$$\sigma_{\mathrm{LS,\,Luzz}}(r) = 1.33(N_i/p)^{1/2}[R(s_m)/s_m], \qquad (18.5.8.3)$$

where $R(s_m)$ is the value of R at some value of $s = s_m$ on the selected Luzzati curve. Equation (18.5.8.3) provides a means of making a very rough statistical estimate of error for an atom with $B = B_{\mathrm{avg}}$ (the average B for fully occupied sites) from a plot of R *versus* $2\sin\theta/\lambda$.

The corresponding equation involving R_{free} is

$$\sigma_{\mathrm{LS,\,Luzz}}(r) = 1.33(N_i/n_{\mathrm{obs}})^{1/2}[R_{\mathrm{free}}(s_m)/s_m]. \qquad (18.5.8.4)$$

18.5.8.3. *Comments on Luzzati plots*

Protein structures always show a great range of B values. The Luzzati theory effectively assumes that all atoms have the same B. Nonetheless, the Luzzati method applied to high-angle data shells does provide an upper limit for $\langle\Delta r\rangle$ for the atoms with low B. It is an upper limit since experimental errors and model imperfections are not allowed for in the theory.

Low-resolution structures can be determined validly by using restraints, even though the number of diffraction observations is less than the number of atomic coordinates. The Luzzati method, based preferably on R_{free}, can be applied to the atoms of low B in such structures. As the number of observations increases, and the resolution improves, the Luzzati $\langle\Delta r\rangle$ increasingly overestimates the true $\sigma(r)$ of the low-B atoms.

In the use of Luzzati plots, the method of refinement, and its degree of convergence, is irrelevant. A Luzzati plot is a statement for the low-B atoms about the maximum errors associated with a given structure, whether converged or not.

References

18.1

Adams, P. D., Pannu, N. S., Read, R. J. & Brunger, A. T. (1999). *Extending the limits of molecular replacement through combined simulated annealing and maximum-likelihood refinement*. *Acta Cryst.* D**55**, 181–190.

Booth, A. D. (1946*a*). *A differential Fourier method for refining atomic parameters in crystal structure analysis*. *Trans. Faraday Soc.* **42**, 444–448.

Booth, A. D. (1946*b*). *The simultaneous differential refinement of co-ordinates and phase angles in X-ray Fourier synthesis*. *Trans. Faraday Soc.* **42**, 617–619.

Bricogne, G. (1997). *Bayesian statistical viewpoint on structure determination: basic concepts and examples*. *Methods Enzymol.* **276**, 361–423.

Brünger, A. T. (1992). *Free R-value – a novel statistical quantity for assessing the accuracy of crystal structures*. *Nature (London)*, **355**, 472–475.

Busing, W. R., Martin, K. O. & Levy, H. A. (1962). *ORFLS*. Report ORNL-TM-305. Oak Ridge National Laboratory, Oak Ridge, Tennessee, USA.

Diamond, R. (1971). *A real-space refinement procedure for proteins*. *Acta Cryst.* A**27**, 436–452.

Hendrickson, W. A. (1985). *Stereochemically restrained refinement of macromolecular structures*. *Methods Enzymol.* **115**, 252–270.

Hughes, E. W. (1941). *The crystal structure of melamine. J Am. Chem. Soc.* **63**, 1737–1752.

International Tables for Crystallography (1999). Vol. C. *Mathematical, physical and chemical tables*, edited by A. J. C. Wilson & E. Prince. Dordrecht: Kluwer Academic Publishers.

International Tables for Crystallography (2001). Vol. B. *Reciprocal space*, edited by U. Shmueli. Dordrecht: Kluwer Academic Publishers.

Kleywegt, G. J. (2000). *Validation of protein crystal structures*. *Acta Cryst.* D**56**, 249–265.

Kleywegt, G. J. & Jones, T. A. (1997). *Model building and refinement practice*. *Methods Enzymol.* **277**, 208–230.

Konnert, J. H. (1976). *A restrained-parameter structure-factor least-squares refinement procedure for large asymmetric units*. *Acta Cryst.* A**32**, 614–617.

Konnert, J. H. & Hendrickson, W. A. (1980). *A restrained-parameter thermal-factor refinement procedure*. *Acta Cryst.* A**36**, 344–350.

Kuriyan, J., Brünger, A. T., Karplus, M. & Hendrickson, W. A. (1989). *X-ray refinement of protein structures by simulated annealing: test of the method on myohemerythrin*. *Acta Cryst.* A**45**, 396–409.

Lamzin, V. S. & Wilson, K. S. (1997). *Automated refinement for protein crystallography*. In *Macromolecular crystallography*, Part B, edited by C. C. & R. Sweet, 269–305. San Diego: Academic Press.

Laskowski, R. A., MacArthur, M. W., Moss, D. S. & Thornton, J. M. (1993). *PROCHECK: a program to check the stereochemical quality of protein structures*. *J. Appl. Cryst.* **26**, 283–291.

McRee, D. E. (1993). *Practical protein crystallography*, p. 386. San Diego: Academic Press.

Murshudov, G. N., Vagin, A. A. & Dodson, E. J. (1997). *Refinement of macromolecular structures by the maximum-likelihood method*. *Acta Cryst.* D**53**, 240–253.

Murshudov, G. N., Vagin, A. A., Lebedev, A., Wilson, K. S. & Dodson, E. J. (1999). *Efficient anisotropic refinement of macromolecular structures using FFT*. *Acta Cryst.* D**55**, 247–255.

Pannu, N. S., Murshudov, G. N., Dodson, E. J. & Read, R. J. (1998). *Incorporation of prior phase information strengthens maximum-likelihood structure refinement*. *Acta Cryst.* D**54**, 1285–1294.

Pannu, N. S. & Read, R. J. (1996). *Improved structure refinement through maximum likelihood*. *Acta Cryst.* A**52**, 659–668.

Prince, E. (1994). *Mathematical techniques in crystallography and materials science*. 2nd ed. Berlin: Springer-Verlag.

Rice, L. M. & Brünger, A. T. (1994). *Torsion-angle dynamics – reduced variable conformational sampling enhances crystallographic structure refinement*. *Proteins Struct. Funct. Genet.* **19**, 277–290.

Sheldrick, G. M. (1993). *SHELXL93. Program for the refinement of crystal structures*. University of Göttingen, Germany.

Tronrud, D. E. (1992). *Conjugate-direction minimization: an improved method for the refinement of macromolecules*. *Acta Cryst.* **48**, 912–916.

Tronrud, D. E. (1996). *Knowledge-based B-factor restraints for the refinement of proteins*. *J. Appl. Cryst.* **29**, 100–104.

Tronrud, D. E. (1997). *The TNT refinement package*. *Methods Enzymol.* **277**, 306–318.

Tronrud, D. E., Ten Eyck, L. F. & Matthews, B. W. (1987). *An efficient general-purpose least-squares refinement program for macromolecular structures*. *Acta Cryst.* A**43**, 489–501.

Vriend, G. (1990). *WHAT IF: a molecular modeling and drug design program*. *J. Mol. Graphics*, **8**, 52–56.

Watenpaugh, K. D., Sieker, L. C., Herriott, J. R. & Jensen, L. H. (1972). *The structure of a non-heme iron protein: rubredoxin at 1.5 Å resolution*. *Cold Spring Harbor Symp. Quant. Biol.* **36**, 359–367.

Watenpaugh, K. D., Sieker, L. C., Herriott, J. R. & Jensen, L. H. (1973). *Refinement of the model of a protein: rubredoxin at 1.5 Å resolution*. *Acta Cryst.* B**29**, 943–956.

18.2

Abramowitz, M. & Stegun, I. (1968). *Handbook of mathematical functions. Applied mathematics series*, Vol. 55, p. 896. New York: Dover Publications.