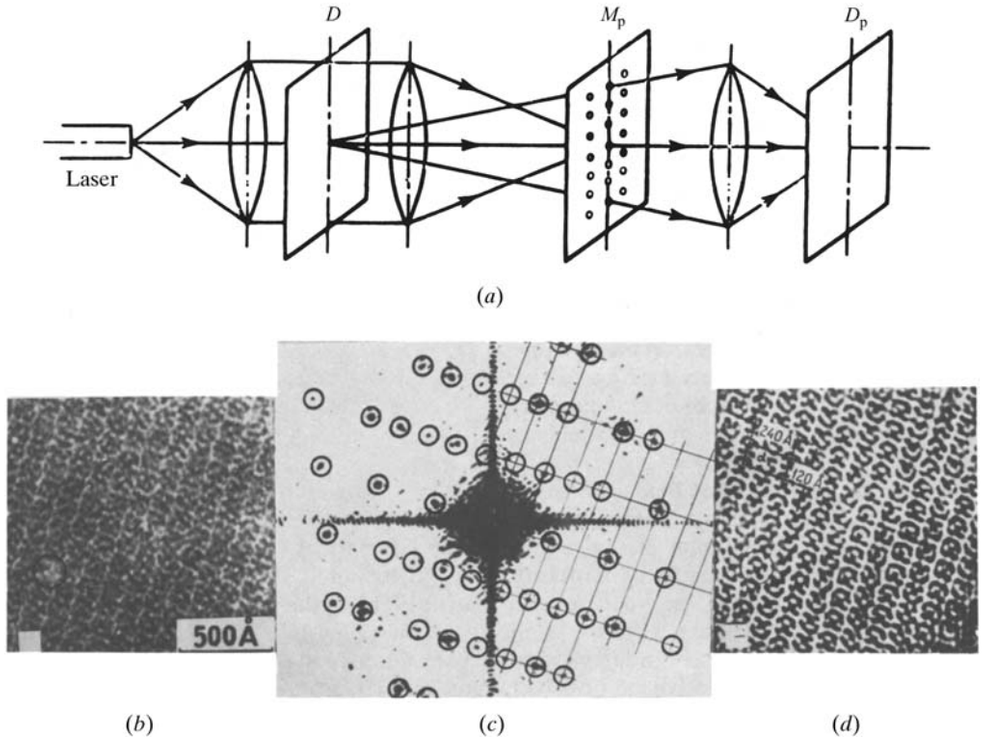

Fig. 2.5.5.2. (*a*) Diagram of an optical diffractometer. *D* is the object (an electron micrograph), M_p is the diffraction plane and a mask that transmits only Φ_{hk}, D_p is the plane of the (filtered) image; (*b*) an electron micrograph of a crystalline layer of the protein phosphorylase *b*; (*c*) its optical diffraction pattern (the circles correspond to the windows in the mask that transmits only the Φ_{hk} diffracted beams from the periodic component of the image); (*d*) the filtered image. Parts (*b*)–(*d*) are based on the article by Kiselev *et al.* (1971).

diffraction, and phases from electron microscopy, and so on (Gurskaya *et al.*, 1971).

Images with point symmetry. If a projection of an object (and consequently, the object itself) has a rotational *N*-fold axis of symmetry, the structure coincides with itself on rotation through the angle $2\pi/N$. If the image is rotated through arbitrary angles and is aligned photographically with the initial image, then the best density coincidence will take place at a rotation through $\alpha = (k2\pi/N)$ $(k = 1, \ldots, N)$ which defines *N*. The pattern averaging over all the rotations will give the enhanced structure image with an $(N)^{1/2}$ times reduced background (Markham *et al.*, 1963).

Rotational filtering can be performed on the basis of the Fourier expansion of an image in polar coordinates over the angles (Crowther & Amos, 1971).

$$I(r, \psi) = \sum_{n=-\infty}^{+\infty} g_n(r) \exp(in\varphi). \qquad (2.5.5.23)$$

The integral over the radius from azimuthal components g_n gives their power

$$p_n \sim \int_0^a |g_n|^2 r \, \mathrm{d}r, \qquad (2.5.5.24)$$

where *a* is the maximum radius of the particle. A set p_n forms a spectrum, the least common multiple *N* of strong peaks defining the *N*-fold symmetry. The two-dimensional reconstructed image of a particle with rotational symmetry is defined by the synthesis (2.5.5.24) with $n = 0, N, 2N, 3N$.

Asymmetric images. In this case, a set of images is processed by computational or analogue methods. The initial selection of images involves the fulfilment of the maximum similarity condition.

The averaging of *n* images in real space gives

$$I_{\mathrm{enh}} = (1/n) \sum_{k=1}^{n} J_k(xy) = \langle I_k \rangle (xy) + (1/n) \sum N_k(xy). \qquad (2.5.5.25)$$

The signal/noise ratio on an average image is $(n)^{1/2}$ times enhanced.

The degree of similarity and accuracy of superposition of two images with an account both of translational and angular shifts is estimated by a cross-correlation function[4] of two selected images J_1 and J_2 (Frank, 1975, 1980).

$$\begin{aligned} k(\mathbf{x}') = J_1 * J_2 &= \int J_1(x) J_2(x + x') \, \mathrm{d}x \\ &= k_{I_1 I_2} + k_{I_1 N_2} + k_{I_2 N_1} + k_{N_1 N_2}. \end{aligned} \qquad (2.5.5.26)$$

The value $k(\mathbf{x}')$ is the measure of image similarity, the x' coordinate of the maximum indicates the shift of the images relative to each other. The first term of the resultant expression (2.5.5.26) is the cross-correlation function of noise-corrected images being compared, the second and third terms are approximately equal to zero, since the noise does not correlate with the signal; the last term is the autocorrelation function of the noise (Cramér, 1954; Frank, 1975, 1980).

The calculation of a correlation function is performed by means of Fourier transformation on the basis of the convolution theorem, since the Fourier transformation of the product of the Fourier transform of function J_1 and the conjugated Fourier transform function J_2 gives the cross-correlation function of the initial functions:

[4] At $I_j = I_k$ this is the autocorrelation function, an analogue of the Patterson function used in crystallography.