

# Hydrodynamic studies on bovine serum albumin in multicomponent solutions

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The partial specific volume and some hydrodynamic properties of bovine serum albumin have been investigated in aqueous solutions of different salts (varying both the cation and anion), buffer systems ( $K_2HPO_4/KH_2PO_4$ , Tris/HCl, glycine/NaOH), sugars (glucose, fructose, sucrose), polyols (glycerol, ethylene glycol, polyethylene glycol), denaturing agents (guanidine.HCl, urea, sodium dodecyl sulfate) under various conditions: variation of the concentration of the additive, pH, addition of further components (e.g. dithiothreitol, 2-mercaptoethanol), pretreatment (e.g. oxidation with performic acid) etc.

The apparent specific volume of the protein depends on the experimental conditions (1-3): predominantly linear increase of the specific volume with increasing concentration of salts, buffer components, sugars, polyols; non-linear decrease upon denaturation. The results can be correlated with characteristics of the additives (e.g. physico-chemical nature of salts, buffer components, number of hydroxyl groups, "denaturing power").

The intrinsic viscosity of the protein, which decreases only slightly with increasing electrolyte concentration, increases significantly upon denaturation. This behaviour is caused by the unfolding of the protein under denaturing conditions; it is still more pronounced in the presence of factors promoting the unfolding process (further additives, pH). The results run parallel to the changes of the specific volume or other molecular parameters.

The sedimentation coefficient (s), as well as the apparent molecular weight (M) of the protein are strongly influenced by high salt concentrations. Applying the experimentally determined values for the specific volume yields correct values for s and M.

The effects observed for serum albumin (and also for a number of other proteins) in multicomponent solutions may be tentatively interpreted as a superposition of charge effects, hydration, preferential ligand binding, unfolding etc. The results may be significant in connection with the application of a variety of methods in multicomponent solutions (cf. (3)): e.g. determinations of M from analytical ultracentrifugation or small-angle scattering, correction of s-values from sedimentation velocity runs, density gradient centrifugation, use of the contrast variation method in small-angle scattering, interpretation of association or dissociation phenomena, calculation of interaction parameters, elucidation of the mechanism of protein crystallization etc.

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- 2) Tuengler, P. et al., (1979), Anal. Biochem., 98, 481.
- 3) Durchschlag, H., Abstr. 5th Int. Conf. on Small-Angle Scattering, Berlin 1980, pp. 135-136.