Fox, D. J. and Boulter, D. Rapid localization of proteins in chromatographic eluates.

The customary procedure for establishing the elution position of proteins from chromatographic columns is to measure the $E^{280}$ mμ in a spectrophotometer. This procedure can become very laborious when the components are few in number and are widely separated on the elution curve. In such cases it has been found possible to localize the protein-containing fractions by rapid visual examination under ultra-violet light from a Black Glass U. V. lamp in the dark. The proteins fluoresce vividly, usually a pale blue color, and the eye can detect any concentration that is measurable with an S. P. 500 spectrophotometer. This method is satisfactory even when the fractions are contained in pyrex test tubes. Once localized the absorption can be measured accurately in the spectrophotometer. Owing to the interference from other absorbing compounds, principally nucleic acids, it is customary to measure the extinction at 280 and 260 mμ and make a Warburg-Christian correction (Warburg and Christian, Biochem. Z. 310, 384, 1941). We have found it profitable, when dealing with large numbers of fractions, to construct a Nomogram from the original Warburg and Christian figures and from this a table converting observed $E^{280}$ and $E^{260}$ to the value of $E^{280}$ that the protein component alone would have given, thus eliminating any errors arising from the variation of aromatic amino acid composition inherent in direct conversion to mg. protein by the Warburg-Christian equation. The only assumption here is that the extraneous absorption is in fact due to nucleic acids or their components.

This corrected $E^{280}$ may at any future time be converted to mg. protein when the conversion factor for the particular proteins are determined after purification.

Using this table, corrections of over 50% have been made to some peaks.--The Hartley Botanical Laboratories, The University, Liverpool.