

Red River Valley Local Section American Chemical Society

Next Meeting

Friday, 14 November 2008 in Grand Forks

Speaker : Dr. Tom Gonnella , Mayville State University

Location: Abbott Hall, Room 138, U N D Campus.

▲ 17:30 Presentation:

5:30 PM

- Following presentation Dinner: Whitey's of East Grand Forks.
121 DeMers Avenue
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Speaker's presentation:

● Evaluation of the ALDH-NADH Interactions with Real-time Fluorescence Analysis.

Within many mammalian systems aldehyde dehydrogenases (ALDH's) convert toxic aldehydes into more manageable carboxylic acids. Nicotinamide adenine dinucleotide (NAD⁺) is an essential coenzyme for this enzymatic process to occur. Through the course of a five step reaction scheme¹, NAD⁺ is converted to its reduced form (NADH) and released. The release of NADH from the ALDH is necessary for further turnover of the enzyme. The addition of magnesium ions to these ALDH systems modulates activity, in some cases increasing activity (ALDH2) and in others, like ALDH1, decreasing activity. The specific role of the magnesium ions in the reaction scheme of the different ALDH's is in the process of being resolved.²

Our research is focused on using the intrinsic fluorescence of NADH to provide fundamental information regarding factors that influence the enzyme-cofactor interactions. By applying time resolved fluorescence spectroscopy we have been able to distinguish between free NADH in solution ($\tau = 0.4$ ns) and NADH bound to recombinant rat ALDH2 ($\tau = 6.0$ ns) in the presence of magnesium ions. With such a dramatic difference in fluorescence lifetimes, we have been able to determine ALDH2 enzyme activity, the steady state NADH-ALDH2 complex concentration, the rate of NADH displacement from NADH-ALDH2 complex and the dissociation constant for NADH with ALDH2 under a wide range of magnesium ion concentrations. From these results we have been able to more clearly identify the influence the magnesium ion has on individual steps in the overall five step reaction. Future studies will involve closely examining the influence other divalent metal cations have on this system.

[†] This project is supported by NIH grant P20 RR016741 from the INBRE Program of the National Center for Research Resources.