



**International
Standard**

ISO 25387

**Microbeam analysis — Analytical
electron microscopy — Procedures
for determining the point resolution
of high-resolution transmission
electron microscope**

Analyse par microfaisceaux — Microscopie électronique analytique — Modes opératoires de détermination de la résolution ponctuelle des microscopes électroniques à transmission à haute résolution

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Foreword

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The procedures used to develop this document and those intended for its further maintenance are described in the ISO/IEC Directives, Part 1. In particular, the different approval criteria needed for the different types of ISO document should be noted. This document was drafted in accordance with the editorial rules of the ISO/IEC Directives, Part 2 (see www.iso.org/directives).

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This document was prepared by Technical Committee ISO/TC 202, *Microbeam analysis*, Subcommittee SC 3, *Analytical electron microscopy*.

Any feedback or questions on this document should be directed to the user's national standards body. A complete listing of these bodies can be found at www.iso.org/members.html.

Introduction

High-resolution transmission electron microscopes (HREM) that can observe ultra-fine structures with sub-nanometre resolution have been applied in various fields such as medical, biological, materials science, classical and innovative materials, and others.

One of the key factors in knowing the limitations of the microscopic performance of HREM is the point resolution. Although there are various categories of TEM resolution definitions, point resolution for HREM is generally defined by the Scherzer resolution.

Scherzer resolution is measured from the high-resolution image of a weak phase object observed under a specific defocusing condition (called the Scherzer focus or the Scherzer condition) derived from the spherical aberration coefficient of the objective lens and the electron wavelength. However, different definitions have been proposed for the Scherzer condition, which is an important factor in the resolution measurement of TEM, depending on the purpose of the observation and the targeting materials. To determine the TEM resolution, this document adopts the Scherzer condition, which is derived from information theory.

Furthermore, although the spherical aberration coefficient is usually provided by the TEM manufacturer, it is important to know the real spherical aberration coefficient to properly evaluate the instrumental performance of the TEM. This document specifies the procedure for measuring real spherical aberration coefficient of objective lens used and the procedure for determining the point resolution (Scherzer resolution) of HREM using the measured real spherical aberration coefficient.

Recently, ultra-high-resolution electron microscopes equipped with a spherical aberration corrector, called “ C_s -corrected TEMs”, have been developed and widely used. These state-of-the-art TEMs can reduce spherical aberration to as close to zero as possible, contributing to a dramatic improvement in TEM resolution beyond the Scherzer resolution. In general, the resolution of these state-of-the-art TEMs is defined by the critical structure size (called the information limit) at which the phase contrast carried by the phase contrast transfer function vanishes by the envelope function. For this reason, the resolution for C_s -corrected TEMs should be treated as a separate category from the Scherzer resolution for HREMs.

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Microbeam analysis — Analytical electron microscopy — Procedures for determining the point resolution of high-resolution transmission electron microscope

1 Scope

This document specifies a procedure for determining the point resolution, called Scherzer resolution, of high-resolution transmission electron microscopes (HREM), which can visualize sample structure with sub-nanometre fineness. This document also specifies the measurement procedure of the real spherical aberration coefficient of the objective lens used.

The procedure specified in this document for measuring the spherical aberration coefficient uses the dark rings that appear in the fast Fourier transform (FFT) pattern of HREM images of amorphous thin films, in which, at least three dark rings need to be observable near the Scherzer focus. Therefore, this document is applicable to HRTEMs equipped with a cold field emission gun (CFEG), Schottky emission gun (SEG) or thermal field emission gun (TFEG), or HREM equipped with a thermionic emission gun (TEG) in which three or more dark rings can be clearly observed in the FFT pattern.

This document does not treat the information limits, lattice resolution and STEM resolution. In addition, this document is not applicable to C_s -corrected TEM.

2 Normative references

There are no normative references in this document.

3 Terms and definitions

For the purposes of this document, the following terms and definitions apply.

ISO and IEC maintain terminology databases for use in standardization at the following addresses:

- ISO Online browsing platform: available at <https://www.iso.org/obp>
- IEC Electropedia: available at <https://www.electropedia.org/>

3.1

chromatic aberration

lens defect which arises because electrons from the same point in the specimen, but of slightly different energies, will be focused at different positions in the image plane

[SOURCE: ISO15932:2013, 3.2.2.1]

3.2

cold field emission gun

CFEG

electron gun (3.5) employing cold field emission

[SOURCE: ISO15932:2013, 3.1.2.1]

3.3

defocus spread

fluctuation of the defocus caused by the *energy spread* (3.6) of the incident electron beam, and fluctuation in the power supply for accelerating voltage and the objective lens exciting current

**3.4
diffractogram**

image showing how the intensity of Fourier transform pattern of the high-resolution TEM image distributes with the structure size

**3.5
electron gun**

component that produces an electron beam with a well-defined kinetic energy

[SOURCE: ISO15932:2013, 3.1]

**3.6
energy spread**

diversity of energy of electrons in the incident beam

[SOURCE: ISO15932:2013, 2.1.1.1]

**3.7
envelope function**

function that expresses the damping of the *phase contrast transfer function* (3.17), caused by *energy spread* (3.6) of the incident electron beam, fluctuation in the power supply for accelerating voltage and the objective lens exciting current, angular aperture of an incident electron beam, and environmental instability

**3.8
fast Fourier transform
FFT**

efficient algorithm to compute the discrete Fourier transform

[SOURCE: ISO15932:2013, 5.4.1.1]

**3.9
first zero**

position where the *phase contrast transfer function* (3.17) first intersects the spatial frequency axis, and the reciprocal of the wave number indicated by this position corresponds to *point resolution* (3.19) of HREM

**3.10
illumination semi-angle**

divergence semi-angle of the incident electron beam to a specimen

**3.11
information limit**

theoretical limit of the resolution of HREM, established by the observation of an amorphous specimen and used as a performance index of a TEM

[SOURCE: ISO15932:2013, 7.3]

**3.12
lattice spacing**

line spacing of periodic structure formed by interference between a scattered wave from a set of lattice planes of the crystal and transmitted wave or between two scattered waves.

**3.13
lattice resolution**

resolution under imaging, which corresponds to the periodic lattice structure of the specimen

[SOURCE: ISO15932: 2013, 7.6]

**3.14
live fast Fourier transform
live FFT**

fast Fourier transform (3.8) processing technology which follows changes in processing target images in real time

3.15

Miller index

notation system in crystallography for planes and directions in crystal lattices, in which a family of lattice planes or directions is determined by three integers h, k, l

[SOURCE: ISO15932: 2013, 8.4]

3.16

phase contrast

image contrast due to the interference of transmitted and phase-shifted diffracted waves, which provides high-resolution TEM image

[SOURCE: ISO15932: 2013, 6.2, modified — “interference of transmitted and” has been added]

3.17

phase contrast transfer function

PCTF

function which provides image contrast modulation caused by the phase shift produced by combination of given spherical aberration and defocus

3.18

phase-distortion function

function representing the equiphase surface distribution of the electron wave passing through the lens, which depends on the defocus amount, spherical aberration coefficient, spatial frequency, and wavelength of electrons

3.19

point resolution

resolution under *Scherzer focus* (3.24), which is defined as the reciprocal of the spatial frequency where the *phase contrast transfer function* (3.17) crosses the abscissa for the first time

[SOURCE: ISO15932: 2013, 7.6]

3.20

projected potential

electrostatic potential of a crystal projected along a low-index zone axis

3.21

region of interest

ROI

sub-dataset picked out from the entire dataset for a specific purpose

[SOURCE: ISO20263: 2017, 3.1.24]

3.22

Scherzer condition

state of observation set to Scherzer focus

3.23

Scherzer defocus

state in which the focus is shifted to *Scherzer focus* (3.24)

3.24

Scherzer focus

$\Delta f_{\text{Scherzer}}$

defocusing condition in which high-resolution images of the *weak phase object* (3.35) are formed by shifting the phase of the scattered beam by $-0,63 \pi$ (phase delay) which maximizes the spatial frequency of the first zero of the contrast transfer function

Note 1 to entry: $\Delta f_{\text{Scherzer}}$ is given by the Formula:

$$\Delta f_{\text{Scherzer}} = 1,12\sqrt{C_s\lambda}$$

where C_s is the spherical aberration coefficient and λ is the electron wavelength.

Note 2 to entry: $\Delta f_{\text{Scherzer}} > 0$ indicates the condition of under focus.

[SOURCE: ISO15932: 2013, 5.9.3, modified — “the diffracted beam by $0,5 \pi$ ” has been replaced by “the scattered beam by $-0,63 \pi$ (phase delay)”, “ $\Delta f_{\text{Scherzer}} = -1,12\sqrt{C_s\lambda}$ ” has been replaced by “ $\Delta f_{\text{Scherzer}} = 1,12\sqrt{C_s\lambda}$ ”, and “Note 2 to entry” has been added.]

3.25

Scherzer resolution

d_{Scherzer}

point resolution (3.19) of HREM defined by the structural size corresponding to the first zero position of the phase contrast transfer function (3.17) at Scherzer focus (3.24)

Note 1 to entry: d_{Scherzer} is given by the Formula:

$$d_{\text{Scherzer}} = 0,668C_s^{\frac{1}{4}}\lambda^{\frac{3}{4}}$$

where C_s is the spherical aberration coefficient and λ is the electron wavelength.

3.26

Schottky emission

thermionic electron emission that takes place under an electric field that enhances emission by lowering the surface barrier

[SOURCE: ISO 15932: 2013, 3.1.1]

3.27

Schottky emission gun

SEG

electron gun (3.5) employing Schottky emission (3.26)

3.28

spatial frequency

reciprocal of structure size

3.29

spherical aberration

lens defect arising from the varying strength of an electromagnetic lens with distance from the optic axis, which causes rays further from the optic axis to be focused more strongly than those nearer the optic axis

[SOURCE: ISO 15932: 2013, 3.2.2.2]

3.30

temporal coherency

correlations between waves observed at different points in time

3.31

thermal field emission gun

TFEG

electron gun (3.5) employing thermal field emission

[SOURCE: ISO 15932: 2013, 3.1.2.3]

3.32

thermionic emission gun

TEG

electron gun (3.5) employing thermionic emission

[SOURCE: ISO 15932: 2013, 3.1.3]

3.33

through-focus technique

image recording method that records images sequentially while varying the focal length at defined intervals

3.34

under focus

focusing condition of the objective lens in which its excitation is adjusted to slightly decreased rather than focusing on a specimen

3.35

weak phase object

WPO

very thin TEM sample to which *weak phase object approximation* (3.36) can be applied

3.36

weak phase object approximation

WPOA

approximation assuming that the *weak phase object* (3.35) does not change the amplitude of the incident wave but shifts the phase slightly in proportion to the *projected potential* (3.20)-

[SOURCE: ISO15932: 2013, 6.2.1, modified — “in which the specimen” has been replaced by “assuming that the *weak phase object* (3.35)” and “slightly shifts the phase” has been replaced by “shifts the phase slightly in proportion to the *projected potential* (3.20)”.]

4 Symbols and abbreviated terms

C_c	chromatic aberration coefficient
CFEG	cold field emission gun
C_s	spherical aberration coefficient
$C_{s(\text{prvd})}$	the spherical aberration coefficient provided by TEM manufacturer
$C_{s(\text{r})}$	the real spherical aberration coefficient of the objective lens used
$d_{hkl(\text{Au})}$	lattice spacing indicated by the (hkl) plane of the colloidal gold nano crystal
d_{Scherzer}	theoretical point resolution of HREM
$d_{\text{Scherzer}(\text{prvs})}$	provisional Scherzer resolution
$d_{\text{Scherzer}(\text{r})}$	reciprocal of the $u_{\text{Scherzer}(\text{r})}$ value, representing measured point resolution of the TEM used
$D_{n(j)}$	diameter (in pixels) of the n -th dark ring of the diffractogram for j -th image
$D_{\text{frg}}(j)$	diffractogram obtained by FFT processing to the ROI set in the $\text{Img}(j)$
$E_c(u)$	envelope function for temporal coherency based on the chromatic aberration
$E_s(u)$	envelope function for spatial coherency based on the effective electron source size

F	objective lens focal length at in-focus position
FFT	fast Fourier transform/transformation
$F_{\text{PCTF}(r)}$	realistic phase contrast transfer function
HREM	high resolution transmission electron microscope
$\text{Img}(j)$	j -th image in the through-focus image series
I_{obj}	objective lens exciting current
$L1(j)$	line connecting the spots reflected Au lattice spacing appeared in the $\text{Dfrg}(j)$
$L1(j)$	line-profile along $L1(j)$
$L2(j)$	line passing through the centre of $\text{Dfrg}(j)$
$L2(j)$	line-profile along $L2(j)$
$N_{hkl}(j)$	in the diffractogram for j -th image, distance (in pixels) between two peaks corresponding to bright spots reflecting the lattice spacing of the (hkl) plane of the colloidal gold nano crystal
PCTF	phase contrast transfer function
ROI	region of interest
$\sin \chi(u)$	phase contrast transfer function (PCTF)
SEG	Schottky emission gun
STEM	scanning transmission electron microscope/microscopy
TEG	thermionic emission gun
TEM	transmission electron microscope/microscopy
TFEG	thermal field emission gun
u	spatial frequency
$u_{n(j)}$	spatial frequency corresponding to the n -th dark ring of the diffractogram for j -th image
$u_{\text{unit}(j)}$	in the diffractogram for j -th image, spatial frequency (in nm^{-1}) corresponding to one pixel
u_{Scherzer}	spatial frequency for the 1st zero of PCTF under Scherzer condition
$u_{\text{Scherzer}(r)}$	spatial frequency corresponding to the 1 st dark ring of $\text{Dfrg}(j)$ obtained by applying the $\Delta f_{\text{Scherzer}(r)}$ to the approximate quadratic equation
V_{acc}	accelerating voltage
WPO	weak phase object
WPOA	weak phase object approximation
α	illumination semi-angle of incident electron beam
ΔE	energy spread of electrons
Δf	amount of defocus

$\Delta f_{\text{ave}(j)}$	average of three defocus values ($\Delta f_{n(j)}$ ($n=1$ to 3)) for the j -th image
$\Delta f_{\text{Scherzer}}$	amount of Scherzer focus
$\Delta f_{\text{Scherzer}(\text{prvs})}$	provisional Scherzer focus
$\Delta f_{\text{Scherzer}(\text{r})}$	real Scherzer focus
$\Delta f_{n(j)}$	defocus value calculated from diameter of n -th dark ring of the diffractogram for j -th image
λ	wavelength of electron
$\chi(u)$	phase-distortion function

5 Definition of point resolution based on information theory

5.1 General

It is widely recognised that the point resolution of a TEM is determined by the Scherzer resolution. However, the coefficients included in the formula defining the Scherzer condition and the Scherzer resolution have slightly different values depending on the target and purpose of observation.^[2] Therefore, there is a possibility that different measurers can have different point resolution values.

In order to eliminate this issue, in this document, the point resolution is defined by applying a value determined on the basis of information theory ^[3] to the coefficients.

5.2 Definition of Scherzer condition

The observation condition (or defocus condition) that produces a phase contrast image reflecting the projection potential of a weak phase object is called the Scherzer condition (or Scherzer focus; represented by $\Delta f_{\text{Scherzer}}$), and the Scherzer resolution is obtained under this condition^[4].

The Scherzer condition is derived from the phase-distortion function ($\chi(u)$),^[5] which describes the phase difference between the electrons scattered by a weak phased object and the unscattered electrons, when they pass through the objective lens and combine at the image plane. The phase-distortion function, expressed as a composite of two phase-shifts induced by spherical aberration and defocusing of the objective lens, is described by [Formula \(1\)](#)^[6].

$$\chi(u) = \delta_{C_s} + \delta_{\Delta f} = 0,5\pi C_s \lambda^3 u^4 - \pi \Delta f \lambda u^2 \quad (1)$$

where

- $\chi(u)$ is the phase-distortion function;
- δ_{C_s} is the phase shift caused by spherical aberration of objective lens;
- $\delta_{\Delta f}$ is the phase shift caused by defocus of objective lens;
- C_s is the spherical aberration coefficient of objective lens;
- λ is the wavelength of the electron;
- u is the spatial frequency;
NOTE u is the reciprocal of the structure size ($1/d$).
- Δf is defocus of objective lens.
NOTE $\Delta f > 0$ is under focus.

A graph of the phase-distortion function for $\lambda = 0,002\,51$ nm and $C_s = 1$ mm is shown in [Figure 1](#). The parameter is defocus values ($\Delta f = 40$ nm, 60 nm, and 80 nm). The variation of the phase contrast with

spatial frequency can be derived from the phase-distortion function. Namely, the phase contrast is zero when the phase-distortion function is an even multiple of $\pi/2$, and the contrast shows maximum when the phase-distortion function is equal to an odd multiple^[7]. The Scherzer condition is a condition for obtaining high phase contrast over a wide spatial frequency range, which is achieved by a defocus condition such that the minimum value (Figure 1, Key 5) of the phase-distortion function is approximately equal to $-\left(\frac{\pi}{2}\right)$ (Figure 1, Key 4). For example, a defocus value of 60 nm (Key 2) in Figure 1 is close to the Scherzer condition.

The relationship between the minimum value ($\chi(u)_{\min}$) of phase-distortion function and defocus (Δf) is expressed by Formula (2).

$$\chi(u)_{\min} = -\frac{\pi}{2} \left(\frac{\Delta f^2}{C_s \lambda} \right) \quad (2)$$

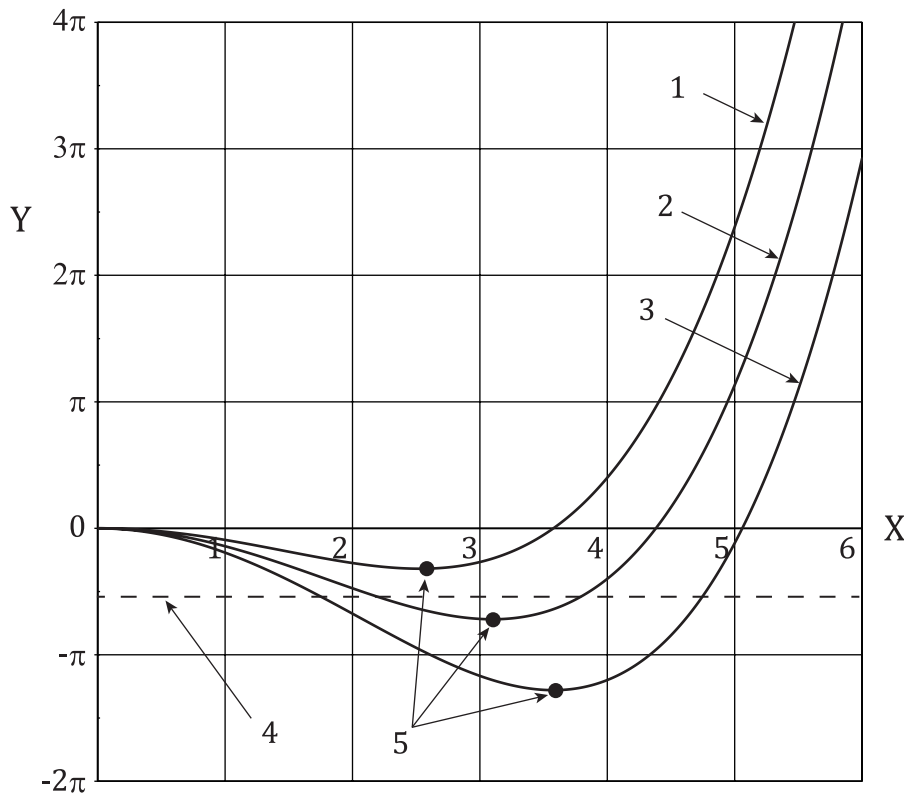
According to information theory,^[3] the maximum information of the sample is transferred to the image when the value of $\chi(u)_{\min}$ is equal to $0,63 \pi$.^[8] In this document, the defocus value expressed in Formula (3) derived from this condition is defined as the Scherzer condition ($\Delta f_{\text{Scherzer}}$).

$$\Delta f_{\text{Scherzer}} = \sqrt{1,26 \times C_s \lambda} = 1,12 (C_s \lambda)^{\frac{1}{2}} \quad (3)$$

For example, under the TEM conditions used to draw Figure 1, $\Delta f_{\text{Scherzer}}$ is calculated as +56 nm (under focus).

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Key

- X spatial frequency ($u \text{ nm}^{-1}$)
- Y phase-distortion ($\chi(u)$) of scattered wave against transmitted wave
- 1 graph for defocus at 40 nm.
- 2 graph for defocus at 60 nm.
- 3 graph for defocus at 80 nm.
- 4 dashed-line indicating $\chi(u) = -\frac{\pi}{2}$.
- 5 black dots indicating the minimum position of each $\chi(u)$ curve.

Figure 1 — Phase-distortion function ($\chi(u)$).
TEM conditions; Accelerating voltage is 200 kV ($\lambda = 0,002 51 \text{ nm}$), and C_s is 1,0 mm.

5.3 Definition of the theoretical point resolution

The change in phase contrast as a function of spatial frequency (u) can be expressed as a sine function of $\chi(u)$, represented by [Formula \(4\)](#).

$$\sin \chi(u) = \sin(0,5\pi C_s \lambda^3 u^4 - \pi \Delta f \lambda u^2) \tag{4}$$

The $\sin \chi(u)$ is known as the phase contrast transfer function (PCTF). Examples of PCTF series due to differences in defocus are shown in [Figure 2](#). The TEM conditions for obtaining these graphs are the same as for [Figure 1](#). The appearance of the graph depends on the defocus value, but their common rule is to oscillate as u increases after a graph crosses the u -axis first (so called 1st zero).

[Figure 2b](#) shows the PCTF under the defocus condition of $\Delta f_{\text{Scherzer}}$. In this graph, it is clear that the $\sin \chi(u)$ is close to -1, i.e., high contrast, over a wide range of u . In contrast, [Figures 2a](#) and [2c](#) show the PCTFs when both defocus conditions are away from the $\Delta f_{\text{Scherzer}}$, indicating a narrower range of high contrast.