THE DEVELOPMENT OF A SPECIFIC ASSAY FOR GLUTARALDEHYDE AND THE IDENTIFICATION OF THE ASSAY REACTION PRODUCT

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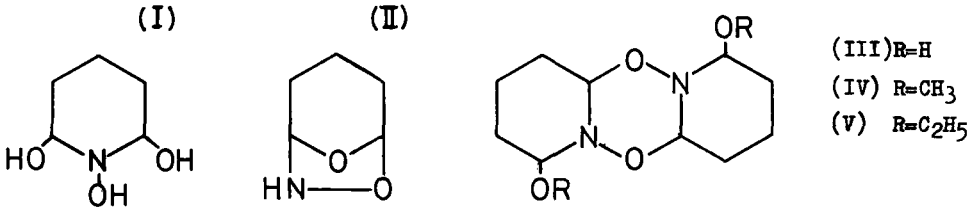
Glutaraldehyde solutions (2%w/v) are commonly used as chemical sterilising agents for heat sensitive equipment. Glutaraldehyde has traditionally been assayed by titrating the acid liberated when it is reacted with hydroxylamine hydrochloride. This method is very non-specific because the reaction is characteristic of carbonyl compounds generally and, hence unsuitable as the stability-indicating assay which we required. It is also subject to interference from the alkaline activators which are added to glutaraldehyde solutions prior to use.

Friedrich and Hadju (1975) described a spectrophotometric method for the determination of glutaraldehyde, sensitive to 0.01%. The method is based upon the reaction with a 25 molar excess of hydroxylamine and comprised the measurement of the difference in absorbance (238nM) between the reaction products and a strong chromophore which is transiently formed in the reaction mixture. The method proved inconvenient to use for our application because the rate of formation of the intermediate is temperature dependent and the time at which its absorbance is measured is critical. Since the reaction product, presumably glutaraldehyde dioxime, did not absorb at 238nM, we were intrigued that the intermediate should be such a strong chromophore; the reaction obviously did not proceed via the straight chain monoxime. Glutaraldehyde could be expected to react with primary amines to form N-substituted piperidines and it seemed that the intermediate was likely to be the trihydroxypiperidine (I).



Since other aldehydes would not undergo such a reaction, its potential as a specific assay for glutaraldehyde was obvious. We have developed such a method which is not only sensitive to glutaraldehyde at the 0.001% level but is also free of interference from other aldehydes and the polymeric compounds formed in the degradation of glutaraldehyde solutions.

To establish the structure of the intermediate it was isolated from a concentrated reaction mixture as a white crystalline solid (A). Reference to the literature revealed that glutaraldehyde monoxime had previously been assigned the unlikely structure (II), (Heimberger et al 1970). Although the melting point and infrared spectrum of compound (A) are consistent with (II) the mass spectrum is indicative of a molecular weight of 230 and accordingly the structure (III) is proposed. An N.M.R. spectrum could not be obtained because of the insolubility of the compound but the proposed structure (III) has been confirmed by the further reaction to give the dialkoxy dipiperidinodioxadiazines (IV and V) the structures of which have been reported previously, Eikelmann et al (1972).

J. Hadju, P. Friedrich (1975) Anal. Biochem. 65 273–80
G. Eikelmann et al (1972) Liebigs Ann. Chem. 759 183–188
W. Heimberger et al (1970) German Patent No. 1928264

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