19.5. Fibre diffraction

BY R. CHANDRASEKARAN AND G. STUBBS

19.5.1. Introduction

Many biopolymers are long helical structures and have a natural tendency to form fibres. This tendency severely impedes the growth of single crystals from these polymers, and even if crystals can be grown, the molecular interactions in the crystals rarely correspond to the biologically significant interactions in the fibres. Conventional macromolecular crystallography is therefore often not applicable to these systems. Fibre diffraction, however, is a powerful technique for determining the structural details of such polymers. It has been used to study a wide variety of biopolymers, ranging from simple polypeptides, polynucleotides and polysaccharides to complex filamentous viruses and cytoskeletal filaments.

Fibres can have relatively high degrees of order, although falling short of true three-dimensional crystallinity. The key difference between fibres and crystals, however, is that in fibres the fundamental structural aggregates, although parallel to each other, are randomly oriented about the fibre axis. Consequently, the diffraction pattern is cylindrically averaged. This cylindrical averaging is the defining characteristic of fibre diffraction.

On the basis of this definition, fibre diffraction may also be considered to include diffraction from many biological membrane specimens, and much of fibre-diffraction theory also applies to membrane diffraction. In general, however, the diffracting units in fibres have helical symmetry, whereas those of membranes do not.

In addition to the loss of information due to cylindrical averaging, fibre-diffraction patterns reflect a generally limited degree of order and rarely extend beyond 3 Å resolution. Consequently, the number of data obtainable from a fibre is considerably less than that from a single crystal having a similar size of asymmetric unit. The use of stereochemical information to supplement the diffraction data is therefore essential. For polymers with small asymmetric units, such as polynucleotides, structural chemical information can be used to construct models consistent with the helical parameters and molecular dimensions obtained from the diffraction data. For the larger asymmetric units found in aggregates, such as viruses, initial models must be constructed in other ways. However, in all cases the combination of diffraction data and stereochemistry can be used to refine both molecular structures and packing parameters. Refinement in this way is very similar to that used in macromolecular crystallography, but because of the limited number of experimental data, stereochemical restraints are particularly important in fibre diffraction. As in crystallography, difference-electron-density maps are used in conjunction with refinement to identify missing portions and determine the correctness of the models and, in favourable cases, to locate ions and solvent molecules associated with the polymers.

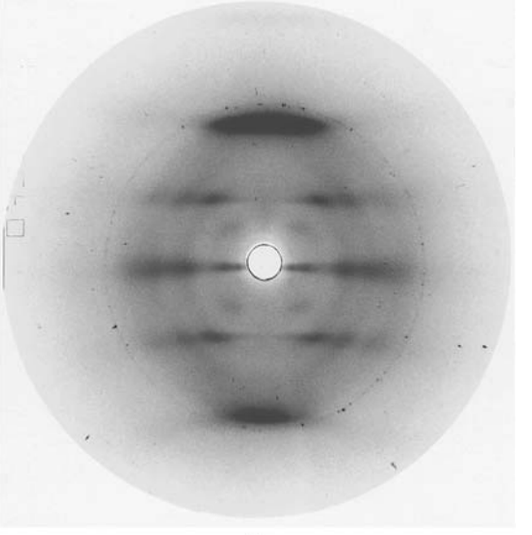
19.5.2. Types of fibres

Fibres fall into essentially two classes with respect to the degree of ordering of the polymer chains. Within each class, there are varying degrees of disorder; furthermore, many fibres exhibit properties intermediate between those of the two ideal classes.

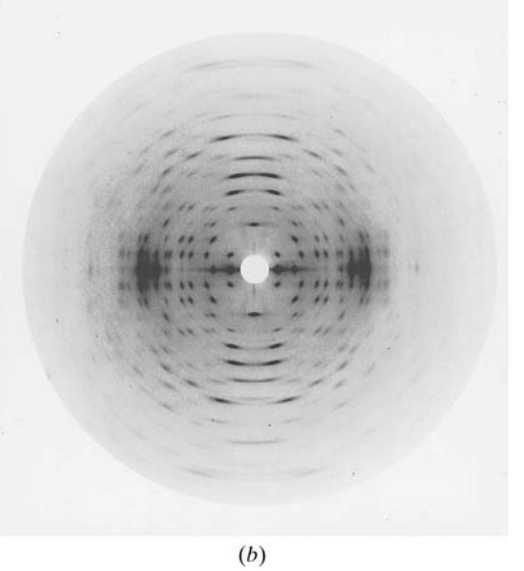
In noncrystalline fibres, the polymers are parallel to each other, but their positions and orientations are otherwise uncorrelated. Diffraction patterns from these fibres are confined to layer lines (Fig. 19.5.2.1*a*) because of the repeating nature of the polymer helix, but are otherwise continuous and correspond to the cylindrical average of the Fourier transform of a single particle.

In polycrystalline fibres, the polymers form fully ordered microcrystallites, and each fibre consists of many such microcrystallites, randomly oriented about the fibre axis. In diffraction patterns from polycrystalline fibres, the layer lines are sampled to form discrete reflections (Fig. 19.5.2.1*b*); the diffraction pattern is the cylindrical average of a single-crystal diffraction pattern and is, in fact, equivalent to the diffraction pattern that would be obtained from a rotating single crystal.

Polycrystalline fibres may be disordered in various ways. For example, the helical polymers may be subject to rotational or translational disorder, and this disorder may be partial (a small number of alternative packings for each particle) or complete (for example, completely random rotational particle orientations). Rotational disorder may be coupled to translational disorder (screw disorder). The resulting diffraction patterns may contain both discrete reflections and continuous diffraction along layer lines; depending upon the type of disorder, the discrete reflections



(*a*)



(*b*)

Fig. 19.5.2.1. X-ray diffraction patterns showing (*a*) continuous intensity on layer lines from an oriented nucleic acid fibre and (*b*) Bragg reflections from an oriented and polycrystalline polysaccharide fibre.

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