

assays: protein

Absorbance 280 nm

The simplest, most straightforward method for estimating protein content is measurement of OD 280 nm. In this assay, ultraviolet light is absorbed by aromatic residues at a wavelength of 280 nm. In order to use this UV absorbance assay, an unknown protein must contain at least one aromatic tyrosine residue to generate a measurable signal. As a rule of thumb, many proteins will yield an OD of about 1.0 for a 1 mg/ml solution.

In spite of its apparent simplicity, the OD 280 nm method suffers from several drawbacks. For example, this method consumes more sample than many other methods because of the residual protein solution left in the cuvette after measurement. In applications in which the protein to be measured is in the presence of buffers or other substances that have UV absorbing components, the method becomes highly unreliable. In nature, the UV absorbing characteristics of proteins can vary widely; therefore, when using this protein quantitation method, a standard curve for each unknown protein must be established to obtain accurate results. In addition, the Absorbance 280 nm method requires expensive quartz cuvettes and a photometer with UV wavelengths.