



AES NEWSLETTER



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Scattered thinking lately? Calm your intellect at the annual meeting of the American Electrophoresis Society, Nov 8-13, 2009

We are pleased to announce the 2009 annual AES meeting will be held in Nashville, TN in conjunction with the annual meeting of the American Institute of Chemical Engineers (AIChE). The AES meeting is designated Topical 3 and consists of 11 sessions running Monday, November 9 through Thursday, November 12. The detailed program is presented as an insert in this newsletter and is also available at <http://aiche.confex.com/aiche/2009/webprogram/T3.html>. The early registration deadline is September 28th and the registration form can be found at the AIChE website <http://www.aiche.org/Conferences/AnnualMeeting/index.aspx>. Remember that membership in AES qualifies for the discounted membership rate and can be checked on the PDF version of the registration form. AES will accept abstracts for submissions to the Topical 3 Poster Session with late-breaking results until October 19th. This year's poster session will include awards for the best student posters, based on judging by 3 members of the AES council, of \$100 for First Place and \$50 for Second Place for student members of the Society. If interested in making a late submission, please send an email to chesteki@uark.edu. The Poster Reception is scheduled for Tuesday, Nov 10, while the AES Banquet will take place on Wednesday, Nov 11 at the Ristorante Volare in the Gaylord Hotel. Tickets for the AES banquet are \$50.00 and can be purchased along with your AIChE registration. We look forward to seeing you there.



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Preparative Gel Electrophoresis of Native Metalloproteins

by Bernd Kastenholz (Forschungszentrum Jülich GmbH), Dr. David E. Garfin (American Electrophoresis Society) and Prof. Jürgen Horst (Westfälische Wilhelms-Universität Münster)

We have developed a method termed “quantitative preparative native continuous polyacrylamide gel electrophoresis” (QPNC-PAGE) that is a high-resolution technique for separating proteins by isoelectric point. We demonstrate this method with conditions that have been used to isolate active, native metalloproteins and to resolve properly- and improperly-folded metal cofactor-containing proteins in complex protein mixtures [1-6].

QPNC-PAGE is based on Bio-Rad’s Model 491 Prep Cell, an instrument for preparative gel electrophoresis of all kinds. The procedure is done in an unmodified Prep Cell [7]. Through use of a high-pH buffer, most proteins are negatively charged and migrate from the cathode to the anode during electrophoresis [1]. Although the pH value (pH 10) of the electrophoresis buffer is not physiological, protein isomers are continuously eluted at physiological pH (pH 8) [2].

In order to ensure full polymerization, the polymerization reaction is allowed to go for 69 hr at room temperature. As a result, the prepared gel is homogeneous, mechanically stable, and free of monomers or radicals. The pore sizes of the prepared gel are very large and therefore sieving effects are minimal [3,4]. Separated metalloproteins (e.g., metal chaperones, prions, metal transport proteins, amyloids, metalloenzymes, metallopeptides) are not dissociated into apoproteins and metal cofactors under the conditions given here. We believe also that metalloproteins do not undergo significant conformational changes during QPNC-PAGE. Consequently, quantitative amounts of highly purified metalloproteins are isolated in a few specific PAGE fractions [3,4].

Fe, Cu, Zn, Ni, Mo, Pd, Co, Mn, Pt, Cr, Cd and other metal cofactors can be identified and quantified by inductively coupled plasma mass spectrometry or by graphite furnace atomic absorption spectrometry in PAGE fractions of human, plant and animal samples [3,4].

The structure-function relationships of isolated metalloproteins in brain, blood or other clinical samples are important because improperly-folded metal proteins, for example, copper chaperone for superoxide dismutase (CCS) or superoxide dismutase (SOD), present in these biomatrices may be responsible for neurodegenerative diseases like Alzheimer’s disease or Amyotrophic Lateral Sclerosis. Active CCS or SOD molecules contribute to intracellular homeostatic control of metal ions (Cu, Zn) in organisms and thus these biomolecules can balance pro-oxidative and anti-oxidative processes in the cytoplasm [3-6].

A combined procedure consisting of QPNC-PAGE, SEC, ICP-MS and solution NMR is an effective approach for elucidating the different structures of physiologically relevant metalloproteins in biofluids (e.g., urine, blood) and to evaluate the relative biochemical impact of metal cofactor-containing (herbal) drugs in patients and probands [3-8]. It is anticipated that the results obtained by this analytical process will contribute to an early diagnosis and therapy of several protein-misfolding diseases, especially Alzheimer’s disease (Figure 1).

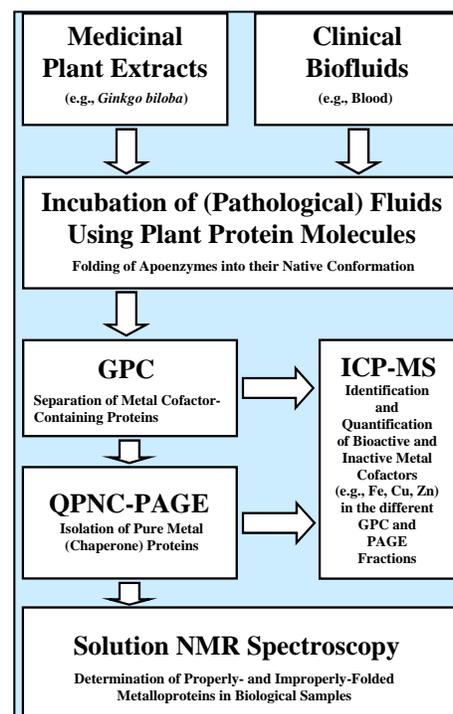


Figure 1. Workflow schemes for analysis of metalloproteins and protein-protein interactions incorporating QPNC-PAGE [5]. Used with permission of Bentham Science Publishers Ltd.

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Electrokinetic-Hydrodynamics (EKHD): A Much Needed Framework in Applied Sensitive Electrical Field Technologies by Jennifer Pascal, Pedro E. Arce, Mario Oyanader (Tennessee Tech University) and Sharon Sauer (Rose-Hulman Inst Technology)

Many problems in current engineering applications are within the family of “applied field sensitive process technologies” that require an external field to move solutes (molecules, particles, drops, bubbles, etc.) in a fluid within a porous domain. The presence of an external field (electrical, magnetic, acoustic, or photon-based) results in an additional conservation variable. The associated potential and forces make the analysis of the system very challenging, and can create a number of new problems for the practicing engineer, or frequently, the student and advisor. To aid in understanding such problems, a unified body of knowledge would be useful to link the variable introduced by the applied field to quantities of the hydrodynamics and solute transport. The electrokinetic-hydrodynamics (EKHD) framework described here (Arce et al., 2005) is directed at achieving this goal.

Unified descriptions of other systems with applied fields have been developed where the physical description of the system is coupled to the hydrodynamics contribution, for example magneto-hydrodynamics (Alfvén, 1942), physicochemical hydrodynamics (Levich, 1962, Probstein, 1991), and microhydrodynamics (Batchelor, 1977; Kim and Karrila, 1991). The word “coupled” here is used in the sense of necessary information from fluid dynamics to obtain a description of the transport behavior of the system.

However, this is not the case for electrical-field based applications since the concepts of electrokinetics and hydrodynamics have generally been decoupled (Saville, 2004), indicating the need for a more cohesive description of these concepts. Traditionally, the subject of electrokinetics has been associated primarily with colloidal sciences (Hunter, 1994) whereas hydrodynamics and solute transport have been more closely aligned with engineering (Bird et al., 1960). While some elements of unification can be found in the classical textbooks (Levich, 1962; Probstein, 1991; Masiyah, 1994), a systematic framework, in particular for the domain of continuum mechanics, seems to be missing.

Some applications utilize low electrical fields, such as separation

of biomolecules, soil remediation, and the motion of colloidal sized particles in porous media, whereas others require high electrical fields, such as high oxidation methods and cold plasma techniques. Motivated by the successful and systematic transport phenomena framework described in Bird et al. (1960), we have formulated a systematic methodology, termed “electrokinetic-hydrodynamics (EKHD),” to effectively model and capture the behavior of the wide variety of systems in which the concepts of electrokinetics as well as hydrodynamics and solute transport play a major role.

Because of the significant role played by hydrodynamics in systems with an applied electrical field that creates bulk flows of the electrolyte solution, i.e., electrohydrodynamic flows, we view problems within EKHD as involving two central aspects: the motion of fluids under an applied electrical field (i.e., electrohydrodynamics) and the motion of solutes/analytes within the fluids (i.e., electro- and convective-diffusive transport) (Delgado et al., 2005; Probstein, 1991). The EKHD method begins by examining these two domains necessary to effectively model systems with applied electrical fields: the fluid domain (continuum domain) and the solute domain (discrete domain).

Qualitative and quantitative information about the behavior of the two-domain system in EKHD can be effectively obtained by sequentially coupling the fluid domain to the solute domain for numerous cases (Bird et al., 1960). An up-scaled or macro-scale description can be achieved by using the mathematically effective spatial averaging technique (SAT) (Whitaker, S., 1999), which involves the use of information from the fluid domain as input into equations describing the solute domain. This approach leads to obtaining information about the macrotransport behavior of the system, such as effective velocity and effective diffusion, without actually solving the system of nonlinear coupled equations. This information can then be used to gain a better understanding of the roles that the various parameters (e.g., applied electrical field, electrophoretic mobility, Peclet number, etc.) play on the overall physical behavior of the systems under study. Moreover, predictions of the system behavior, for example the efficiencies of separation of biomolecules or the removal of contaminants from soil, can be made.

We hope that by recognizing the rich domain of EKHD, new collaborative efforts can be fostered and a more systematic and didactic/learning framework identified. EKHD is full of non-linear

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problems and highly coupled equations that need 1) new and innovative methods to achieve their modeling, 2) solutions that help with the development of new technologies and 3) the improvement of those technologies already in existence.

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New Review: 25th Annual Meeting of the American Electrophoresis Society by David E. Garfin

In *Expert Review of Proteomics*,
June 2009, 6: #3, 239-41.

Go to: www.expert-reviews.com/toc/epr/6/3 for the pdf of this review of the 2008 AES Meeting in Philadelphia.

Late breaking abstracts for the Poster Session will be accepted until Oct 19th

The abstracts will be printed in a poster booklet available at the meeting and also posted on the web site. AES student posters will be judged at the Poster Reception and awards presented for 1st (\$100) and 2nd (\$50) place. Submit late-breaking abstract to Dr. Christa Hestekin chesteki@uark.edu