radii of gyration and their diffusion coefficients will be determined, allowing for the construction and subsequent refinement of hydrated bead models.

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Hydrodynamic characterization of *Chromobacter viscosum* lipase

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Lipases have growing commercial potential in the food and soap industries. For example, in detergent formulations it is considered that they may be effective, particularly at low washing temperatures, in removing fatty soil [1]. Microbial lipases exhibit molecular masses generally in the range 25000–50000 with some higher values reported [2, 3]. Very recently, the high resolution crystallographic X-ray structure of *Mucor miehei* phase (M_r ~ 29500) has appeared [4].

In this study, we consider the hydrodynamic properties of the lipase from *Chromobacter viscosum* in terms of (i) molecular mass in solution and (ii) sedimentation velocity behaviour. It has been the particular intention of this study to examine the possibility of self-association for *C. viscosum* lipase. Self-association has been suggested (possibly via a hydrophobic interaction) for *Aspergillus* sp. lipase [5] and, we believe, is also a possibility for *Geotrichum candidum* lipase [6].

Sedimentation velocity experiments were performed using an MSE Centriscan analytical ultracentrifuge equipped with scanning absorption and Schlieren optics, and the $s_{20,w}$ determined in the usual way [7]. Low-speed sedimentation equilibrium experiments were performed using a Beckmann Model E analytical ultracentrifuge equipped with a 5 mW He–Ne laser light source and Rayleigh interference optics. A low loading concentration of ~ 0.8 mg/ml was used throughout to minimize the possibility of thermodynamic non-ideality effects. 'Whole cell weight average' molecular masses, $M^0_{r,w}$, were determined according to the procedure of Creeth & Harding [8]. A value for $\bar{v}$ of 0.73 ml/g, calculated from the amino acid composition [9], was employed throughout.

The following values were obtained for $M^0_{r,w}$: 35000 ± 2000 in a phosphate chloride buffer, pH 6.8, I 0.1 M at 23.8°C; 35000 ± 2000 in a 40% (w/w) 1,4-dioxan/phosphate chloride buffer mixture, pH 6.8, I 0.1 M at 23.8°C and 40000 ± 2000 in phosphate chloride buffer at 10°C. These values are in agreement with molecular masses determined independently via SDS/PAGE (36000) and using the empirical equation of Squire & Himmel [10] from the infinite dilution sedimentation coefficient, $s^0_{20,w}$, of 3.17S (38700) (Fig. 1). The value for $s^0_{20,w}$ was obtained from a linear least-squares analysis of the data in Fig. 1. Taken in isolation these results appear to indicate that *C. viscosum* lipase has a molecular mass of 38000 ± 2000. However, close inspection of Fig. 1 may suggest, from the positive slope at low concentration, some self-association behaviour, although the absence of an observed decrease in $M^0_{r,w}$ for the 1,4-dioxane and low-temperature buffer systems would appear to suggest that the association, if present, is not hydrophobic in nature.

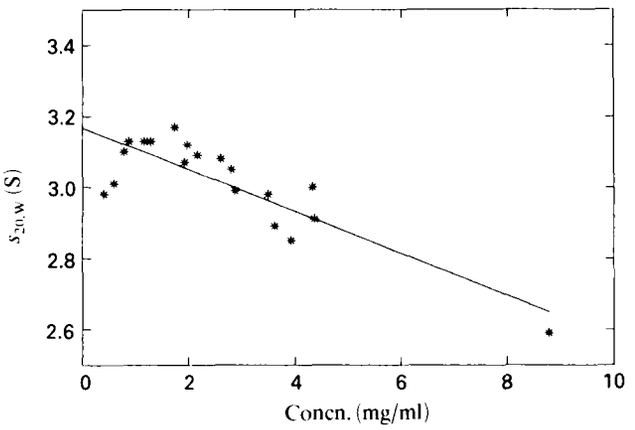

Fig. 1. *Sedimentation velocity profile for C. viscosum lipase*

Sedimentation velocity experiments were performed in a phosphate chloride buffer, pH 6.8, I 0.1 M, at 20.0°C, using a rotor speed of 40000 rev./min.

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