A description is given for the preparation of adenosine deaminase, adenylic deaminase, xanthine oxidase, uricase, nucleoside phosphorylase, and guanase in sufficient purity to be used as analytical reagents for the measurement of the purines on which they act. [The SCP indicates that this paper has been cited in over 1,185 publications since 1955.]

Herman M. Kalckar
Department of Chemistry
Boston University
Boston, MA 02215

April 30, 1984

The principle of determining enzymatic reactions by following optical changes at specific wavelengths had not, as far as I knew in 1944, been used in the US, at least not in the ultraviolet region. In Berlin, Otto Warburg, who built ultraviolet spectrophotometers in his biochemistry lab, introduced the principle in his studies of redox enzymes in 1935.1

"In 1943, working at the Public Health Institute of the City of New York, Inc., I had available to me for the first time a Beckman ultraviolet spectrophotometer. I now explored some known spectral changes in the more shortwaved ultraviolet spectra of purines, purine nucleosides, or nucleotides which had been observed by changing the pH from the acid to the alkaline range. It soon dawned upon me that these sensitive spectral changes could be used in the field of enzymology of nucleosides and nucleotides.

"This was first published in Federation Proceedings,2 where I also introduced a new active ester ribose-1-phosphate and its enzymic reaction with purines to form nucleosides (in surprising good yields). The details of this work appeared in 1947 in the Journal of Biological Chemistry. This appeared in four papers entitled 'Differential spectrophotometry of purine compounds by means of specific enzymes, I-IV' by myself (with the technical assistance of Manya Shafran)3-5. The third paper is the one being classified as one of the most-cited items in the biochemistry literature. This paper focuses on the determination of a variety of enzymes of purine, nucleoside, and nucleotide metabolism. Some of these enzymes, like uricase or xanthine oxidase, as well as their respective substrates, also happen to have clinical interest. Hence, when ultraviolet spectrophotometers became a common item in clinical labs, the methods were probably deemed of additional interest."

"A clinical derivation of the principle of ultraviolet spectrophotometry of purines was presented in the 1947 paper, through the ultraviolet determination of uric acid in serum (see, for instance, the Sigma catalog). This is a sensitive method for use in cases like arthritic urica, or in infants, to spot the serious inborn error, the Lesch-Nyhan syndrome (lack of the enzyme of HGPRT)."

"For recent references, see Murphy et al.6 and Tritsch."7