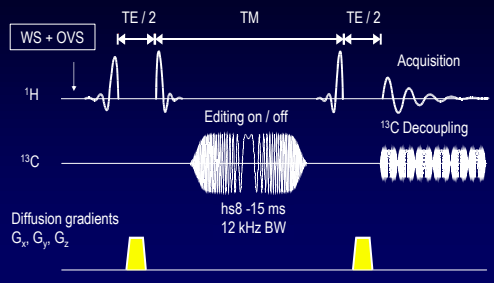
Conclusions

- high resolution and sensitivity at high magnetic fields
  → full advantage of ^{1}H methods for localization
- in vivo detection of ^{13}C labeling in multiple positions:
  → energy metabolism and neurotransmission
- ^{13}C label in glutamine, lactate, GABA, and aspartate
- simultaneous monitoring of glucose, lactate, and phosphocreatine

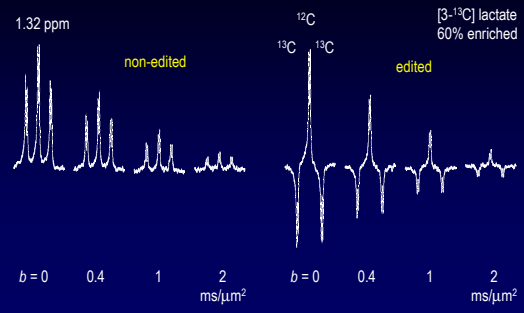
Diffusion-weighted spectroscopy of 13C-labeled lactate in rat glioma in vivo

- diffusion-weighted ^{1}H NMR spectroscopy difficult to perform in vivo due to low concentration of cerebral metabolites
- diffusion-weighted ^{13}C NMR spectroscopy only done in vitro with very long acquisitions times [Malveau C et al. JMR 1998]
- indirect ^{1}H detection of ^{13}C label is more sensitive - many technical issues (e.g. hardware, carbon decoupling)
- ^{13}C labeling provides more specific answers and eliminates unwanted signals (e.g., lipid overlap with lactate signal)

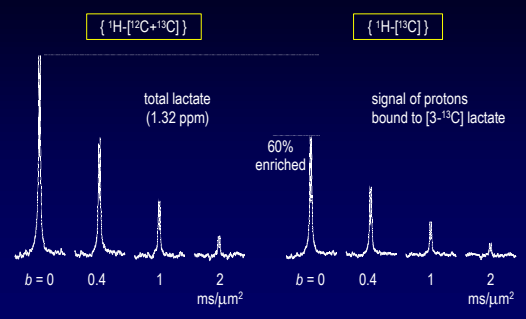
Diffusion-weighted ACED sequence



Editing of [3-13C] lactate - phantom data

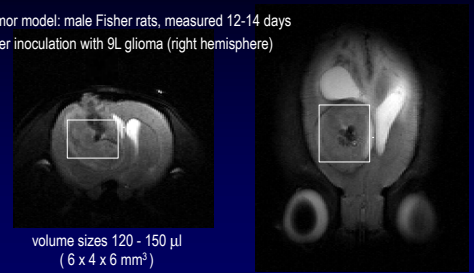


Diffusion-weighted ACED – with decoupling



Diffusion-weighted spectroscopy of 13C-labeled lactate in rat glioma in vivo

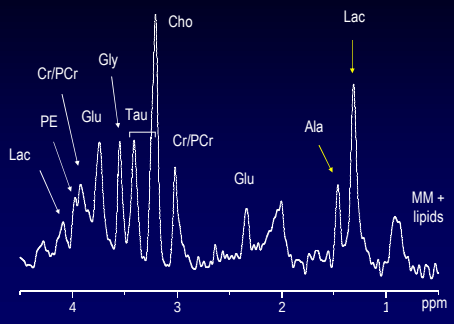
tumor model: male Fisher rats, measured 12-14 days after inoculation with 9L glioma (right hemisphere)



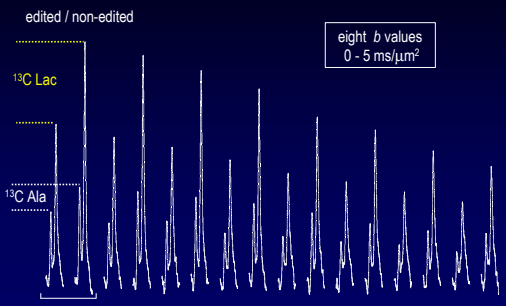
volume sizes 120 - 150 μl
(6 x 4 x 6 mm^3)

continuous infusion of 70%-enriched [1-^{13}C] glucose:
[3-^{13}C] lactate turnover into steady state after one hour

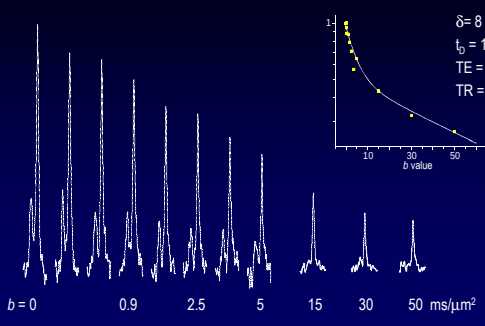
1H spectrum of 9L glioma



Diffusion-weighted series of in vivo lactate and alanine

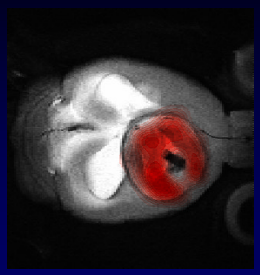


In vivo DW [3-13C] lactate and [3-13C] alanine



Features of diffusion-weighted 13C spectroscopy

- specific information by ^{13}C labeling depending on metabolism
  normal: Glc → Glu/Gln, tumor: Glc → Lac/Ala
- selectivity and localization in metabolically-active tissue
- detection of lactate without lipid contamination
- simultaneous DW ^{1}H spectral information: Cho, Cr, Glu, Gly, PE
- dynamic information → labeling time course from turnover
  → changes of metabolite ADC reflect location
  e.g., CSF, extracellular, intracellular compartment
- lactate in 9L glioma is metabolic product of glucolysis
  → significant signal from intracellular space



Spectroscopic Imaging: lactate map

9L tumor in rat (4.7T)

J.C. Lin et al., 2000

Selected Literature

- Bachelard H, Badar-Goffer R (1993) J Neurochem 61(2) 412-429
- Brand A, Leibfritz D (1997) Cell Mol Biol 43(5), 645-657
- de Graaf (1998) in vivo NMR spectroscopy, Wiley
- Gruetter R (2002) Neurochem Int 41(2-3),143-54
- Magistretti PF, Pellerin L (1999) Phil Trans R Soc Lond B 354,1155-63
- McIlwain H, Bachelard HS (1985) Biochemistry and the Central Nervous System
- Pfeuffer J et al (1999) J Magn Reson 141, 104-120; Magn Reson Med 41, 1077-83
- Ross B, Michaelis T (1994) Magn Reson Q 10(4), 191-247
- Ross B, Bluml S (1996) NMR Biomed 9, 279-296; (2001) Anat Rec 265, 54-81
- Rothman DL (1994), Ch 21, NMR in Physiology and Biomedicine
- Siegel GJ et al (1999) Basic Neurochemistry, Lippincott-Raven
- Ugurbil K et al (2000) Annu Rev Biomed Eng 2, 633-60

Questions? Reprints? josef.pfeuffer@tuebingen.mpg.de