Conclusions

We have shown evidence for the bridging flocculation of PSL by β-LG. The change in surface charge of the PSL spheres resulting from adsorption of β-LG molecules correlates with the formation of aggregates bridged by protein molecules. To develop models of the kinetics of bridging flocculation, some measurements of aggregation as a function of time and temperature have been carried out and will be reported elsewhere. Future work will aim to describe the kinetics of aggregation quantitatively and the diffusion of PSL in more highly aggregated (pre-gel) and gelled whey protein systems will be investigated using p.c.s. and forced Rayleigh scattering.

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Gel permeation chromatography–multi-angle laser light scattering characterization of the molecular mass distribution of 'Pronova' sodium alginate

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A relatively recent innovation in total intensity laser light scattering has been to replace the isolated sample cell containing a quantity of solution with a cell in the form of a glass block with a channel of narrow bore through which the solution flows at a constant rate. If this solution has just passed through a suitable gel permeation chromatography (g.p.c.) column (upstream of the block of glass), then it will have been fractionated according to molecular size. This means that, provided the light scattering and associated equipment can respond quickly enough, the distribution of molecular masses (as well as just the average molecular mass) can be measured. Such an experimental arrangement is easier to realise in practice if the traditional 'one photomultiplier tube' is replaced by an array of light-detecting diodes. Not surprisingly, such an apparatus is best controlled by a computer and the resulting data analysed with a suite of software on the computer.

We have used a Wyatt Technology Dawn F multi-angle laser light-scattering (MALLS) photometer [1] to examine the molecular mass distribution of sodium alginate. A sample, known commercially as 'Pronova', was obtained from Protan of Drammen, Norway. Solutions of the sample were made up in phosphate–chloride buffers of ionic strengths (I) of 0.1 M and 0.3 M with concentrations (c) in the range 0.5–5.0 mg/ml. These same buffers were then used as the mobile phase to wash the sample from the rheodyne, through the g.p.c. column and into the light-scattering cell. It is necessary for the analysis of the data that the refractive index (n) of the solution be monitored and this is done by placing a differential refractometer in series after the light-scattering cell. We have used the value of $dn/dc = 0.154$ ml/g, as measured by Mackie *et al.* [2]. (These authors show that their value is consistent with many others quoted in the literature.)

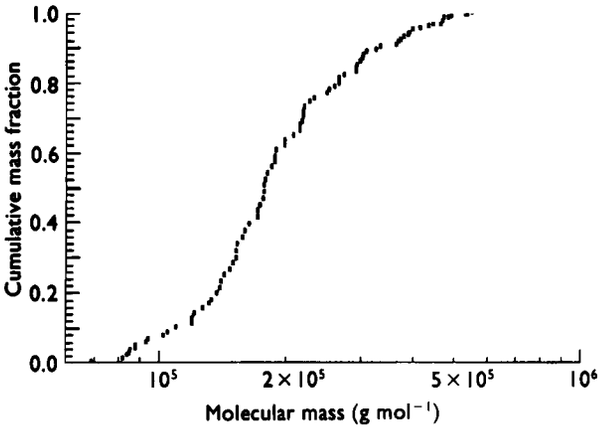
Although it is common to talk about the 'loading concentration' of the solution as it is injected into the system, it must be remembered that when the solution meets the running phase, it is diluted considerably. (This 'loading concentration' does not carry the same connotation as it does in some other fields, such as analytical ultracentrifugation.) This aspect should be compared with some other methods (such as sedimentation equilibrium ultracentrifugation), where the strong non-ideality of a substance such as alginate (and other polysaccharides) means that a value of molecular mass independent of non-ideality can only be obtained by making a number of measurements at different concentrations and extrapolating the apparent molecular mass back to zero concentration. Clearly, this procedure is not necessary with the g.p.c./MALLS

Abbreviations used: g.p.c., gel permeation chromatography; MALLS, multi-angle laser light scattering.

Fig. 1

Cumulative Molecular Mass distribution

Cumulative molecular mass distribution of sodium alginate at a concentration, $c \approx 1$ mg/ml in phosphate chloride buffer of ionic strength, $I_s = 0.3$ M.



technique because of the low concentrations involved. Another advantage of this technique is that the g.p.c. columns filter the samples in the sense that dust and aggregates are separated from the sample before it reaches the light-scattering cell. (This advantageous feature is shared with analytical ultracentrifugation.) The software which performs the data analysis allows for the effect of the second virial coefficient, B. We set B to 7×10^{-3} ml mol g^{-2} as measured by Martinsen *et al.* [3]. This gives values of the mass average molecular mass of our 'Pronova' sample to be (232 ± 6) kg mol^{-1} and (236 ± 2) kg mol^{-1} for $I = 0.1$ M and $I = 0.3$ M, respectively. Ignoring the effect of the virial coefficient and setting B to zero gives values of (196 ± 9) kg mol^{-1} and (203 ± 6) kg mol^{-1} respectively.

Associated with the program ASTRA, which runs the Dawn F and processes the raw data, is another program called EASI. This analyses the processed data to produce molecular mass distributions. Fig. 1 shows a typical cumulative molecular mass distribution (for $c \approx 1$ mg/ml and $I = 0.3$ M).

We have thus seen that g.p.c./MALLS is a technique which is able to measure both the average molecular mass and the molecular mass distribution of even strongly non-ideal materials from just the one concentration.

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Photon correlation spectroscopy study of a 2311 bp relaxed circular DNA — applicability of Rouse–Zimm and wormlike chain models

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Introduction

Circular polymers are of considerable theoretical interest since models for the dynamics of flexible and semi-flexible polymers can substantially be simplified for circular chains compared with their linear counterparts [1, 2]. Especially monodisperse circular chains are therefore good candidates to test the predictions of various theoretical models. We measured the dynamics of a monodisperse, relaxed circular DNA with 2311 bp by means of photon correlation spectroscopy (p.c.s.) in dilute solution. Scattering vector (q)-dependent measurements revealed both translational and internal motions at two different ionic strengths for this molecule.

Abbreviation used: p.c.s., photon correlation spectroscopy.

Results

The frequency distribution functions obtained by an inverse Laplace transform of the first-order $g_1(q,t)$ correlation functions show a q^2-dependent peak that can be attributed to the translational motion of the chain. The amplitude of a second peak in the frequency spectra increases with increasing q as predicted for internal modes [3]. The translational diffusion coefficient of the circular chain is in agreement with the predictions of the circular wormlike cylinder model of Fujii & Yamakawa [4], if a persistence length of 45 nm in salt solution (0.2 mol of NaCl/l) and a cylinder diameter of 2.5 nm is assumed. These values have been found to be typical for linear DNAs [5].

To compare the obtained correlation functions with theoretical predictions, we calculated