Ultrasonic and viscometric study of soya protein in aqueous solution

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The interaction of proteins, compatibility of biomaterials and molecular dynamics, solute in solvent have been investigated by ultrasonic technique. Ultrasonic velocities, viscosities and densities of aqueous solution of soya protein were measured as a function of concentration and resonance frequencies in the range 3-10 MHz ultrasonic waves. The acoustic parameters such as adiabatic compressibility, aquatic impedance and viscosity relaxation time were calculated at different aqueous concentration of soya protein. The adiabatic compressibility of soya protein has been found to decrease with rise in concentration at both frequencies. However, the decreasing tendency was stiffer at 10 MHz as compared to 3 MHz. It may be due to the weak interaction of water molecules with protein chains at 3 MHz. The acoustic impedance (Z) increases with increase in concentration at both frequencies of ultrasound. The viscosity relaxation time of soya protein decreases with concentration at ultrasonic frequency of 10 MHz, however, at lower frequency of ultrasound at 3 MHz, the value τ increases linearly with concentration.

Keywords: Acoustic impedance, Adiabatic compressibility, Relaxation time

1 Introduction

The soyabean is one of the most nutritious foods which belong to the family of legumes or pulses. It is grounded up to make soya flour which is then mixed with water to remove the soluble carbohydrate. The residue is formed into its final shape - chunks, mince or flakes. Soya protein is called as intact protein that means these are polypeptides containing purified forms of native proteins.

The application of ultrasonic methods for probing the structural dynamics of biopolymers such as proteins and polysaccharides has been the subject of wide research. The study of ultrasonic absorption of macromolecules including proteins was likely to have a contribution from a coupling between the thermal motion of the protein and water molecules, which changes the strength of their interaction with the protein. Only a few biologically important macromolecules have been subjected to thorough ultrasonic experiments. For hemoglobin, protein and dextran, the interaction between solvent and solute is probably the principal mechanism of acoustic absorption. Several researchers have used ultrasonic velocity (u) together with density (ρ) and viscosity (η) measurements to study protein-solvent interaction. The study of viscometric on the interaction and compatibility of protein solution has already been investigated. The compatibility and interaction studies of protein probed by sound velocity and related acoustic parameters are reported in Refs 11 and 12.

Polymers which are water soluble such as soya protein, have been studied because of their versatile pharmaceutical, biomedical and industrial applications. These include the preparation of many biomedical products, such as biocompatible hydrogels and anticoagulants through degradation and grafting onto these proteins. Because of this extensive applicability, a comparative study of acoustic parameters and viscosity of the pharmaceutically active soya protein in aqueous solution have been reported in the present paper. The protein-water compatibility and their acoustic properties by ultrasonic technique have been studied.

2 Experimental Details

Soyabean protein sample was supplied by the HiMedia Laboratories Pvt Ltd, Mumbai, India and used without further purification. The water used for the experiments was doubly distilled over alkaline permanganate and deionized.

An ultrasonic interferometer (Mittal Enterprises, New Delhi, India) is a simple and direct device to determine the ultrasonic velocity in liquids with a high degree of accuracy. The principle used in the measurement of velocity (U) is based on the accurate determination of the wavelength (λ) in the medium. Ultrasonic waves of known frequency (f) are produced by a quartz plate fixed at the bottom of the cell. The waves are reflected by a movable metallic plate kept parallel to the quartz plate. If the separation
between these plates is exactly a whole multiple of the sound wavelength, standing waves are formed in the medium. The acoustic resonance gives rise to an electrical reaction on the generator driving the quartz plate and the anode current of the generator becomes maximum.

Ultrasonic velocity of soya protein was measured at frequencies of 3 and 10 MHz with varying the concentration (C) at temperature of 30°C. The viscosity (η) of the sample solution was measured at 30°C with an Ubbelohde suspended level viscometer. The density (ρ) of the sample solution was determined with a specific gravity bottle with a 25 ml capacity.

The ultrasonic acoustic parameters were calculated using the standard formulae as follows.

(a) Ultrasonic velocity (υ) — If the distance is increased or decreased and the variation is exactly one half wavelengths (λ/2) or multiple of it, anode current again becomes the maximum.

\[ υ = \frac{λ}{2} \times f \]

where λ is the wavelength in mm and f is frequency in MHz

(b) Adiabatic compressibility (βs) — \[ β_s = \frac{1}{ρυ^2} \], where ρ is the density and υ is the ultrasonic velocity

(c) Acoustic impedance (Z) — \[ Z = ρ \times υ \], where ρ is the density and υ is the ultrasonic velocity

(d) Viscosity relaxation time (τ) — \[ τ = \frac{4η}{3ρυ^2} \], where ρ is the density and η is the viscosity

### 3 Results and Discussion

The values of ultrasonic velocity (υ) in viscosity (η) and density (ρ) of soya protein are presented in Table 1. To understand the structural effect and molecular interactions occurring in aqueous solution of soya protein, several acoustic and thermodynamical parameters were derived from different experimental results of ultrasonic velocities, viscosities and densities. The variation of density in aqueous solution of soya protein is shown in Fig. 1. It is found that density of soya protein increases gradually with increasing concentration of the solution in the range 0.01-0.1 (w/v) %.

In Fig. 2, viscosities of soya protein progressively have been found to increase with variation of concentrations. It is noticed that viscosity of the solution increases by 10% with increasing concentration 0.01-0.1 (w/v) %. This increase in viscosity with the rise of concentration may be due to the enhanced tendency of protein molecule to associate in the protein-water system.

Ultrasonic velocities were measured with frequency of ultrasound i.e. at 3 and 10 MHz with variation

Table 1 — Comparative values of ultrasonic velocity υ at frequency of 3 and 10 MHz, density (ρ), viscosity (η), adiabatic compressibility (βs), acoustic impedance (Z), viscosity relaxation time (τ) for soya protein at different concentrations (w/v%)

<table>
<thead>
<tr>
<th>C (w/v)%</th>
<th>ρ × 10^3 Kg/m^3 Unit</th>
<th>η, SI Unit</th>
<th>υ, MHz</th>
<th>Z, SI unit</th>
<th>τ × 10^7 min</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.9562</td>
<td>1</td>
<td>1480</td>
<td>1415.17</td>
<td>6.36</td>
</tr>
<tr>
<td>0.01</td>
<td>0.9570</td>
<td>1.023</td>
<td>1512</td>
<td>1446.98</td>
<td>6.23</td>
</tr>
<tr>
<td>0.025</td>
<td>0.9583</td>
<td>1.058</td>
<td>1518</td>
<td>1379.98</td>
<td>7.27</td>
</tr>
<tr>
<td>0.05</td>
<td>0.9598</td>
<td>1.079</td>
<td>1524</td>
<td>1379.98</td>
<td>7.09</td>
</tr>
<tr>
<td>0.075</td>
<td>0.9613</td>
<td>1.089</td>
<td>1530</td>
<td>1470.78</td>
<td>6.45</td>
</tr>
<tr>
<td>0.1</td>
<td>0.9639</td>
<td>1.126</td>
<td>1540</td>
<td>1480.40</td>
<td>6.36</td>
</tr>
</tbody>
</table>

Fig. 1 — Variation of densities with concentration in aqueous solutions of soya protein
of concentration of the solution. At both the frequencies, ultrasonic velocities increase gradually with increase of concentrations. However, increase of velocity at low frequency i.e. 3 MHz is proportionally less than the velocity at high frequency. That means the plot at frequency of 3 MHz is stiffer as compared to the velocity at 10 MHz (Fig. 3). This may be due to active interaction of ultrasound wave with the aqueous solution of soya protein. The protein chain may have hindered rotation or conformational inter conversion that occurred at high frequency of sound wave. At all concentrations, the velocities vary almost linearly which indicated the compatible nature of protein in aqueous solution.

The protein chain assumes a variety of conformational changes under different experimental conditions. Protein chains are more compressible because of their chain like structure. It can be shown by a decrease in adiabatic compressibility with increase in concentration (Fig. 4). This result was verified at both frequency of concentration (3 and 10 MHz) with different values of concentration.

Comparing the results at frequency 3 and 10 MHz, it is assumed that the acoustic sound wave was perturbed at 3 MHz, the segmental motion of soya protein chain was in such a way as to cause rearrangement of the water molecules which are weakly interacting with the protein chain. This result
was in good agreement with the result obtained due to the ultrasonic relaxation in aqueous solution of dextran and PVA-Dextran bending.

In Fig. 5, the acoustic impedance (Z) of soya protein increase with increase in concentration at both frequency of ultrasound. The increasing trend of Z with concentration could be interpreted on the basis that most of the solvent molecules were engaged in interaction with them by cross-linking via hydrogen bond and compatible in all range of concentration and ultrasound.

The variation of τ with concentration is shown in Fig. 6. It is found that viscosity relaxation time decreases with concentration at ultrasonic frequency of 10 MHz. But at low frequency ultrasound of 3 MHz, the value of τ increases linearly with concentration. This indicates that the interaction and compatibility of water-protein solution were prominent via structural change.

4 Conclusions
The ultrasonic results are described in this paper with acoustic thermodynamical magnitudes of the interaction parameters between the protein and the water. So a remarkable compatibility in aqueous solution in all range of concentration, the interesting and significant outcome of our study may enable the development of pharmaceutically active molecules and various biomedical applications.

Acknowledgement
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