Acta Cryst. (1964). **17**, 611

The differential rotation function.

By Yoshio Sasada*, *Medical Research Council Laboratory of Molecular Biology, Hills Road, Cambridge, England*

(*Received* 25 *September* 1963)

Rossmann & Blow (1962) proposed the rotation function in order to find the relative orientation between sub-units in one crystal or between molecules in different crystal forms. They applied this method to haemoglobin (Rossmann & Blow, 1962), insulin (Hodgkin, Harding, Rossmann & Blow, 1962) and α-chymotrypsin (Blow, Rossmann & Jeffrey, 1963) with successful results. However, this method takes a considerable length of time for computation. Rossmann & Blow estimated the computation time to be about 50 hours on EDSAC II for a crystal of horse haemoglobin if they explored the whole range of Eulerian angles with 20° intervals using 1000 reflexions. They cut down their computing time by using larger intervals with a limited number of reflexions to find the approximate peak positions, and then refined these with closer angular intervals and more reflexions.

In general, the background region of the rotation function has little physical significance. Hence, once the rough peak position has been determined, the major interest is to determine the accurate angular coordinates for which the rotation function has the maximum values. This situation suggests that a technique similar to the differential synthesis for the usual electron density distribution (Booth, 1946) can be applied to the present problem.

Let us assume that an approximate rotation between two identical units is known and is represented by angular coordinates θ_1, θ_2 and θ_3. Let $\Delta\theta_1$, $\Delta\theta_2$ and $\Delta\theta_3$ be the small angular displacements of this point from the true maximum of the rotation function. If they are small, then

$$\left(\frac{\partial^2 R}{\partial\theta_1^2}\right)\Delta\theta_1+\left(\frac{\partial^2 R}{\partial\theta_1\partial\theta_2}\right)\Delta\theta_2+\left(\frac{\partial^2 R}{\partial\theta_1\partial\theta_3}\right)\Delta\theta_3+\left(\frac{\partial R}{\partial\theta_1}\right)=0 \text{ etc.},$$

where $(\partial R/\partial\theta_1)$ and $(\partial^2 R/\partial\theta_1^2)$ *etc.* are the first and second derivatives of the rotation function at $(\theta_1, \theta_2, \theta_3)$.

The rotation function is defined as

$$R=\int^U \varrho(\mathbf{x})\varrho(\mathbf{x}')d\mathbf{x} \tag{1}$$

or

$$R=\int^U \varrho(\mathbf{x})\varrho(\mathbf{x}')\exp\left[-a\mathbf{x}^2\right]d\mathbf{x} \tag{2}$$

where $\mathbf{x}'$ and $\mathbf{x}$ are related by the rotation $[C]$ according to the relationship

$$\mathbf{x}'=[C]\mathbf{x}.$$

The function (2) has been called the 'shaded rotation function' (Rossmann, 1962). It was designed to increase the peak to background ratio. If U is taken as a sphere centred on the origin, then the rotation function can be written:

$$R=\sum_{\mathbf{h}}\sum_{\mathbf{p}}|F_{\mathbf{h}}|^2|F_{\mathbf{p}}|^2 G_{\mathbf{hp}}\,.$$

Now G is the function of the distance, H, between the origin and the point $(\mathbf{h}+\mathbf{h}')$, where

$$\mathbf{h}'=[\tilde{C}]\mathbf{p}\,,$$

and

$$G_{\mathbf{hp}}=\frac{3\,(\sin 2\pi Hr-2\pi Hr\cos 2\pi Hr)}{(2\pi Hr)^3}$$

for the usual rotation function or

$$G_{\mathbf{hp}}=\frac{1}{Hr}\int_0^1 r'\exp\left[-ar'^2\right]\sin 2\pi Hrr'dr'$$

for the shaded rotation function. The first derivative with respect to θ is then

$$\left(\frac{\partial R}{\partial\theta_1}\right)=\sum_{\mathbf{h}}\sum_{\mathbf{p}}|F_{\mathbf{h}}|^2|F_{\mathbf{p}}|^2\left(\frac{r}{2H}\right)\left\{\frac{3}{Hr}\left(\frac{\sin 2\pi Hr}{2\pi Hr}-G\right)\right\}\left(\frac{\partial H^2}{\partial\theta_1}\right)$$

for the usual rotation function, or

$$\left(\frac{\partial R}{\partial\theta_1}\right)=\sum_{\mathbf{h}}\sum_{\mathbf{p}}|F_{\mathbf{h}}|^2|F_{\mathbf{p}}|^2\left(\frac{r}{2H}\right)\left\{\frac{-A+2\pi HrB}{(Hr)^2}\right\}\left(\frac{\partial H^2}{\partial\theta_1}\right)$$

for the shaded rotation function, where

$$A=\int_0^1 r'\exp\left[-ar'^2\right]\sin 2\pi Hrr'dr'\,,$$

$$B=\int_0^1 r'^2\exp\left[-ar'^2\right]\cos 2\pi Hrr'dr'\,,$$

$$\left(\frac{\partial H^2}{\partial\theta_1}\right)=2\sum_{i=1}^{3}\left\{a_i^{*2}(h_i+h_i')+\sum_{i\neq j\neq k}^{3}\sum a_i^*a_j^*\cos\alpha_k^*(h_j+h_j')\right\}\left(\frac{\partial h_i'}{\partial\theta_1}\right),$$

$$\left(\frac{\partial h_i'}{\partial\theta_1}\right)=\sum_{j=1}^{3}\left(\frac{\partial C_{ji}}{\partial\theta_1}\right)p_j$$

and a_i^*, α_i^* $(i=1\sim 3)$ are the dimensions of the reciprocal unit cell. Similar expressions can be written for $(\partial R/\partial\theta_2)$ and $(\partial R/\partial\theta_3)$.

The second derivative is given by

$$\left(\frac{\partial^2 R}{\partial\theta_1\partial\theta_2}\right)=\sum_{\mathbf{h}}\sum_{\mathbf{p}}|F_{\mathbf{h}}|^2|F_{\mathbf{p}}|^2\left[\left(\frac{r}{2H}\right)\left\{\frac{3}{Hr}\left(\frac{\sin 2\pi Hr}{2\pi Hr}-G\right)\right\}\times\left(\frac{\partial^2 H^2}{\partial\theta_1\partial\theta_2}\right)+\left(\frac{r^2}{4H^2}\right)\left\{\frac{3}{(Hr)^2}\left(5G-6\frac{\sin 2\pi Hr}{2\pi Hr}+\cos 2\pi Hr\right)\right\}\times\left(\frac{\partial H^2}{\partial\theta_1}\right)\left(\frac{\partial H^2}{\partial\theta_2}\right)\right]$$

for the usual rotation function, or

$$\left(\frac{\partial^2 R}{\partial\theta_1\partial\theta_2}\right)=\sum_{\mathbf{h}}\sum_{\mathbf{p}}|F_{\mathbf{h}}|^2|F_{\mathbf{p}}|^2\left[\left(\frac{r}{2H}\right)\left\{\frac{-A+2\pi HrB}{(Hr)^2}\right\}\left(\frac{\partial^2 H^2}{\partial\theta_1\partial\theta_2}\right)+\left(\frac{r^2}{4H^2}\right)\left\{\frac{3A-6\pi HrB-4\pi^2(Hr)^2C}{(Hr)^3}\right\}\left(\frac{\partial H^2}{\partial\theta_1}\right)\left(\frac{\partial H^2}{\partial\theta_2}\right)\right]$$

for the shaded rotation function, where

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* Present address: Institute for Protein Research, Osaka University, Kita-ku, Osaka, Japan.

$$C = \int_0^1 r'^3 \exp\left[-ar'^2\right] \sin 2\pi Hrr'dr' \,,$$

$$\left(\frac{\partial^2 H^2}{\partial\theta_1\partial\theta_2}\right) = 2\sum_{i=1}^{3}\Bigg[\left\{a_i^{*2}(h_i+h_i') + \mathop{\sum\sum}_{i\neq j\neq k}^{3} a_i^* a_j^* \cos \alpha_k^*(h_i+h_j')\right\} \times \left(\frac{\partial^2 h_i'}{\partial\theta_1\partial\theta_2}\right) + a_i^{*2}\left(\frac{\partial h_i'}{\partial\theta_1}\right)\left(\frac{\partial h_i'}{\partial\theta_2}\right) + a_j^* a_k^* \cos \alpha_i^* \left\{\left(\frac{\partial h_j'}{\partial\theta_1}\right)\left(\frac{\partial h_k'}{\partial\theta_2}\right) + \left(\frac{\partial h_k'}{\partial\theta_1}\right)\left(\frac{\partial h_j'}{\partial\theta_2}\right)\right\}\Bigg]$$

and

$$\left(\frac{\partial^2 h_i'}{\partial\theta_1\partial\theta_2}\right) = \sum_{j=1}^{3}\left(\frac{\partial^2 C_{ji}}{\partial\theta_1\partial\theta_2}\right) p_j \,.$$

It is necessary to examine the nature of the first and second derivatives of the rotation function. As $(\partial H^2/\partial\theta)$ is a function of $(\mathbf{h}+\mathbf{h}')$ and $\mathbf{p}$, $|F_{\mathbf{p}}|^2(r/H)(\partial H^2/\partial\theta)$ as a whole does not change so drastically with H and $\mathbf{p}$. Therefore, it is concluded that the change of $(\partial R/\partial\theta)$ with Hr is controlled mainly by the factor

$$\frac{3}{Hr}\left(\frac{\sin 2\pi Hr}{2\pi Hr} - G\right)$$

for the usual rotation function or

$$\frac{-A + 2\pi HrB}{(Hr)^2}$$

for the shaded rotation function. These two factors are plotted against Hr in Fig. 1 by full and dotted lines, respectively. We now see that the values of the first derivatives do not exceed about 5% of their maximum value if Hr is larger than 1·8. Similar consideration for the second derivatives showed that they are controlled by the factor

$$\frac{3}{(Hr)^2}\left(4G - 5\frac{\sin 2\pi Hr}{2\pi Hr} + \cos 2\pi Hr\right)$$

for the usual rotation function or

$$\frac{2A - 4\pi HrB - 4\pi^2(Hr)^2C}{(Hr)^3}$$

for the shaded rotation function. These are also shown in Fig. 1. The values for the second derivatives do not exceed 5% of their maximum values when Hr is greater than 2·0. These properties of the derivatives suggest that they may be evaluated by considering only a few of the terms in the summation over $\mathbf{p}$, as is also true for the calculation of the rotation function itself. It is obvious from Fig. 1 that the shaded rotation function is more favourable than the usual one with respect to this point.

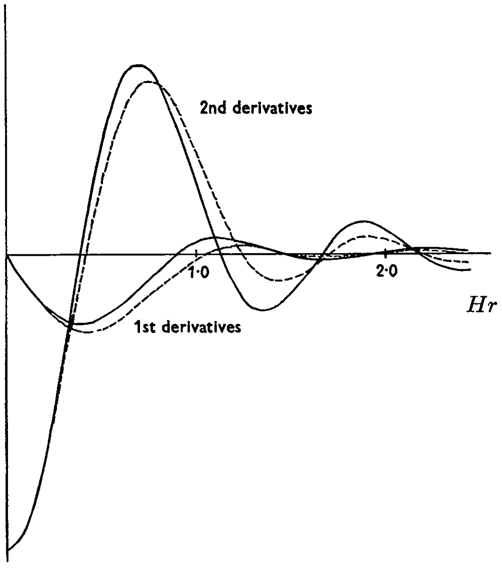

Fig. 1. Variation of the first and second derivatives of the rotation function with Hr. Full lines for the usual and dotted for the shaded rotation function, on a correctly normalized scale.

A program for the differential rotation function was written for the IBM 7090 computer. It was tested on the 10Å chymostrypin data, in order to determine the accurate angular relationship between the two independent crystallographic molecules in the asymmetric unit of the $P2_1$ lattice. The progress of a typical refinement is given in Table 1. The time for one cycle using about 280 independent reflexions and 27 terms in the summation over $\mathbf{p}$ was about 1·75 minutes. An exploration of the peak area by the evaluating the rotation function at say 30 points might have taken half an hour.

Table 1. *An example of progress of a refinement by the differential rotation function, using* 10Å *α-chymotrypsin data*

The shaded rotation function was used with a radius of 30 Å and $a = 2·0$. The total refinement required 7·76 minutes

Cycle number	$\varkappa$	ψ	φ	R
0	175·0	85·0	85·0	—
1	179·4	86·5	88·2	$0·5478 \times 10^9$
2	179·7	88·0	88·7	$0·5709 \times 10^9$
3	180·1	89·9	89·4	$0·5856 \times 10^9$
4	180·0	90·0	89·3	$0·5984 \times 10^9$

(The exploration suggests the maximum at $\varkappa = 180·0$, $\psi = 90·0$, $\varphi = 89·0$).

I wish to express my sincere thanks to Dr M. F. Perutz for his encouragement, to Dr M. G. Rossmann for suggestion of this problem and for his help and discussions throughout this investigation, and to Drs D. M. Blow & M. G. Rossmann for permission to use the α-chymotrypsin data. This study, made in Cambridge, was made possible by a grant from the Rockefeller Foundation.

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