CC/NUMBER 16
APRIL 21, 1986

This Week's Citation Classic®

Metzler D E, Ikawa M & Snell E E. A general mechanism for vitamin B_6-catalyzed reactions. *J. Amer. Chem. Soc.* 76:648-52, 1954.
[Biochemical Institute and Department of Chemistry, University of Texas, and Clayton Foundation for Research, Austin, TX]

From studies of nonenzymic model reactions of pyridoxal (the aldehyde form of vitamin B_6), the structural features needed for enzymic catalysis involving pyridoxal phosphate were deduced. A general mechanism that makes use of the electron-accepting properties of the protonated pyridine ring was proposed. [The *SCI*® indicates that this paper has been cited in over 585 publications since 1955.]

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March 26, 1985

During the early 1940s, Esmond E. Snell showed that pyridoxine, vitamin B_6 alcohol, could be oxidized to a biologically active aldehyde, pyridoxal, which was converted to an amine, pyridoxamine, upon heating with amino acids. Snell suggested that vitamin B_6, through the interconversion of pyridoxal and pyridoxamine, might serve as a coenzyme for transamination.[1]

When I started my graduate studies with Snell in 1948 at the University of Wisconsin, the role of pyridoxal 5'-phosphate in transaminases had already been established and amino acid decarboxylases had been shown to require the same coenzyme. Snell wanted to study the nonenzymic transamination quantitatively. I was a logical choice for the project since I liked analytical chemistry and had done undergraduate research under Ernest H. Swift at the California Institute of Technology. As I was struggling with erratic analytical values, Snell encouraged me to try more compounds and new conditions. One day he handed me a bottle of EDTA. It inhibited our reactions strongly, suggesting metal catalysis. Now, by adding known concentrations of catalytic metal ions, I was able to make quantitative measurements.

We were surprised to learn that heating serine with pyridoxal and Al^{3+} gave pyruvate. We then showed that the corresponding enzyme was activated by pyridoxal phosphate. On the other hand, threonine was cleaved to acetaldehyde. Racemization of amino acids also occurred. By this time we had moved to the University of Texas where Joanne Olivard, Jack Longenecker, and Miyoshi Ikawa joined in the effort.

Ikawa synthesized several compounds including 2-formyl-3-hydroxypyridine, which was also a good catalyst. From the study of these compounds, we concluded that the 3-hydroxyl group of the coenzyme was needed to form a metal chelate with a Schiff base of pyridoxal and the amino acid. The powerful electron-withdrawing ability of the N-protonated pyridine ring was also needed for catalysis. Taking some clues from the newly published *Chemistry of the Metal Chelate Compounds*, by Martell and Calvin,[2] we quickly deduced the common mechanism for all of the catalyzed reactions.

There are several reasons that our paper has been cited often. People like the fact that so many different enzymic reactions can be understood from the underlying chemical properties of the coenzyme. Organic chemists like the versatility of the coenzyme and inorganic chemists the metal chelation, which, however, is not present in enzymes.

I felt bad that we did not properly cite a short paper by Braunstein and Shemyakin published in 1952[3] and a full-length account published in 1953.[4] We did cite a *Chemical Abstracts* account,[5] but it was not possible to know that Braunstein and Shemyakin had proposed many aspects of our theory. Happily, our paper, with its emphasis on chemical model reactions, was nicely complementary to theirs; their detailed 1953 paper[4] is usually cited along with ours. Much new information in this field is summarized in a treatise.[6]

1. **Snell E E.** Summary of known metabolic functions of nicotinic acid, riboflavin and vitamin B_6. *Physiol. Rev.* 33:509-24, 1953. (Cited 95 times since 1955.)
2. **Martell A E & Calvin M.** *Chemistry of the metal chelate compounds.* New York: Prentice-Hall, 1952. 613 p. (Cited 1,665 times since 1955.)
3. **Braunstein A E & Shemyakin M M.** Theory of processes of amino acid metabolism that are catalyzed by pyridoxal enzymes. *Dokl. Akad. Nauk SSSR* 85:1115-18, 1952. (Cited 35 times since 1955.)
4. ----------. Teoriia protsessov aminokislotnogo obmena, kataliziruemykh piridoksalevymi enzimami. *Biokhimiya* 18:393-411, 1953. (Cited 170 times since 1955.)
5. ----------. Theory of processes of amino acid metabolism that are catalyzed by pyridoxal enzymes. (Abstract.) *Chem. Abstr.* 47:626, 1953.
6. **Christen P & Metzler D E,** eds. *Transaminases.* New York: Wiley, 1985. 643 p.

