

quaternary structure all affect absorbance, therefore factors such as pH, ionic strength, etc. can alter the absorbance spectrum.

Aromatic residues, like tyrosine and tryptophan, absorb UV light at 280 nm. So, if an extinction coefficient is known for protein (ϵ), the absorbance can be measured in a UV/Vis spectrometer and calculate the concentration of protein using Beer's law ($A = \epsilon lc$, where l is the path length of the spectrometer).

In BIURET COLORIMETRIC ASSAY OF SERUM PROTEIN, Proteins react with copper ions in alkaline solution to form a violet- colored complex that absorbs light at 550 nm. This reaction is the basis of the biuret assay for protein. Biuret reagent is a solution of CuSO_4 in NaOH . When you add biuret reagent to a protein solution the reaction produces a solution of the protein-biuret complex in a concentration that is the same as the original concentration of protein. Therefore, by measuring the concentration of the complex, using A_{550} (Absorbance at 550 nm), you are also measuring the concentration of protein. The assay consists of two parts that will be conducted simultaneously. The first part consists of establishing a standard curve by using several known quantities of a pure protein. In this experiment bovine serum albumin (BSA) will be used as the protein to be reacted with the biuret reagent. Since all proteins react with the biuret agent in the same manner, this standard curve will give us an E value that can be applied to any protein solution reacted with biuret. The second part consists of reacting measured quantities of serum (protein) with the biuret reagent and measuring the Absorbance of each reaction mixture.

Bradford Assay and BCA Assay are other methods of calculating the concentration of the protein sample.

Conclusion:

The UV/Vis spectroscopy results show that plant sample has a higher concentration of protein in directly diluted protein sample.

References: