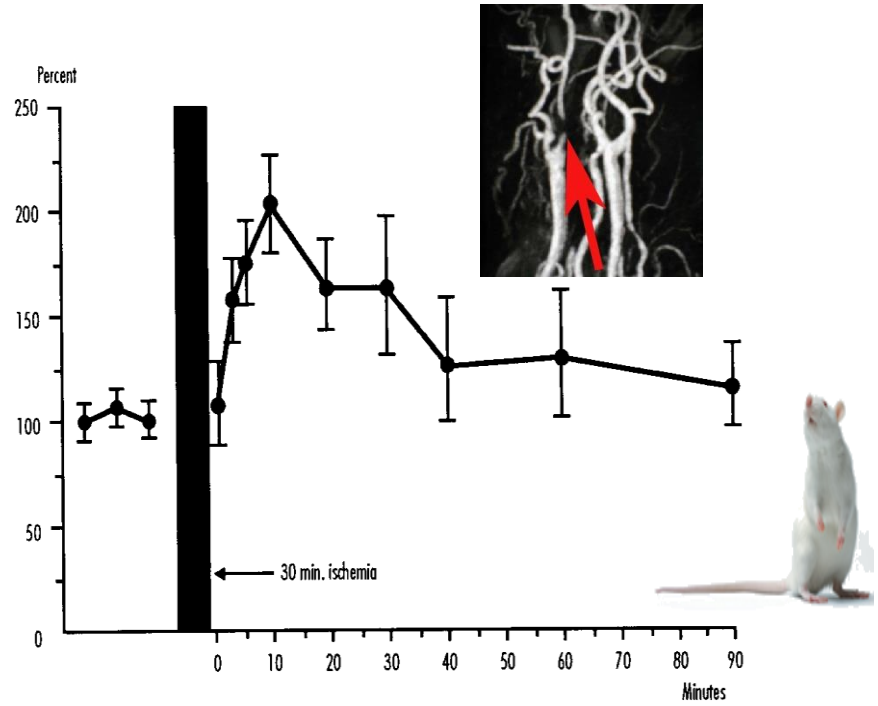
Development of trace analytical methods for biological microenvironment monitoring

Still to be Answered : What cause brain damage in acute ischemic stroke and traumatic stress?

Production of Free Radicals in Fetal Brain



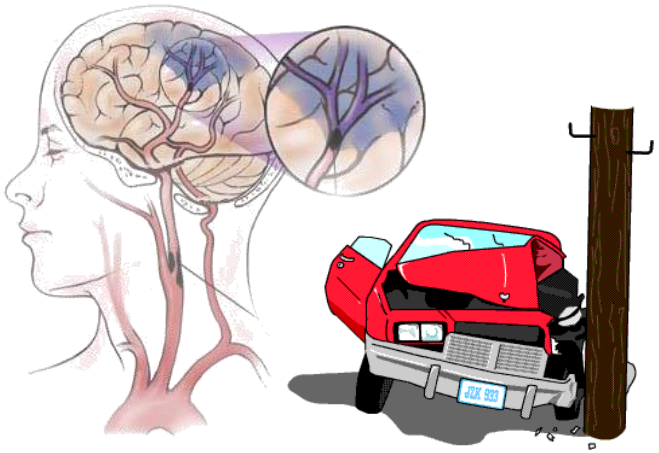
http://www.nichd.nih.gov/publications/pubs/acute/acute.cfm

Acute stress facilitates long-lasting changes in cholinergic gene expression

Daniela Kaufer*, Alon Friedman*†, Shlomo Seidman* & Hermona Soreq*

In light of the recent observation of stress-related breaks in the blood-brain barrier, the mechanism of metal ion accumulation in the brain should be re-evaluated.

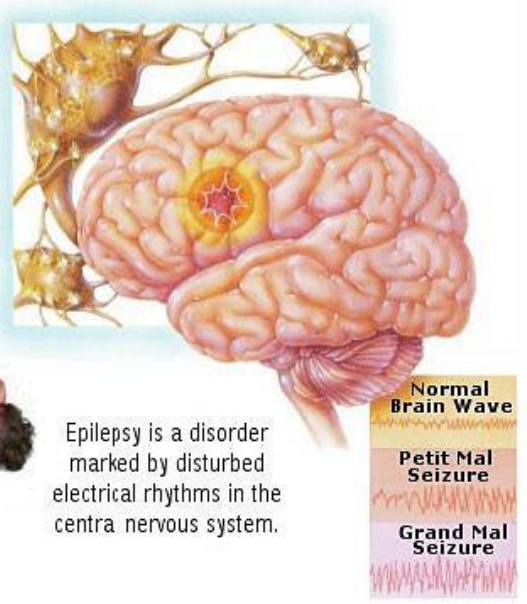
Nature, 393, 1998, 373



doi:10.1093/brain/awl318 *Brain* (2007), **130**, 521–534

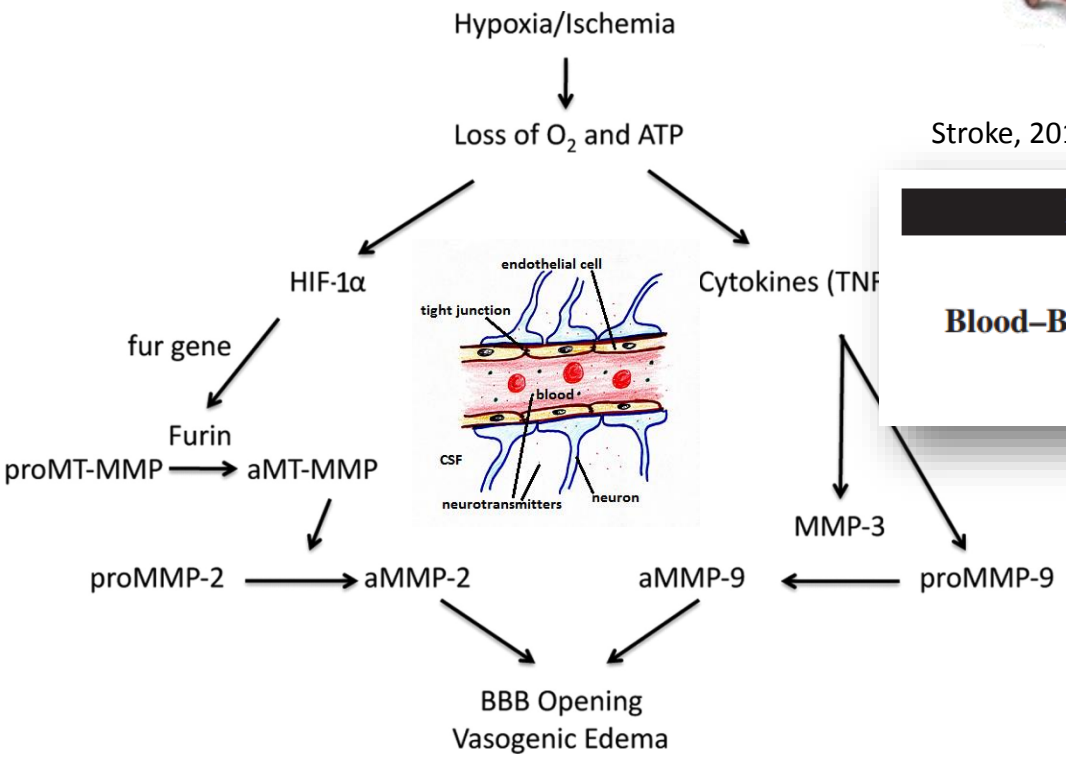
Blood–brain barrier leakage may lead to progression of temporal lobe epilepsy

E. A. van Vliet,[1,2] S. da Costa Araújo,[2] S. Redeker,[3] R. van Schaik,[2] E. Aronica[3] and J. A. Gorter[1,2]



Epilepsy is a disorder marked by disturbed electrical rhythms in the centra nervous system.

Blood–brain barrier (BBB) disruption in hypoxia/ischemia



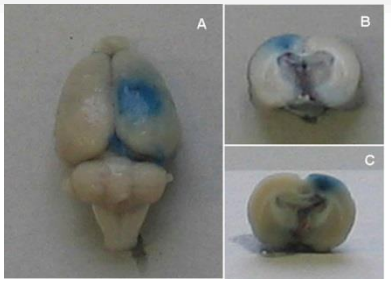
Stroke, 2011; 42: 3323-3328

Basic Science Advances for Clinicians

Section Editors: Anna Planas, PhD, and Richard Traystman, PhD

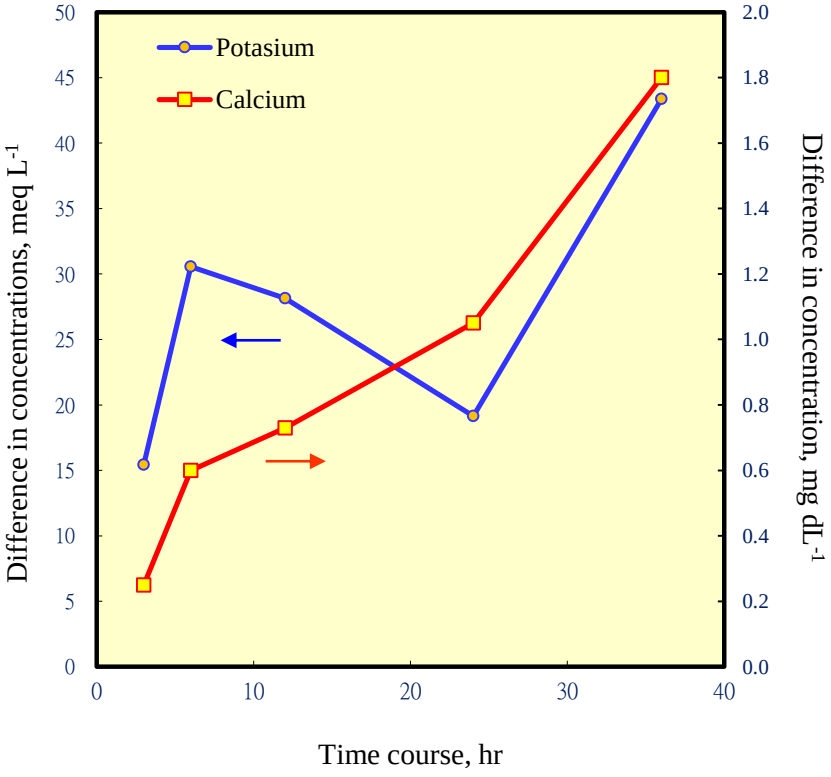
Blood–Brain Barrier Breakdown in Acute and Chronic Cerebrovascular Disease

Yi Yang, MD, PhD; Gary A. Rosenberg, MD

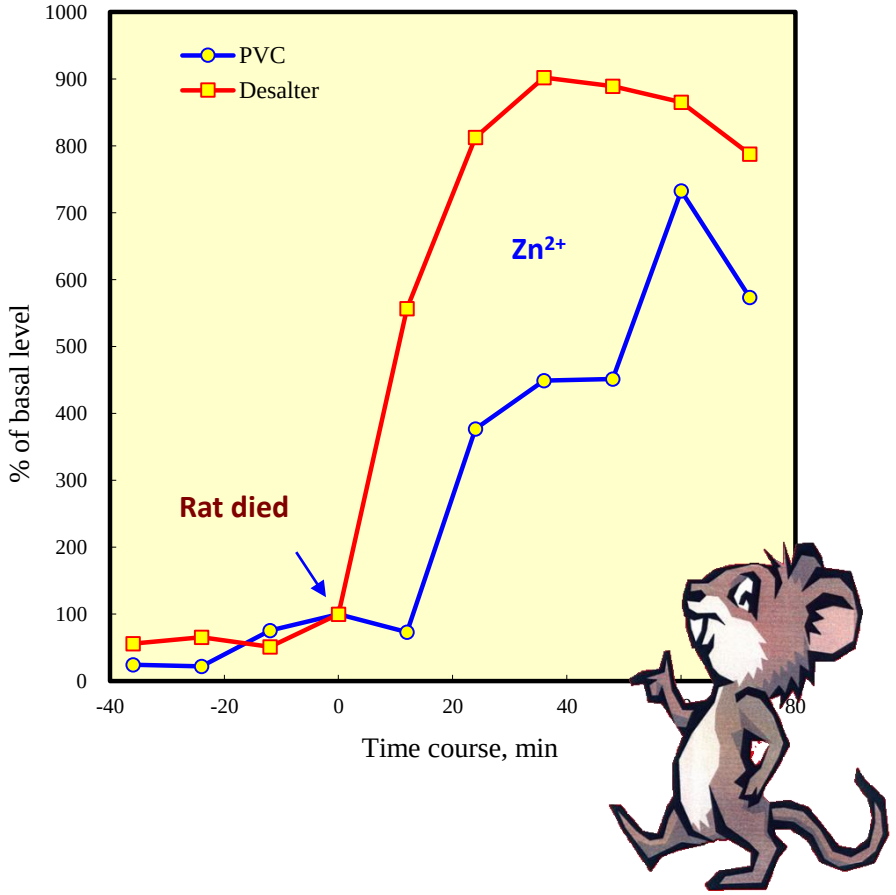


Postmortem Changes in CSF metal ions concentrations

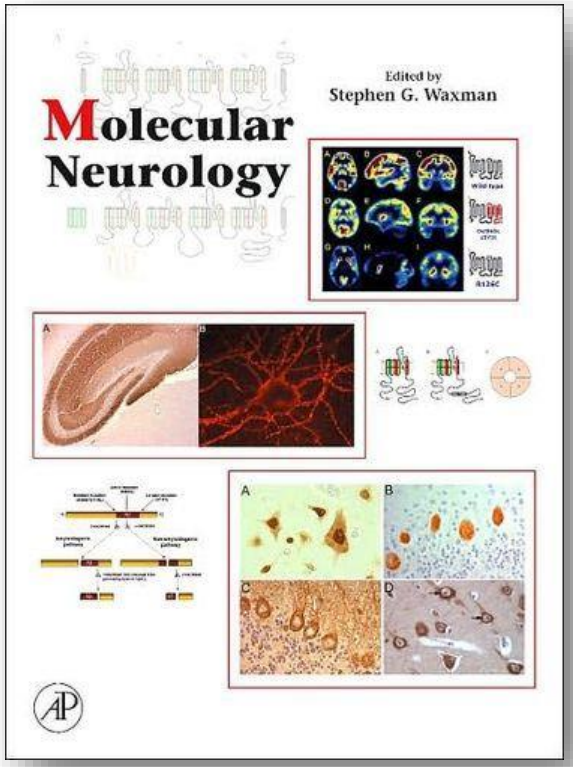
Schoning P. et al., J. Forensic Sci., 1980, 25:60-66.

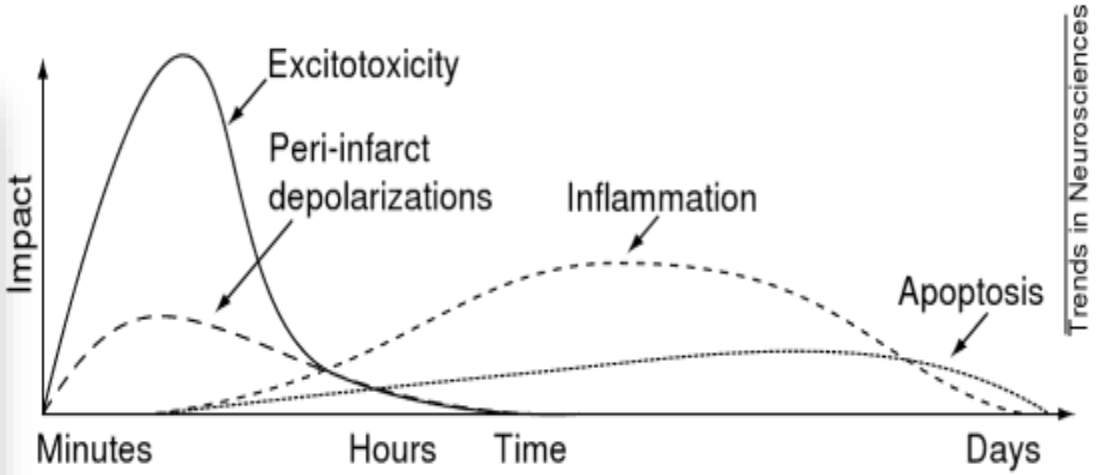


Sun Y.C. et al., J. Mass Spectrom. Rev., 2010, 29: 392-424 .



Post ischemic changes over time





Early after the onset of the focal perfusion deficit, excitotoxic mechanisms can damage neurons and glia. Later changes include peri-infarct depolarizations, inflammation, and apoptosis.

Molecular Neurology (2007) ed. S Waxman, Elsevier, Amsterdam

The Journal of Neuroscience, July 30, 2003-23(17):6847-6855

Cellular/Molecular

Evidence for Chelatable Zinc in the Extracellular Space of the Hippocampus, But Little Evidence for Synaptic Release of Zn

Alan R. Kay
Biological Sciences, University of Iowa, Iowa City, Iowa 52242

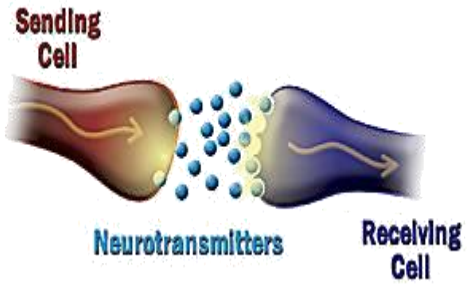
Between 10 and 30 µM

300 µM

Only 6 nM

Therefore, the concentration of Zn^{2+} released was still unclear.

The extracellular concentration of Zn^{2+} released has been estimated at between 10 and 30 μM, although Assaf *et al.* estimated it was about 300 μM under extreme conditions.[2,4,8,9] However, recently, Kay estimated it was only 6 nM, and Yang reported a decrease of extracellular Zn^{2+} in the cortex as determined by microdialysis during focal ischemia in gerbils.[10,11] Therefore, the concentration of Zn^{2+} released was still unclear. In our study, the extracellular Zn^{2+} level in hippocampal CA1 area transiently increased during ischemia, and reached a maximum of about 600 nM. This result is not consistent with the result of Yang, despite that the same method was used. Although the cause of the discrepancy is unknown, differences of animal species, ischemia model, and measurement site might be blame. In the present study, the concentration of glutamate reached about 20 times the basal level, but the concentration of Zn^{2+} was only about twice the basal level, suggesting that the amount of Zn^{2+} released during ischemia is not excessive.



Why *in-vivo* monitoring is "The worst of the worst" to Analytical Chemistry

Low analyte concentration

- 10^{-1} μg/L < $[M^{n+}]$ < 10^{2} μg/L

Limited sample volume

- Sampling rate=0.5-1 μL/min
- Sample volume=10 μL/20 min

Sensitivity

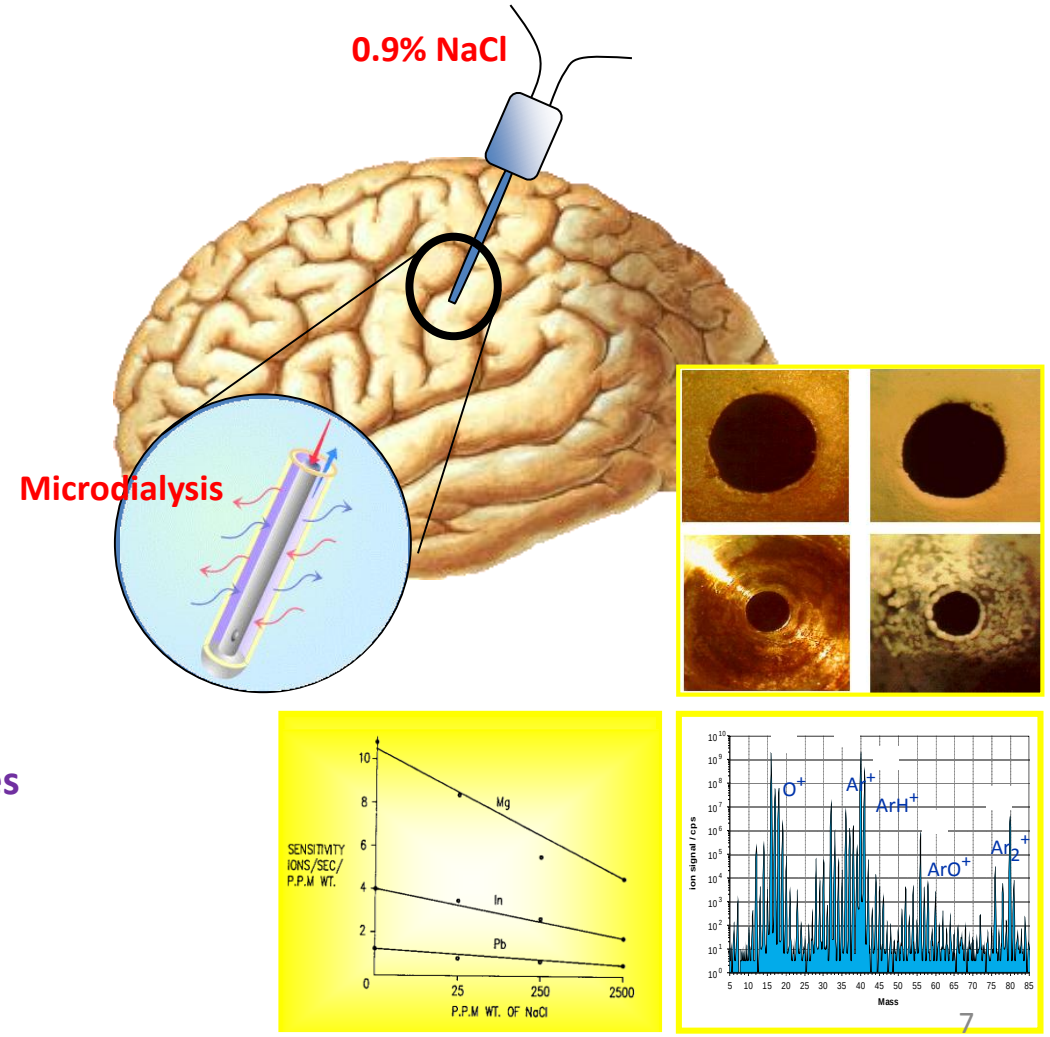
- 1 μg/L×10 μL= 10 ×10^{-12} g

Selectivity

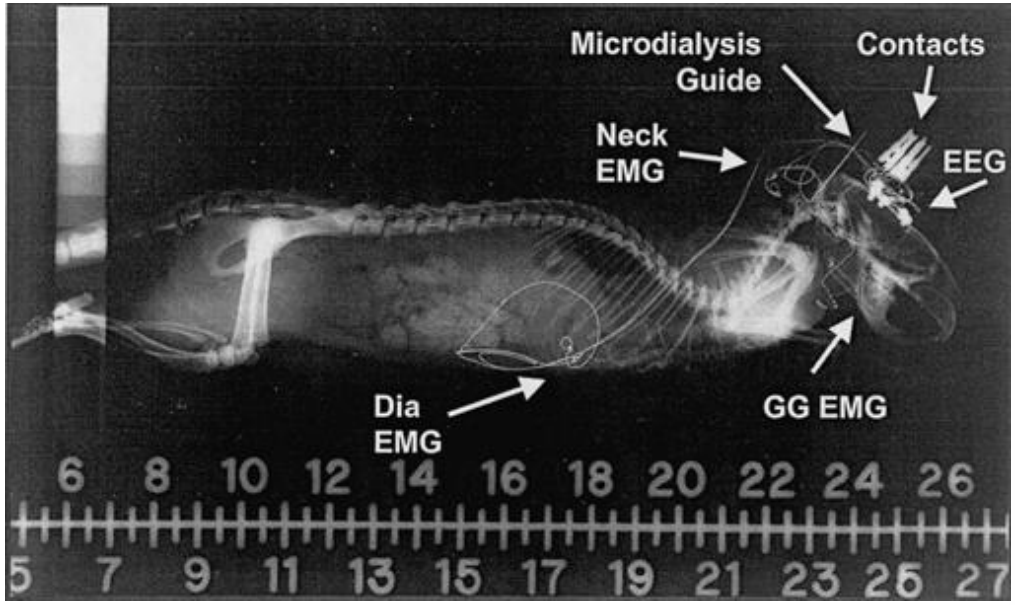
- Normal saline=0.9% NaCl
- Target anionic and cationic species

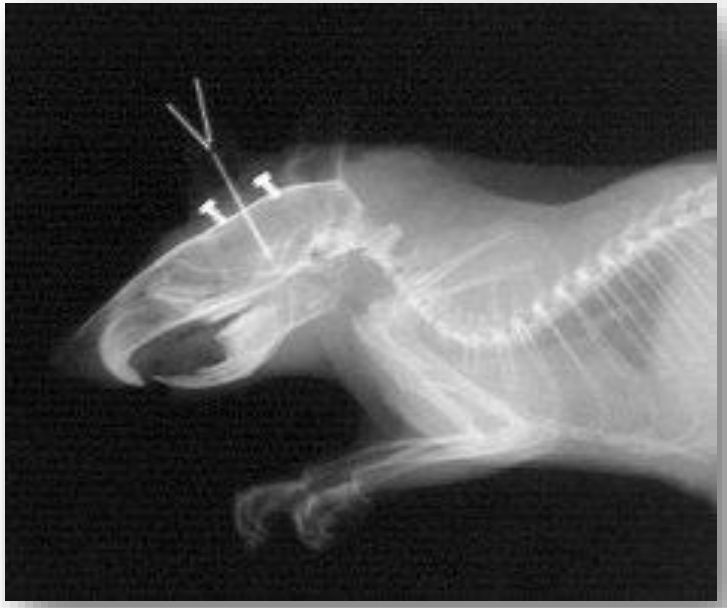
Robustness

- 12 h continuous monitoring
- Blank control



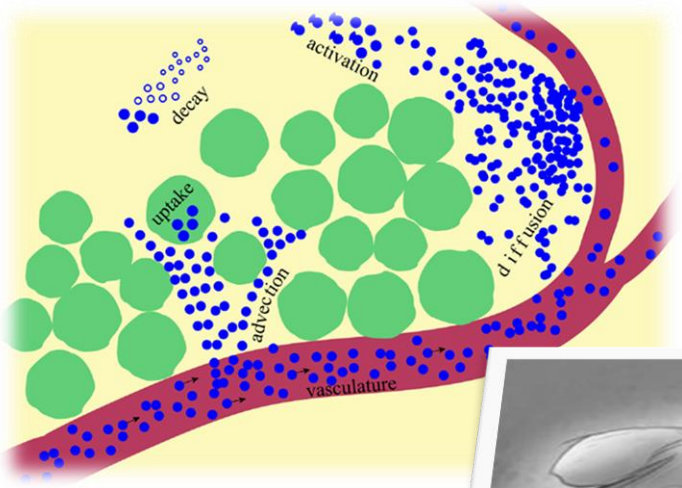
X-ray showing location of microdialysis guide cannula

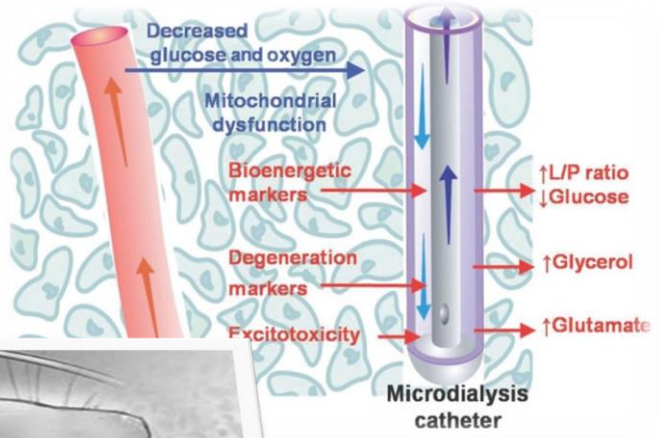


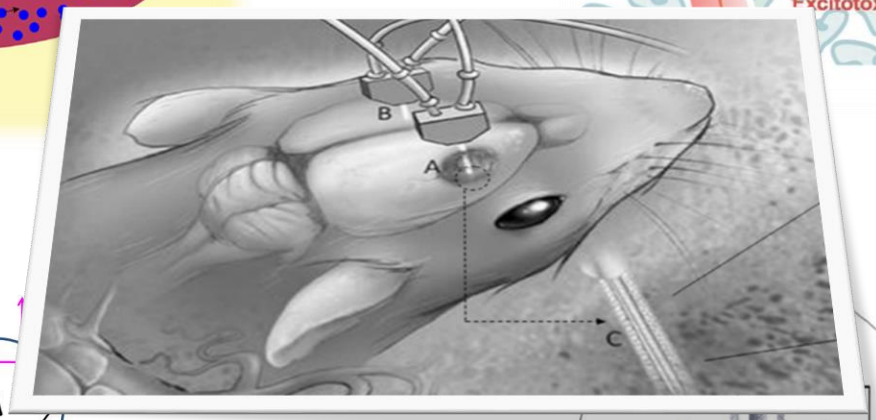


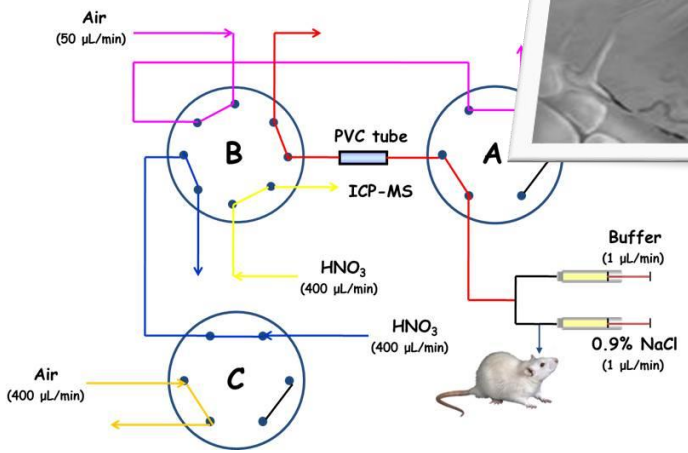
The Journal of Physiology
Volume 532, Issue 2, pages 467-481, 5 AUG 2004 DOI: 10.1111/j.1469-7793.2001.0467f.x
http://onlinelibrary.wiley.com/doi/10.1111/j.1469-7793.2001.0467f.x/full#f1

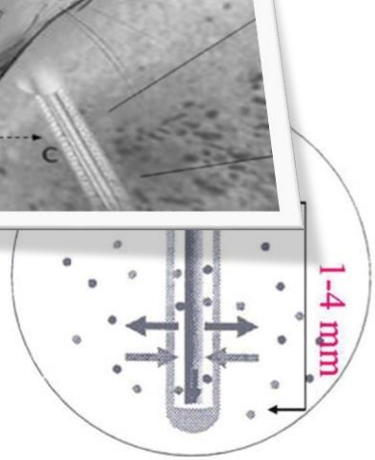
Microdialysis *in vivo* sampling technique









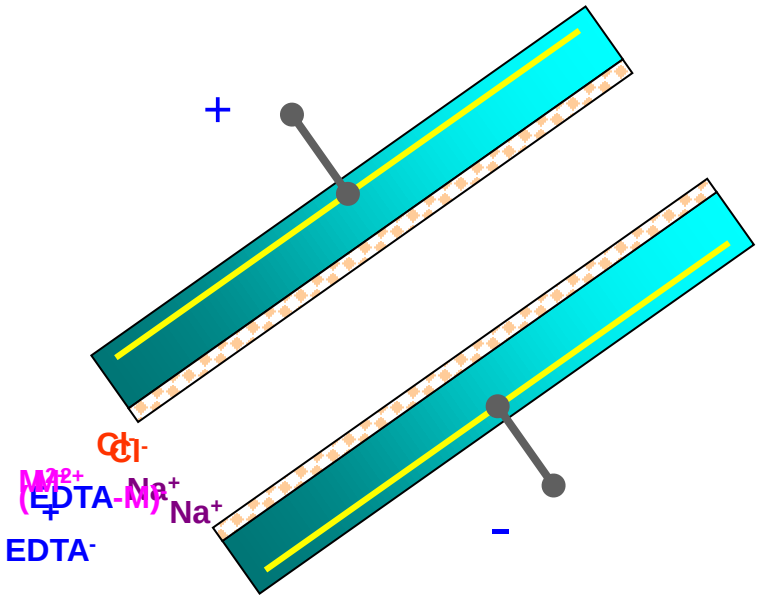


Anal. Chem. **2007,** *79,* 8900−8910

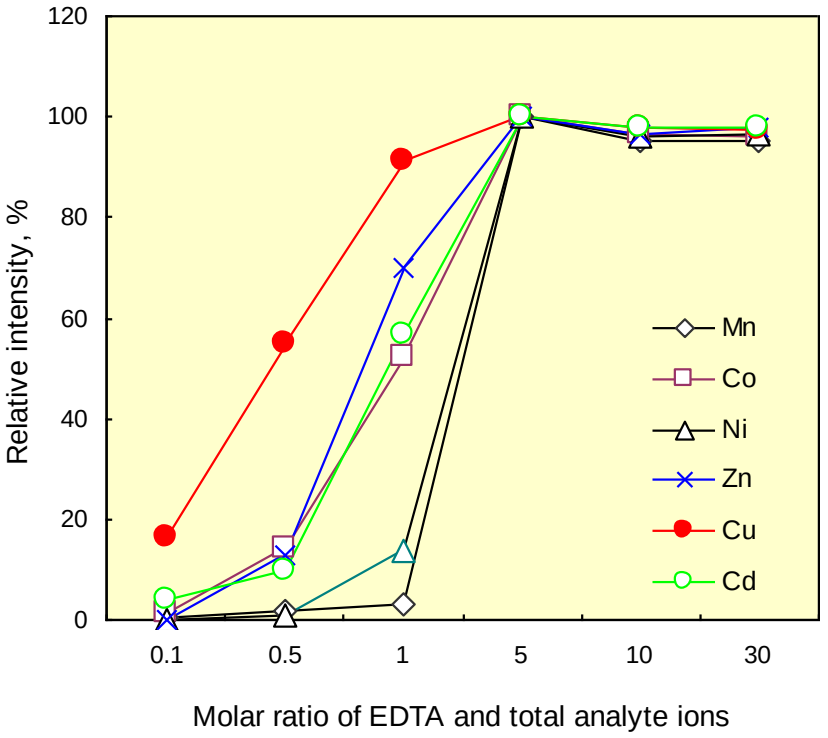
In Vivo Monitoring of Multiple Trace Metals in the Brain Extracellular Fluid of Anesthetized Rats by Microdialysis−Membrane Desalter−ICPMS

Y. T. Chung,† Y. C. Ling,† C. S. Yang,§ Y. C. Sun,*,‡ P. L. Lee,‡ C. Y. Lin,‡ C. C. Hong,‡ and M. H. Yang‡

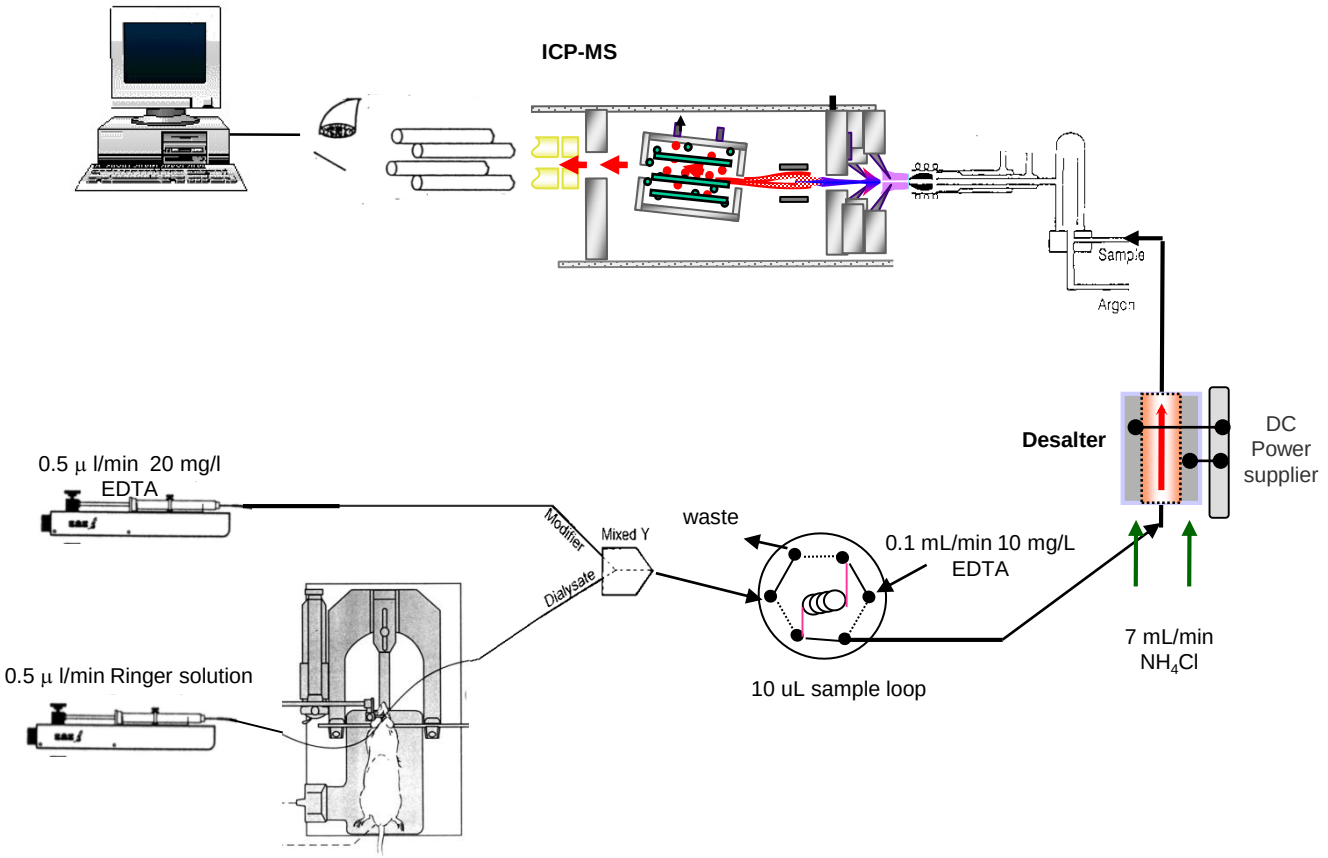
Department of Chemistry, and Department of Biomedical Engineering and Environmental Sciences, National Tsing-Hua University, Hsinchu, Taiwan, and Department of Applied Chemistry, National Chi-Nan University, Nantou, Taiwan



EDTA concentration vs. Retention efficiency



Schematic illustration of the on-line MD–desalter–ICPMS hyphenation system



Published in: Y. T. Chung; Y. C. Ling; C. S. Yang; Y. C. Sun; P. L. Lee; C. Y. Lin; C. C. Hong; M. H. Yang; *Anal. Chem.* **2007,** 79, 8900-8910.
DOI: 10.1021/ac070981z

Time course of the concentration of trace metals in the ECF of a living rat

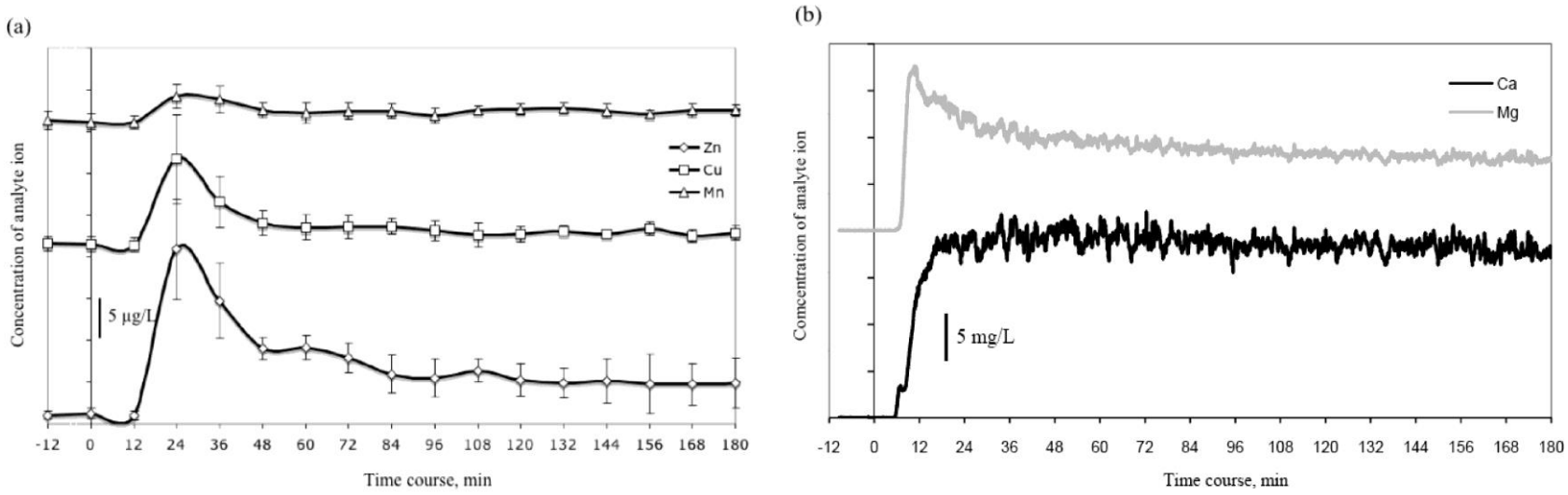


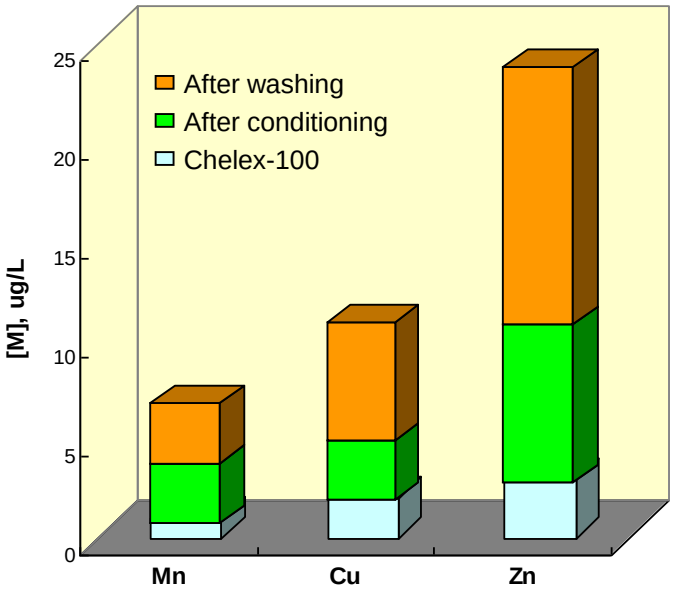
Figure 5 Time course of the concentration of trace metals in the ECF of a living rat following insertion of a microprobe into the brain: (a) Cu, Zn, and Mn with prior collection of microdialysate in a 10 μL loop; (b) Ca and Mg, real-time measurement without use of the sample loop. The error bars represent the standard deviations (n = 3). The MD probe was inserted at time zero.

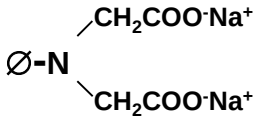
Published in: Y. T. Chung; Y. C. Ling; C. S. Yang; Y. C. Sun; P. L. Lee; C. Y. Lin; C. C. Hong; M. H. Yang; *Anal. Chem.* **2007,** 79, 8900-8910.
DOI: 10.1021/ac070981z

Anal. Chem. **2008,** *80,* 6959–6967

Online In-Tube Solid-Phase Extraction Using a Nonfunctionalized PVC Tube Coupled with ICPMS for in Vivo Monitoring of Trace Metals in Rat Brain Microdialysates

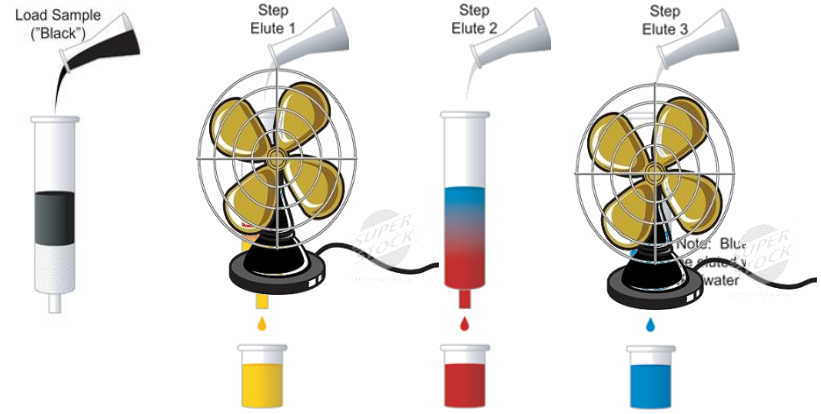
C. K. Su,[†] T. W. Li,[†] and Y. C. Sun*[,†,‡]





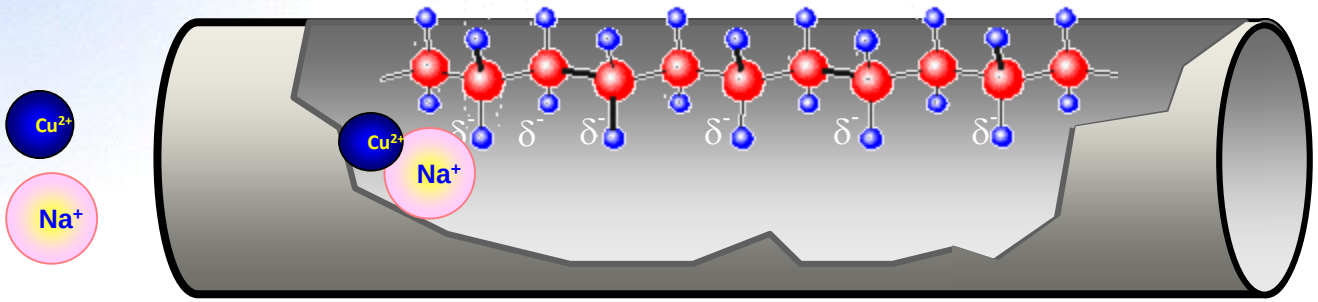
Iminodiacetate chelating resin

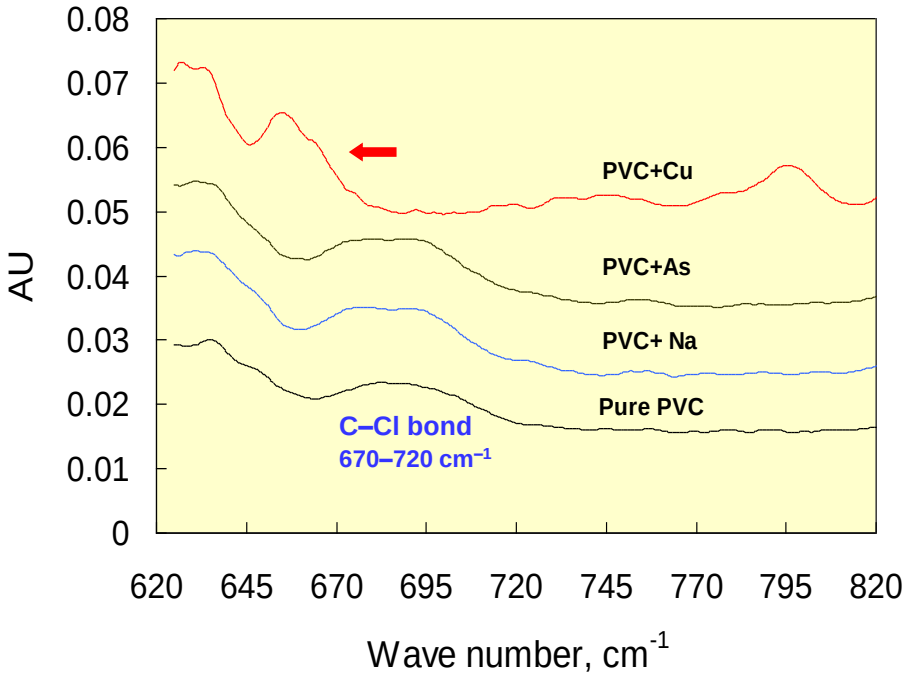
Sample loading **Washing** **Elution** **Conditioning**

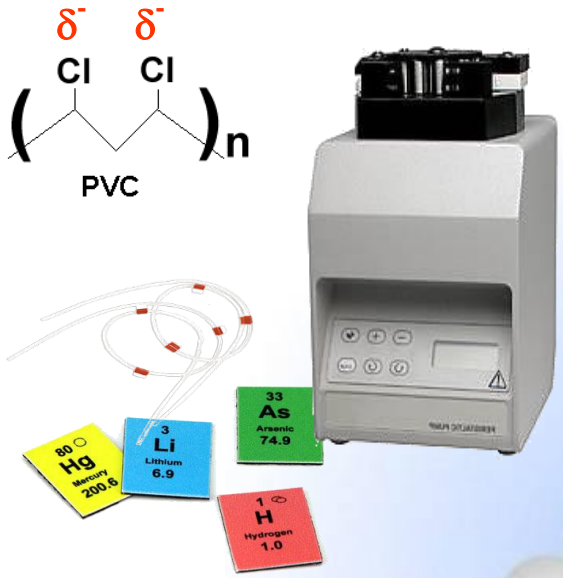


In-tube PVC Solid Phase Extraction

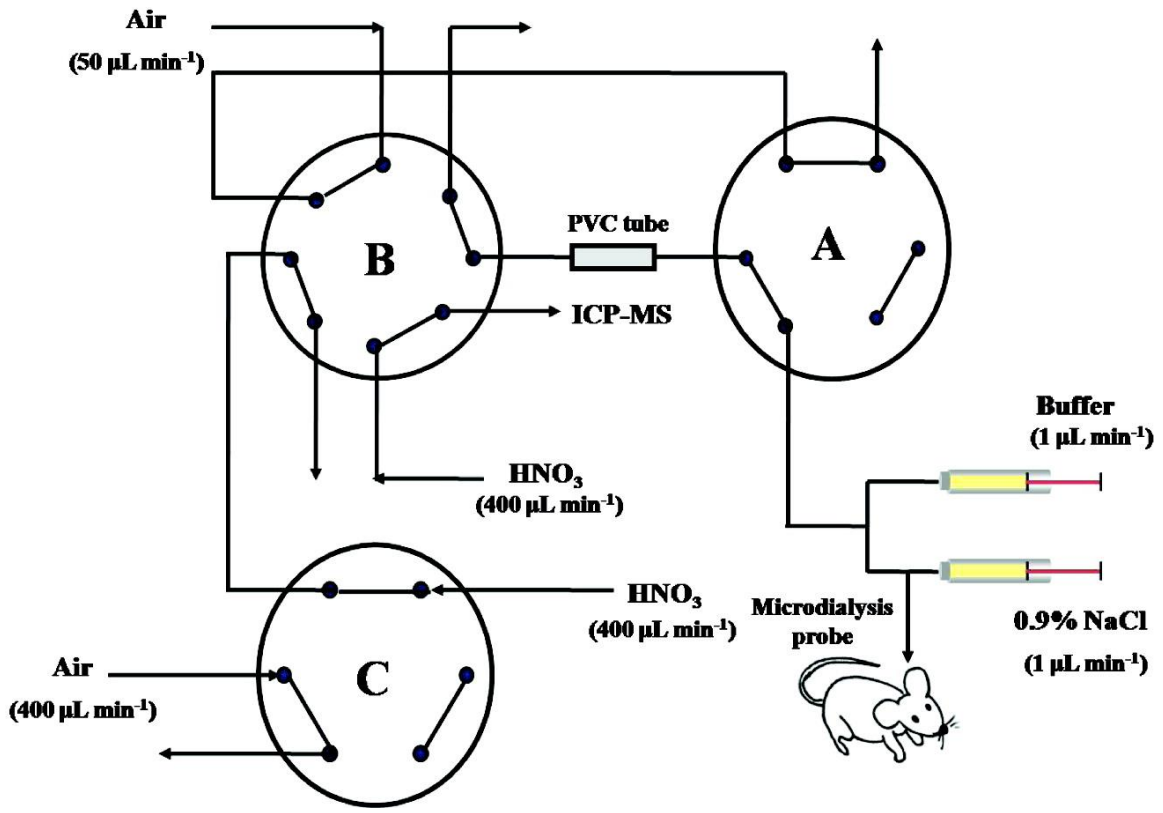
Ion-Dipole Forces $\propto |Z|\mu/r^2$







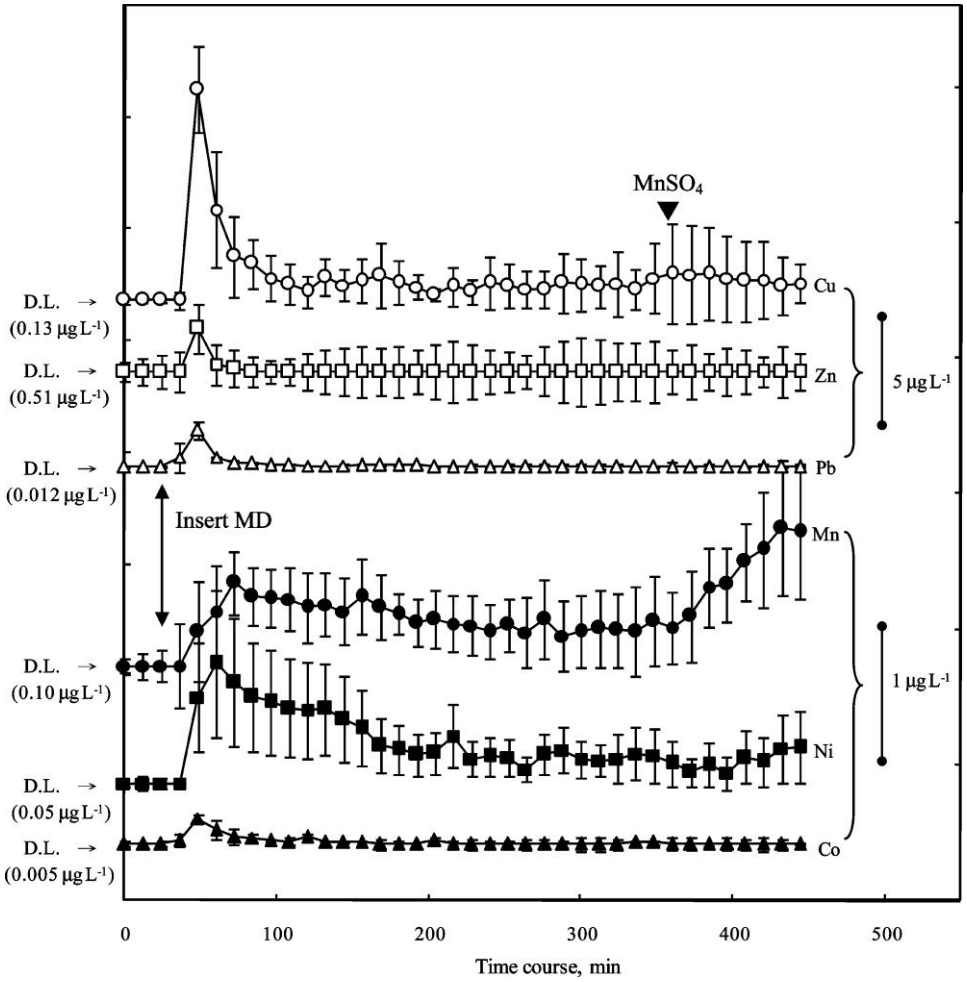
Schematic representation of the flow injection laboratory-on-valve system

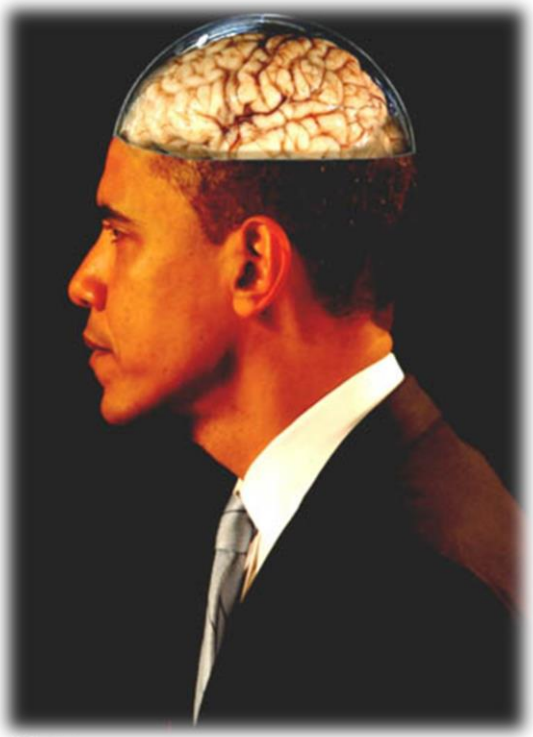


Schematic representation of the flow injection laboratory-on-valve system for trace element preconcentration via complexation with a nonfunctionalized PVC tube. VA and VC: six-port rotary valves. VB: eight-port rotary valves.

Published in: C. K. Su; T. W. Li; Y. C. Sun; *Anal. Chem.* **2008,** 80, 6959-6967.
DOI: 10.1021/ac800802e

Time course of the concentrations of trace metals in the brain extracellular fluid of a living rat





Published in: C. K. Su; T. W. Li; Y. C. Sun; *Anal. Chem.* **2008,** 80, 6959-6967.
DOI: 10.1021/ac800802e

JAAS

Cite this: *J. Anal. At. Spectrom.*, 2012, **27**, 56

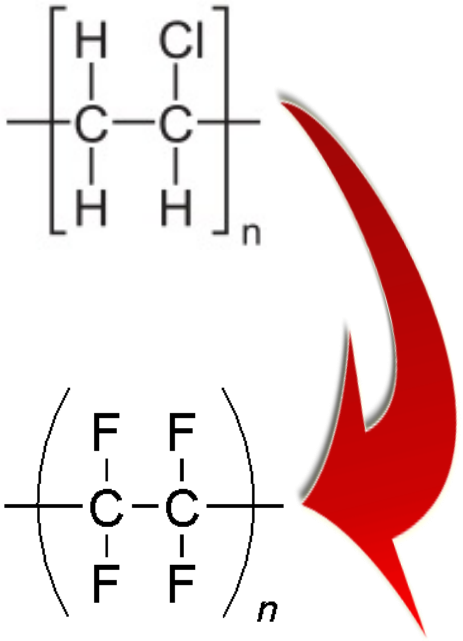
www.rsc.org/jaas

PAPER

Simultaneous *in vivo* monitoring of multiple brain metals using an online microdialysis-in-loop solid phase extraction-inductively coupled plasma mass spectrometry system†

Cheng-Kuan Su,[a] Yan-Ling Lin[a] and Yuh-Chang Sun*[ab]

Received 3rd June 2011, Accepted 26th September 2011
DOI: 10.1039/c1ja10168d



Using a non-functionalized small-bore polytetrafluoroethylene (PTFE) sample loop as a preconcentrator, a novel hyphenated system—comprising microdialysis sampling, online automatic in-loop solid phase extraction (SPE), and inductively coupled plasma mass spectrometry (ICP-MS)—for monitoring the levels of trace metal ions in living rat brains was developed. Taking advantage of the selective polymer–ion interaction to online separate metal ions from highly saline operating in the in-loop PTFE SPE system, it could online remove the salt matrix selectively from the microdialysate of rat brain extracellular fluid after slightly adjusting the pH; the next analytical cycle could be performed without the need for a pre-conditioning procedure after the prior elution with HNO_3. Owing to the simplicity and convenience of the developed operation sequence, this method combines extremely low blank level and detection limits (0.003–0.5 μg L^{-1}) and acceptable spike recoveries (90–98%) with the ability to analyze multiple elements. To demonstrate analytical reliability and compatibility, the analysis of standard reference material NIST 1643e (trace elements in water) and 2670a (trace elements in human urine) as well as continuous long-term monitoring of the variations of multiple trace metal ions in rat brain were performed in this work. According to the analytical results, it showed that the developed hyphenation system had the ability to analyze the SRM samples accurately, and the basal values of Ni (0.36 ± 0.16 μg L^{-1}), Zn (3.09 ± 0.31 μg L^{-1}), and Mn (1.31 ± 0.06 μg L^{-1}) in rat brain extracellular fluid could also be measured.