

This Week's Citation Classic

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Bencze W L & Schmid K. Determination of tyrosine and tryptophan in proteins.

Anal. Chem. **29**:1193-6, 1957.

[Dept. Medicine. Harvard Med. Sch., and Massachusetts Gen. Hosp., Boston, MA]

A spectrophotometric method is described which relies on the pattern of tyrosine and tryptophan ultraviolet absorption peaks rather than on the exact location of the absorption maxima. By addition of tyr and try stock solutions to the native protein the degree of the inherent background or extraneous absorption of the protein can be estimated. [The SC[®] indicates that this paper has been cited over 695 times since 1961.]

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"Upon my immigration to the US in 1954 my task was to contribute new data towards the characterization of human acid glycoprotein. I arrived in Boston as a postdoctoral fellow coming from the University of Zürich, Switzerland, where I had earned my PhD in organic chemistry under the guidance of the late Hans Schmid. My new master at Massachusetts General Hospital was Karl Schmid. Although both Schmid's were born in the same county of Switzerland, they were no kin. Both of them were research oriented teachers, endowed with the exactitude of a Swiss watchmaker, assiduously implanting their meticulous techniques in their students and co-workers. I had been duly impressed with the painstaking care which afforded the pure glycoprotein and pledged that I would devote the same heaping measure of care to each milligram of the protein that was entrusted to me.

"Recalling the skilled eagle eyes of my teacher in Switzerland, who would cast a glance on the ultraviolet absorption spectrum of a plant extract and then proclaim that it contained a coumarin but no chromone, I commenced to record the spectra of varying mixtures of tyr and try. It was then convincingly clear that the pattern of the ultraviolet absorption curve could reliably tell whether the ratio of tyr:try was 3:7 or 2:8, respectively. Consequently, a method was at hand that was independent from the bathochromic shift of the absorption maxima. The only problem awaiting to be tackled was the imponderable extraneous absorption. Again, the stock solutions of tyr and try offered their invaluable aid. Addition of known amounts of tyr and try to the solution of the native protein will furnish a new ultraviolet absorption pattern. Repeated additions of tyr and/or try and calculations should lead to an approximate assay of the extraneous absorption of the protein. We set up plans to study the tyr and try content of large proteins and to investigate light scattering as a part of the extraneous ultraviolet absorption.

"However, the printers ink on this publication wasn't even dry when I abandoned my engagement with proteins. I have turned to medicinal chemistry. Karl Schmid remained true and loyal to the proteins and keeps on turning out a steady stream of new data on his favorite glycoproteins.

"Perhaps it is the enticing simplicity of the method that turned the publication into a *Citation Classic*. However, the task to unravel the nature of the extraneous absorption remains a difficult and a disturbing problem. It may very well be the case that a substantial portion of the citations are cursing the method and its advocates in an outright fashion. Luckily, the computer merely counts the citations and will keep silent about the complaints."