Zusammenfassung der Valenzen 5–5' und 6–6':

$$x_{\mu} = +0,4654 \quad y_{\mu} = 0 \quad z_{\mu} = -1,6684.$$

c) Die Valenzen 7_s' und 7_a' (s. Abb. 3)

$$\operatorname{tg} \beta = \frac{z_7 - z_1}{x_7 - x_1}, \quad \beta = 56,21^\circ.$$

$$\alpha_{7_s'} = 110,95^\circ,$$

$$\alpha_{7_a'} = 1,47^\circ,$$

Die Einheitsvektoren:

$$7_s' \quad x_{\mu} = -0,3574 \quad y_{\mu} = 0 \quad z_{\mu} = +0,9339$$

$$7_a' \quad x_{\mu} = +0,9997 \quad y_{\mu} = 0 \quad z_{\mu} = +0,0257$$

$$x_{\mu} = +0,6423 \quad y_{\mu} = 0 \quad z_{\mu} = +0,9596$$

Zusammengefaßt ergeben sich für die 6 C–Cl-Bindungen des Hexachlorbicycloheptens folgende Vektor-

komponenten:

$$1' + 4' \quad x_{\mu} = +0,4892 \quad z_{\mu} = +0,5148$$

$$5' + 6' \quad x_{\mu} = +0,4654 \quad z_{\mu} = -1,6684$$

$$7_s' + 7_a' \quad x_{\mu} = +0,6423 \quad z_{\mu} = +0,9596$$

$$x_{\mu} = +1,5969 \quad z_{\mu} = -0,1940$$

Mit Benutzung der Bindungsmomente:

$\mu_B = 2,15$ D für die C–Cl-Valenzen 1' + 4' (aus den Momenten von tert.-Butyl- und tert.-Amylchlorid),

$\mu_B = 1,40$ D für die C–Cl-Valenzen 5' + 6' (aus den Momenten von *cis*-2,3-Dichlor-buten-(2), o-Dichlorbenzol und 2,3-Dichlor-naphthalin) und

$\mu_B = 1,95$ D für die C–Cl-Valenzen 7_s' + 7_a' (aus den Momenten von 2,2-Dichlorpropan und 1,1-Dichlorcyclopentan)

ergibt sich für das 1.4.5.6.7.7-Hexachlor-bicyclo-[2.2.1]-hepten-(5) (I) ein Gesamtmoment von 3,02 D, das in guter Übereinstimmung mit gemessenen Werten^{5, 6} steht.

Considerations of image contrast in Electron Microscopy of objects composed of low atomic number

By M. L. DE

Biophysics Division, Saha Institute of Nuclear Physics, Calcutta-9, India

(Z. Naturforschg. 17 b, 728—731 [1962]; eingegangen am 17. März 1962)

Defects of the electron scattering formula based on Thomas-Fermi atom model and of Leisegang's formula, are first briefly discussed in relation to image contrast problem in Electron Microscopy. A few aspects of the electron scattering formula of Lenz are then considered particularly for dealing with elements of low atomic number. From a consideration of this formula it is shown that the image contrast of a given mass thickness of object is liable to be dependent on the atomic number of the object.

It is well known that contrast in the electron microscope image of an object, arises due to differential scattering of the beam electrons in the different parts of the object. The question of electron scattering and image contrast resulting therefrom, have been studied both theoretically as well as experimentally by several authors. The earlier work of SCHIFF¹, BOERSCH² and BORRIES³ were based on Thomas-Fermi atom model and BORRIES particularly, by using the elastic and inelastic scattering formula of MOLIERE⁴ and KOPPE⁵ respectively, had come to a number of interesting conclusions. But in the electron microscopy of biological samples, one naturally deals with matter composed of low atomic

weight elements for which the TF statistical model does not provide a good representation particularly in the small angle region ($\sim 10^{-2}$ radian) of electron microscopic work. Further, the experimental results of LEONHARD⁶ and COUPLAND⁷ also showed the invalidity of the Molieré-Koppe formulae for low as well as for high atomic weight samples. LEISEGANG⁸, after first Born approximation, had derived electron scattering formula for multiple elastic collisions, and showed good agreement of the formula with his experimental results at fairly large scattering angles for comparatively thick samples of Au (150 Å), Al (270 Å) and Ag (>120 Å). This elastic scattering equation has

¹ L. I. SCHIFF, *Physic. Rev.* **61**, 72 [1942].

² H. BOERSCH, *Z. Naturforschg.* **2a**, 615 [1947].

³ B. V. BORRIES, *Z. Naturforschg.* **4a**, 51 [1949].

⁴ G. MOLIERE, *Z. Naturforschg.* **2a**, 133 [1947].

⁵ H. KOPPE, *Z. Physik* **124**, 658 [1947].

⁶ F. LEONHARD, *Z. Naturforschg.* **9a**, 1019 [1954].

⁷ J. H. COUPLAND, *Proc. physic. Soc.* **69 B**, 46 [1956].

⁸ S. LEISEGANG, *Z. Physik* **132**, 183 [1952].

been used by several authors⁹⁻¹³ for estimation of mass thickness of biological substances by measurement of contrast of electron microscope image. In these cases, however, the inelastic scattering cross-section has been totally disregarded although in the small angle region of electron microscope work, the inelastic scattering is more predominant than the elastic scattering process.

LENZ¹⁴, by making use of the elastic and inelastic scattering equations of WENTZEL¹⁵ and MORSE¹⁶, developed scattering formula based on Hartree atom model; and they were therefore very suitable for low atomic weight elements and for small scattering angle. Lenz's equations made satisfactory agreement with the earlier experimental results of BIBERMAN et al.¹⁷ and with the subsequent works of other authors¹⁸⁻²⁰; further, SILVESTER and BURGE²¹ on the basis of these equations, computed for biological matter, the 'aufhellungsdicke', which denotes the upper limit of mass thickness which can be usefully examined with the electron microscope.

SADHUKHAN²² made certain modifications in the Lenz's equations by considering the atomic radius on Wentzel model ($R = a_H \cdot Z^{-1/2}$); and these modified equations were used by COSSLETT²³ in his paper dealing with quantitative electron staining.

In the present paper some aspects of Lenz's equations have been considered particularly for elements of low atomic weight, showing the nature of dependence of the scattering cross-sections of the various atoms on their atomic numbers. Considerations are then given to the resulting image contrast expected on the basis of these scattering cross-sections.

Consideration of Lenz's Scattering Formula

According to the equation of Lenz, the expressions for the differential cross-section of an atom (at. No. Z), for elastic and inelastic scattering, can be written as,

$$\frac{d\sigma_e}{d\Omega} = \frac{4Z^2}{a_H^2} \frac{1}{\left(q^2 + \frac{1}{R^2}\right)^2} \quad (1)$$

$$\frac{d\sigma_{in}}{d\Omega} = \frac{4Z}{a_H^2 q^4} \left[1 - \frac{1}{(q^2 R^2 + 1)^2} \right] \quad (2)$$

with

$$q = \frac{4\pi}{\lambda} \cdot \sin \frac{\alpha}{2} \quad (3)$$

where a_H is the Bohr radius of the Hydrogen atom, λ is the de-Broglie electron wavelength, α is the scattering angle, and R is the screening radius of the atom. R has been represented by LENZ as

$$R = (\Theta/6Z)^{1/2} \quad (4)$$

where the parameter Θ is related to the electron density distribution in the atomic shell. Θ has been expressed as $\Theta = k \cdot a_H^2$ where k is a numerical factor, values of which for several atoms on Hartree model have been tabulated by WYRWICH and LENZ²⁴. With these tabulated values of Θ combined with equation (4) one gets values of the screening radius of several atoms as represented in Fig. 1, here for comparison, the corresponding Wentzel radii have also been shown. It is seen that while the Wentzel radius decreases smoothly with increase of Z , the Hartree radius is a discontinuous function of Z .

Fig. 1. Variation of the screening radius of atoms with Atomic Number Z , calculated on Hartree and Wentzel atom models.

Fig. 2 shows the angular variation of differential cross-section of carbon for elastic scattering calculated

- ⁹ E. ZEITLER and G. F. BAHR, Exp. Cell Res. **12**, 44 [1957].
- ¹⁰ M. L. DE and P. SADHUKHAN, Nature [London] **182**, 1008 [1958].
- ¹¹ E. ZEITLER and G. F. BAHR, J. appl. Physics **30**, 940 [1959].
- ¹² G. BLOOM and E. ZEITLER, Exp. Cell Res. **17**, 13 [1959].
- ¹³ L. H. SOBIN, G. BLOOM and E. ZEITLER, Exp. Cell Res. **17**, 472 [1959].
- ¹⁴ F. LENZ, Z. Naturforsch. **9a**, 185 [1954].
- ¹⁵ G. WENTZEL, Z. Physik. **40**, 590 [1927].
- ¹⁶ P. M. MORSE, Physik. Z. **33**, 443 [1932].
- ¹⁷ L. M. BIBERMAN, E. N. WTOROW, I. A. KOWNER, N. G.

- SRUSCHKIN and B. M. JAWORSKIT, C. R. [Doklady] Acad. Sci., U.R.S.S. **69**, 519 [1949].
- ¹⁸ G. KEMPF and F. LENZ, Proc. Stockholm Conf. Electron Microscopy, p. 67, Almqvist and Wiksell, Stockholm 1957.
- ¹⁹ C. E. HALL and T. INOUE, J. appl. Physics **28**, 1348 [1957].
- ²⁰ M. E. HAINE and A. W. AGAR, Brit. J. appl. Physics **10**, 341 [1959].
- ²¹ N. R. SILVESTER and R. E. BURGE, Nature [London] **188**, 641 [1960].
- ²² P. SADHUKHAN, J. appl. Physics **29**, 1235 [1958].
- ²³ V. E. COSSLETT, J. Roy. microscop. Soc. **78**, 18 [1958].
- ²⁴ H. WYRWICH and F. LENZ, Z. Naturforsch. **13a**, 515 [1958].

on Hartree and Wentzel models with Lenz's equations and on TF model with Moliere's equation. It is seen that in the angular range of 10^{-4} to 10^{-2} radian and for 50 kV electrons, the Hartree cross-section is about 2.5 times greater than the Wentzel value and about 4 times smaller than TF cross-section.

Fig. 2. Variation of the differential elastic scattering cross-section of carbon atom with angle, for 50 kV electrons, calculated on the basis of TF, Hartree and Wentzel atom models.

The expressions for the total elastic and inelastic cross-section can be written after suitable transformation as in the Table below.

Total scattering cross-section	HARTREE	WENTZEL
Elastic: $(\sigma_e)_0^\pi = \int_0^\pi d\sigma_e$	$0.053 k Z \lambda^2$	$0.316 Z^{4/3} \lambda^2$
Inelastic: $(\sigma_{in})_q^\pi = \int_q^\pi d\sigma_{in}$	$0.106 k \lambda^2 \ln \frac{6Z}{q^2 k a_H^2}$	$0.632 \lambda^2 Z^{1/3} \ln \frac{Z^{2/3}}{q^2 a_H^2}$
$\frac{\sigma_{in}}{\sigma_e}$	$\frac{2}{Z} \ln \frac{6Z}{q^2 k a_H^2}$	$\frac{2}{Z} \ln \frac{Z^{2/3}}{q^2 a_H^2}$

Table I. Total scattering cross-sections on Hartree and Wentzel Models.

In the above expressions, the parameter q denotes the minimum inelastic scattering angle and is given by

$$q = \frac{2\pi}{\lambda} \frac{\Delta E}{U} \quad (5)$$

where ΔE is the ionisation potential of the atom and U is the electron energy, both in volts. From the tabulated values of ΔE for the various atoms as is given by LANDOLT and BORNSTEIN²⁵ one finds that, for U in the

²⁵ LANDOLT and BORNSTEIN, Zahlenwerte und Funktionen, 1, 1, 211, 6. Auflage, 1950.

range of 50–100 kV, $\ln q$ is practically constant, so that both σ_e and σ_{in} can be plotted respectively as σ_e/λ^2 and σ_{in}/λ^2 against Z , as shown in Fig. 3. Here curves (1) and (2) are drawn on Hartree model and curves 1(A) and 2(A) on Wentzel model. The ratio (σ_{in}/σ_e) for the various atoms has been found to be practically the same on the two models, and curve 3 in Fig. 3 represents the said ratio, while curve 3(A) has been taken from Fig. 7 of LENZ's paper¹⁴.

Fig. 3. Dependence of total elastic and inelastic scattering cross-sections of atoms on Z ; curves 1 and 2 with Hartree radius, curves 1A and 2A with Wentzel radius. Curve 3 denotes the ratio of $\frac{\text{Total inelastic cross-section}}{\text{Total elastic cross-section}}$ on both atom models; curve 3A taken from Fig. 7 of LENZ's paper¹⁴.

Since for the evaluation of image contrast, the parameter of importance is $\int_\alpha^\pi d\sigma = (\sigma)_\alpha^\pi$ i. e. the value of the cross-section for scattering beyond the effective objective angle α , $(\sigma)_\alpha^\pi$ for carbon has been plotted in Fig. 4 for 50 kV electrons against various values of aperture angles normally used in electron microscopy (10^{-3} – $3 \cdot 10^{-2}$ radian). Both the elastic (σ_e) as well as the inelastic (σ_{in}) cross-sections, as well as the total cross-section i. e. $\sigma_e + \sigma_{in}$ are shown in Fig. 4, calculated on both Hartree and Wentzel radius basis. In Fig. 5 is represented the scattering cross-section per gm., S , of various substances for two aperture angles viz. 3.5×10^{-3} , and 10^{-2} radians, for 50 kV electrons, S being given by

$$S = \frac{N_A}{A} \cdot (\sigma_e + \sigma_{in}) \quad (6)$$

where $A = \text{At. wt.}$, $N_A = \text{Avogadro's No.}$

Fig. 4. Angular variation of elastic and inelastic scattering cross-section of carbon atom for 50 kV electrons.

Fig. 5. Variation of scattering cross-section per gm. of various elements with Atomic Number Z .

Conclusion

From the figures reproduced above one finds that, owing to the regular nature of variation of the Wentzel radius with Z (Fig. 1, curve a), the corresponding scattering cross-section also show a continuous variation with atomic number [Fig. 3, lines 1(A) and 2(A)], while the total scattering cross-section per gm. S is remarkably constant that is independent of variation in Z (Fig. 5, curves w). But it has been shown²⁶ already that if one uses the Lenz's equation with Wentzel radius, the calculated values of scattering cross-section do not agree satisfactorily with values obtained from experimental results.

On the other hand, due to the irregular variation of the Hartree radius with Z (Fig. 1, curve b) there is discontinuous variation with Z of the scattering cross-section per atom (Fig. 3, curves 1, 2); also the total cross-section per gm. S , is seen to vary with Z rather irregularly (Fig. 5, curve H). This means that the image contrast for the same amount of object matter is liable to vary with atomic number.

The same conclusion is also reached from the experimental standpoint as has been shown by the author in a recent communication²⁷.

The author thanks Prof. N. N. DAS GUPTA of Calcutta University for his kind interest and also the Ministry of Scientific Research and Cultural Affairs for financial support.

²⁶ M. L. DE, Nature [London] **192**, 547 [1961].

²⁷ M. L. DE, Naturwissenschaften **49**, 296 [1962].