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This Week's Citation Classic

Boyer P D. Spectrophotometric study of the reaction of protein sulfhydryl groups with organic mercurials. *J. Amer. Chem. Soc.* **76**:4331-47, 1954. [University of Minnesota. Department of Agricultural Biochemistry, Minneapolis, MN]

As stated in the summary of the paper, 'A procedure is described for the sensitive and rapid spectrophotometric measurement of the extent and rate of reaction of various organic mercury compounds, particularly *p*-mercuribenzoate, with sulfhydryl groups.' Included with the paper are examples of differing reactivities of sulfhydryl groups in proteins, and the effect of ionic composition and pH on rate and extent of reaction. [The *SCI*® indiates that this paper has been cited over 1315 times since 1961.]

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"In common with a number of its companion 'Citation Classics' the principal reason my paper has been frequently cited is probably because it fulfills a definite methodological need. Another reason is that the results demonstrate simply and quantitatively differing reactivities of sulfhydryl groups in proteins. This paper was a direct outgrowth of a need for quantitation of the number and reactivity of sulfhydryl groups in enzymes.

"H. L. Segal, as a graduate student in my laboratory, had obtained some of the first convincing evidence for acyl-S-enzyme formation as part of the coupling mechanism of glyceraldehyde 3-phosphate dehydrogenase.[1] We wanted more information about the number and reactivity of the sulfhydryl groups.

"The reaction of mercurials with sulfhydryl groups of proteins had been widely studied prior to this paper, but methods available were cumbersome. In considering the need and the problem, it seemed plausible that the orbital electrons in the benzene ring of *p*-mercuribenzoate would be sufficiently perturbed when a hydroxide ligand on the mercury is replaced by sulfur to allow detection by change in ultraviolet absorption. Experimental measurements showed that this was indeed the case; there was sufficient increase in absorption accompanying mercaptide formation to give a sensitive measure.

"It is fortunate for the application of the procedure that the change in ultraviolet spectrum is considerable in the 250-260 nm region where there is a minimum in the absorption of proteins. Since this publication a number of other measures have been found for sulfhydryl groups, but the spectrophotometric procedure with *p*-mercuribenzoate remains useful for measurement of mercaptide formation.

"I recall another interesting aspect of this work. It was supported by a modest grant from the National Science Foundation, and included in the approved budget were sufficient funds to allow me to travel to a scientific meeting. The research was scheduled for presentation at the 123rd Meeting of the American Chemical Society in Los Angeles, but my use of the National Science Foundation funds was blocked by an administrator of the University of Minnesota on the basis that this very large university had already approved attendance of another staff member from another department at this meeting.

"My objections were of no avail, and the paper was presented at my personal expense. This is an example of poor administration at a fine university, which in general has been administered much better than most. More importantly, such action was a forerunner of the present condition of science support where the time expended and restrictions required for conformity to regulation often stifle progress."

1. **Segal H L & Boyer P D.** The role of sulfhydryl groups in the activity of glyceraldehyde 3-phosphate dehydrogenase. *J Biol. Chem.* **204**:264-81, 1953.