

## ***This Week's Citation Classic***

**Tanford C.** Protein denaturation. *Advan. Prot. Chem.* **23**:121-282, 1968.  
[Dept. Biochemistry, Duke Univ. Medical Center, Durham, NC]

**This paper reviews work that had been done in my laboratory during the preceding several years on the characterization of proteins denatured by urea and guanidine hydrochloride, and similar work done elsewhere on other protein denaturants. [The SC® indicates that this paper has been cited over 680 times since 1968.]**

Charles Tanford  
Department of Biochemistry  
Duke University Medical Center  
Durham, NC 27710

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"Proteins, alive and functioning, carry within their structure the secret of virtually all life processes. But when they are denatured, they are dead, like a car in a junkyard. Who today would want to study denatured proteins? I don't know, and don't know why there should be more than scattered citations of my review in the literature.

"In the 1960s there was a valid biological reason for investigating denatured proteins. There was a growing conviction that the three-dimensional structure and biological activity of proteins are uniquely determined by the amino acid sequence of their constituent polypeptide chains. The conviction was based on the spontaneous recovery of structure and function after denaturation by guanidine hydrochloride and subsequent removal of the denaturing agent. But that experiment actually does not prove anything, unless it can be demonstrated that all secondary and tertiary structure had been lost when the protein was transiently in the denatured

state. (In fact, had the argument been based on recovery from denaturation by sodium dodecyl sulfate, it would have been invalid. As Jacqueline Reynolds showed later, this denaturing agent does not cause loss of protein secondary structure.<sup>1</sup>)

"Purely by chance, I had in my laboratory at the time two exceptionally good polymer chemists, Kazuo Kawahara from the University of Osaka and Savo Lapanje from the University of Ljubljana, and they agreed to help me study the state of a number of proteins in guanidine hydrochloride solution. Our approach was rigorous and objective, and we had no vested interest in what the result would be. As it turned out, the denatured proteins did in fact behave as truly structureless polymer chains ('random coils'), and the sequence/structure/function hypothesis was proved. The original work (which, I should mention, ultimately involved several students and postdoctoral fellows in addition to Kawahara and Lapanje) was published in about a dozen papers. I summarized what we had learned in the review that is the subject of this article, and added to it what was then known about other denaturing agents, such as alcohols, heat, etc.

"Live proteins are more interesting than dead ones, and I have not pursued the subject of denaturation. Looking through the papers published from my laboratory during the last six years, I find that we ourselves have cited the denaturation review only twice, both times in connection with demonstrating that a protein we were studying resisted denaturation by guanidine hydrochloride."

1. **Reynolds J A & Tanford C.** The gross conformation of protein-sodium dodecyl sulfate complexes. *J. Biol. Chem.* **245**:5161-5, 1970.