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The Interaction of Surfactant with Bovine Milk Casein by Viscometric Method

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ABSTRACT

Viscosity of bovine milk casein and sodium lauryl sulphate (SLS) suspensions have been investigated at different concentrations of protein as well as surfactant and viscometric constants such as intrinsic viscosity, specific viscosity, viscosity number, hydrodynamic radius and interaction index were calculated using standard equations. The instrinsic viscosity data and other parameters calculated from viscosity and it were used to explain the changes produced due to operating up of the protein molecule in the presence of the added surfactant. The mode of linkage of surfactant to protein is primarily electrostatic, while secondary association of further surfactant also takes place through non-polar attraction.
Keywords: Bovine milk casein, Viscometer.

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INTRODUCTION

Polymer–surfactant interactions [1-8] have been extensively investigated by researchers due to their manifold applications in the field of the food and pharmaceutical industry and in analytical biochemistry. The interactions of protein with mostly cationic and anionic surfactants ingredients are the particular interact because they are used co-operatively in formulated complexes. The mechanism for protein-surfactant interactions are polyelectrolyte absorption [9], hydrophobic [10] and ionic interaction [11] depending on the substrate and type of proteins involved. Different types of physiocochemical techniques, like surface tension, conductivity, viscosity etc. have been used to investigate the interactions between cationic and anianic surfactants with protein [12-14]. These interactions in aqueous media give rise to the formation of association structures thereby modifying the solution and interfacial properties [15,16]. The study on the effect of proteins on the properties of surfactants provides important information for interactions between surfactant and protein. Among the anionic surfactant, sodium lauryl sulphate occupied unique significance. Their interaction with proteins has been extensively investigated mainly due to its biochemical applications. The viscosity values have been used for the calculation of shape parameters like hydrodynamic radius as influenced by the different concentrations of the anionic surfactants. A possible mechanism of the viscometric changes has also been proposed.

MATERIAL AND METHODS

Sodium molydate (E.Merck) was dissolved in distilled water and its molybdenum was determined gravimetrically. Buffers and other solutions were prepared from reagent grade chemicals. A potassium chloride (BDH) solution was used for maintenance of ionic strength of reaction mixtures.

Protein Solution

Soyabean protein (SBP) was extracted from soyabean powder (BDH) by alkali extraction followed by the gradual adition of HCl to lower the pH to the isoelectronic point. It was dissolved in dilute alkali solution and centrifused to obtain the clean solution. The concentration of protein solution was determined by colorimetric method. It was stored in a refrigerator and purified toluene was added to check its surface denaturation.

pH-measurement

There measurements were carried out by a systronic pH-meter using a wide range glass electrode. The instrument was standardized against 0.05 molar potassium hydrogen phthalate (pH 4.0) and 0.05molar sodium borate (pH 9.20) for acidic and basic ranges, respectively. The standardized pH-meter was used to prepare the buffer solution of required pH values.

Viscosity Measurement

Ostawald Viscometer of relatively long capillary having flow time of water 65 seconds was used for viscosity measurement at temperature 25ºC in a water thermostat. Kinetic energy correction was found to be a negligible. Viscometer stand was arranged in a manner that it was always situated in the same position in the bath. Protein and surfactant stock solutions were centrifuged at 16000 rpm for 60 minutes to remove particulate matters. The densities of solvent and solution were determined with the help of pyknometer. The viscosity values were determined by the following relation:

$$\eta_{rel} = \frac{\eta}{\eta_0} = \frac{t.p}{t_0.p_0}$$

η_{rel} is relative viscosity, t and ρ are the flow time and density of the solution. Whereas to and ρ_0 are time of flow and density for the water used as solvent.

RESULT AND DISCUSSION

The values of viscosity, relative viscosity and specific viscosity of protein in absence and presence of surfactant were calculated by the use of proper relation. Following relation applies for dilute suspensions of solid particles.

$$\eta_{sp} = \eta_{rel} - 1 = \frac{\eta - \eta_0}{\eta_0} = V\phi \qquad (i)$$

Where η_{sp} is the specific viscosity, η is the viscosity of suspensions, η_0 is the viscosity of pure solvent, V is the volume fraction of the particles in the suspensions and ϕ is the shape factor. At higher concentrations of the solute-solute interaction is considered and a quadric term is to be added.

$$\eta_{SP} + V\phi + K\phi^2 \qquad (ii)$$

The value of ϕ in equation (ii) can be expressed in term of the product VC where V is specific solute volume and C is the concentration in g/ml.

$$\eta_{sp} = \gamma V_c + KV^2C^2 \qquad (iii)$$

Dividing by C gives

$$\eta_{sp}/C = \gamma V + KV^2 C^2 \qquad\qquad (iv)$$

The limiting value of η_{sp}/C as $C \to O$ is called intrinsic viscosity or intrinsic viscosity number $[\eta]$. It is a measure of effective hydrodynamic volume per gm of macromolecule. Equation (iv) now assumes the following form:

$$\eta_{sp}/C = [\eta] + K^1[\eta]^2 C \qquad\qquad (v)$$

Where K^1 is Huggins constant.

$$Ln\frac{\eta_{rel}}{C} = [\eta] + K[\eta]^2 \qquad\qquad (vi)$$

The quantity of $Ln(\eta_{rel}/C)$ intrinsic viscosity $[\eta]$ is also used in determining the hydrodynamic radius according to following equation.

$$Re = \left[\frac{3M}{10\pi N}[\eta]\right]^{1/3} \qquad\qquad (vii)$$

Where, M is the molecular weight of macromolecule, N is Avogadro number. Hence by the use of above equation $[\eta]$. K (Interaction Index) and Re were calculated by Viscosity measurement of protein–surfactant system.

Table I : Intrinsic viscosity and other parameters for Bovine milk-casein – SLS System at pH 8.0 and at 30ºC temperature.

Concentration of SLS added in gm/gm of protein	Intrinsic Viscosity (mlg^{-1})	Hydrodynamic radius Re (Aº)	Interaction Index (K)
0.00	39.40	85.35	5.20
0.50	54.95	91.23	4.11
0.62	62.80	96.04	3.60
1.20	72.60	104.86	3.00
2.40	102.00	113.68	2.25
3.60	118.70	119.56	1.95
4.80	126.50	120.54	1.86
6.00	135.34	120.54	1.66

It is also observed from the results that viscosity as well as intrinsic viscosity of protein–surfactant system at pH 8.0 increase with increase concentration of the surfactant. The total effect of surfactant in solubilization of protein and also unfolding it to permit the dispersed chain to form fibers. This behaviour depends upon chain length nature and distribution of polar

groups, flexibility of chains, tightness of packing and on the number of cross links. The presence of adsorbing surface helps the unfolding of the mild casein molecule, which is then following by gradual aggregation. The increase in viscosity of the system with increasing surfactant concentrations supports the above mentioned fact. The increase in the value of $[\eta]$ Table I is due to corresponding extension in the shapes of molecules and consequently due to the increase in the dissymmetry of suspended units.

CONCLUSION

The values of Re increase with increase $[\eta]$ and the progressive increase in Re values confirmed the swelling of protein molecule with using SLS surfactant concentration.

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