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Microbeam analysis — Analytical electron microscopy — Methods for calibrating image magnification by using reference materials with periodic structures

Analyse par microfaisceaux — Microscopie électronique analytique — Méthodes d'étalonnage du grandissement d'image au moyen de matériaux de référence de structures périodiques

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Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

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 ~~documents~~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).~~www.iso.org/directives).

~~Attention is drawn~~ISO draws attention to the possibility that ~~some of~~ the ~~elements~~implementation of this document may ~~be~~involve the ~~subject~~use of (a) patent(s). ISO takes no position concerning the evidence, validity or applicability of any claimed patent rights~~.~~ in respect thereof. As of the date of publication of this document, ISO had not received notice of (a) patent(s) which may be required to implement this document. However, implementers are cautioned that this may not represent the latest information, which may be obtained from the patent database available at www.iso.org/patents. ISO shall not be held responsible for identifying any or all such patent rights.~~ Details of any patent rights identified during the development of the document will be in the Introduction and/or on the ISO list of patent declarations received (see www.iso.org/patents).~~

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For an explanation ~~on~~of the voluntary nature of standards, the meaning of ISO specific terms and expressions related to conformity assessment, as well as information about ISO's adherence to the World Trade Organization (WTO) principles in the Technical Barriers to Trade (TBT~~) see the following URL: www.iso.org/iso/foreword.html.~~), see www.iso.org/iso/foreword.html.

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

This ~~second~~third edition cancels and replaces the ~~first edition (ISO 29301:2010), which has been technically revised.~~

~~The main changes compared to the previous edition are as follows:~~

~~— Annexes B and C have been changed to informative;~~

~~— the Foreword has been revised;~~

~~— the Introduction has been revised;~~

~~— Clause 1 has been revised;~~

~~— Clause 2 has been updated;~~

~~— ISO 5725-1 and ISO/IEC Guide 98-3 have been added to the Bibliography;~~

— the heading and first paragraph of Clause 3 have been updated;

— the terms "accuracy" (3.1) and "under focus" (3.35) have been added to Clause 3;

— the terms "beam damage", "goniometer" and "lattice spacing" have been deleted from Clause 3;

— the term "just focus" has been replaced by the term "focus" (3.10) in Clause 3;

— the term "image file format" has been replaced by the term "image file" (3.14) in Clause 3;

— the terms "image scanner" (3.17) and "ROI: region of interest" (3.26) have been revised;

— the term "standard excitation condition" (3.32) has been updated;

— the keys 11 and 15 in Figure 1 have been updated;

— 6.1, 6.2, 6.3 g), i), l), m), p) and q), 6.4, 6.5.1, 6.5.2, Formula (1), 6.6.1, 6.6.2 b), d), e) and f), 6.7 a), 6.8.2, 6.8.3, 6.9.2 and 6.9.3 have been revised and updated;

— Clause 7 has been revised;

— Clause 8 has been revised;

— Formulae (18), (19), and (20) have been revised;

— the first paragraph and e) in 9.2 have been revised;

— D.1 has been revised;

— URLs have been added to D.2.2, D.2.3 and D.2.4;

— D.2.4 and D.5 have been added;

— the first paragraph in Annex E has been revised;

— in Annex E, Calibration Results (For photographic film or imaging plate use) and Calibration Results (For digital camera use) have been revised;

— the figures have been modified;

— the Bibliography has been updated.

This third edition replaces the second edition (ISO 29301:2017), of which has been it constitutes a minor revision with editorial corrections. The changes are as follows:

The change compared to the previous edition is as follows:

— the element name of Silver in Table D.1 has been changedcorrected to Silicon.

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

The transmission electron microscope (TEM) is widely used to investigate the micro/nano-structure of a range of important materials such as semiconductors, metals, nano-particles, polymers, ceramics, glass, food and biological materials. The technique used involves the transmission of electrons through an ultra-thin specimen, interacting with the specimen as they pass through. This interaction results in a magnified image which is focused onto an imaging device, such as a photographic film, an imaging plate, or an image sensor built into a digital camera. A TEM is capable of imaging at significantly higher resolutions than ordinary (light) microscopes. It can be used to examine fine details as small as a single atomic column in a given specimen. This document addresses the need for magnification calibration of the images. It describes the requirements for calibration of the image magnification in the transmission electron microscope using a certified reference material or a reference material with periodic structures.

Microbeam analysis — Analytical electron microscopy — Methods for calibrating image magnification by using reference materials with periodic structures

1 Scope

This document specifies a calibration procedure applicable to images recorded over a wide magnification range in a transmission electron microscope (TEM). The reference materials used for calibration possess a periodic structure, such as a diffraction grating replica, a super-lattice structure of semiconductor or an analysing crystal for X-ray analysis, and a crystal lattice image of carbon, gold or silicon.

This document is applicable to the magnification of the TEM image recorded on a photographic film, or an imaging plate, or detected by an image sensor built into a digital camera. This document also refers to the calibration of a scale bar.

This document does not apply to the dedicated critical dimension measurement TEM (CD-TEM) and the scanning transmission electron microscope (STEM).

2 Normative references

The following documents are referred to in the text in such a way that some or all of their content constitutes requirements of this document. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

ISO 17034, *General requirements for the competence of reference material producers*

ISO Guide 35, *Reference materials~~-~~ — General and statistical principles for certification*

ISO/IEC 17025:2005, *General requirements for the competence of testing and calibration laboratories*

3 Terms and definitions

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

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

— ISO Online browsing platform: available at ~~https://www.iso.org/obp~~https://www.iso.org/obp

— IEC Electropedia: available at ~~http://www.electropedia.org/~~https://www.electropedia.org/

3.1
accuracy
closeness of agreement between a test result and the accepted reference value

Note 1 to entry: A "test result" is the calibrated magnification obtained by the procedure outlined in this document.

Note 2 to entry: The term "accepted reference value" is the magnification given by the TEM manufacturer.

[SOURCE: ISO 5725-1:1994, 3.6, modified — new Notes 1 and 2 to entry have been added.]

3.2
alignment
series of operations to align the incident direction of the electron beam to the *optical axis* (3.22) using deflectors and/or mechanical knobs

3.3
certified reference material
CRM
reference material, accompanied by a certificate, one or more of whose property values are certified by a procedure which establishes its traceability to an accurate realization of the unit in which the property values are expressed, and for which each certified value is accompanied by an uncertainty at a stated level of confidence

Note 1 to entry: For the purposes of this document, a CRM possesses periodic structure(s), with the desired range of periodic interval and accuracy, to be used for the calibration of the *image magnification* (3.15).

3.4
contamination
formation of a deposited layer of any material due to the interaction of the electron beam with the sample and/or its immediate environment

3.5
crystal orientation
direction of crystal which is represented by crystal index

Note 1 to entry: During TEM imaging, it is often useful to have a crystalline specimen aligned such that a specific (low index) *zone axis* (3.36) is parallel, or nearly parallel, to the beam direction [*optical axis* (3.22)].

3.6
diffraction grating replica
shadow-casting carbon replica film constituting a grating which contains 500 to 2 000 parallel grooves per millimetre, or cross-line grating with a similar line spacing

Note 1 to entry: A diffraction grating replica can be used as a *reference material* (3.25) for calibration of the *image magnification* (3.15) in the low to medium-low magnification range.

3.7
digital camera
device that detects the *image* (3.13) using a chip-arrayed *image sensor* (3.18), such as a charge-coupled device (CCD) or complementary metal-oxide semiconductor (CMOS), that converts a visual image to an electric signal

3.8
dynamic range
range of detectable electron doses illuminated on the detector, in which the image signal can be detected properly

3.9
excitation current
electric current applied to the coil of the magnetic lens

3.10
focus

focusing condition in which the specimen height coincides with the object plane of the objective lens

3.11
glass scale
ruler on which a fine scale is drawn and utilized as the reference scale to measure the distance in the digitized image after digitizing it with an *image scanner* (3.17)

Note 1 to entry: The transparency and thermal stability of the glass scale are convenient to get the digitized reference image with a transmitted image scanner and to make the contact image on the *imaging plate* (3.16).

3.12
horizontal field width
HFW
original length corresponding to full width in the horizontal direction on a magnified image

3.13
image
two-dimensional projection of the specimen structure generated by *TEM* (3.34)

Note 1 to entry: A *photographic film* (3.23), an *imaging plate* (3.16), and an *image sensor* (3.18) built into a digital camera are examples of devices for detecting the *image* (3.13).

[SOURCE: ISO 16700:2016, 3.2, modified — the term “SEM” has been replaced by the term “TEM”.]

3.14
image file
computer file containing information relating to the digitized image

3.15
image magnification
ratio of the linear dimension of the specific structure/scaling on the image detector, such as a *photographic film* (3.23), an *imaging plate* (3.16), and an *image sensor* (3.18) built into a digital camera, to the corresponding linear dimension of the structure/scaling on the *specimen* (3.27)

3.16
imaging plate
IP
electron image detector consisting of a film with a thin active layer embedded with specifically designed phosphors

3.17
image scanner
device that converts an analogue image into a digitized image with the desired pixel-resolution

Note 1 to entry: There are mainly two different types of scanners: flatbed type and drum type.

3.18
image sensor
device, such as a charge-coupled device (CCD) array or complementary metal-oxide semiconductor (CMOS) sensor, that converts visual image information to an electric signal, built-in digital camera or other imaging devices

3.19
image wobbler
deflection coil used to change the direction of incident electron beam onto the *specimen* (3.27)

Note 1 to entry: This coil is activated in a periodic manner with the aim of identifying easily the place of *focus* (3.10).

3.20
lattice image
image (3.13) consisting of interference fringes formed by the interaction between the transmitted electron beam and diffracted electron beam from a specific crystal plane

Note 1 to entry: Lattice fringes can be used to calibrate *image magnification* (3.15) at the high end of the magnification range.

3.21
magnetic hysteresis
physical phenomenon related to the magnetizing loop in which the magnetic field strength depends on the direction of the adjustment of the exciting current for the magnetic lens

3.22
optical axis
straight line passing through the symmetrical centre of the magnetic field of the electron lens

Note 1 to entry: The path of an electron beam along this axis goes through the lens without changing the direction.

3.23
photographic film
negative film
sheet or a roll of thin plastic coated by photographic emulsion for recording an *image* (3.13)

3.24
pixel-resolution
number of imaging pixels per unit distance of the detector

Note 1 to entry: The typical unit is sometimes expressed as dots per inch (dpi).

3.25
reference material
RM
material or substance, one or more of whose property values are sufficiently homogeneous and well-established to be used for the calibration of an apparatus, the assessment of a measurement method, or for assigning values to materials

Note 1 to entry: For the purpose of this document, an RM possesses periodic pattern(s) with the desired range of periodic interval and accuracy, to be used for the calibration of the *image magnification* (3.15).

3.26
region of interest
ROI
region of the *image* (3.13) selected for a specific reason

3.27
specimen
small portion of a sample for observation

Note 1 to entry: For *TEM* (3.34), a specimen has to be thin enough to transmit the electron beam.

3.28
specimen cartridge

part of the *specimen holder* (3.31) which supports a *specimen* (3.27) and is attached to the tip of the specimen holder for use

3.29
specimen drift
unintentional movement of the *specimen* (3.27) due to any source (thermal, mechanical, electric, charging)

3.30
specimen height
specimen position along the *optical axis* (3.22) of the objective lens

Note 1 to entry: “Specimen height = 0” corresponds to the specimen position in correct focus under the *standard excitation condition* (3.32) of the objective lens.

Note 2 to entry: See Reference [6].

3.31
specimen holder
device that supports a *specimen* (3.27) in the right position in the pole-piece gap of the objective lens

3.32
standard excitation condition
setting condition for excitation current to derive the highest performance of the objective lens

Note 1 to entry: Under this condition, *specimen height* (3.30) shall be set so that the *image* (3.13) is focused.

Note 2 to entry: This condition is provided by the TEM manufacturer for each instrument.

Note 3 to entry: *Image magnification* (3.15) is generally measured under this condition; however, as long as reproducible conditions are established, the magnification can be calibrated at any of the instrument settings.

3.33
super-lattice
stable periodic structure which is fabricated by alternating layers of at least two different kinds of materials

Note 1 to entry: The super-lattice can be used as a *reference material* (3.25) for calibration of *image magnification* (3.15) from a medium-high to high magnification range.

3.34
transmission electron microscope
TEM
instrument that produces magnified images or diffraction patterns of the *specimen* (3.27) by an electron beam which passes through the specimen and interacts with it

3.35
under focus
focusing condition in which the specimen height is further from the objective lens than its object plane

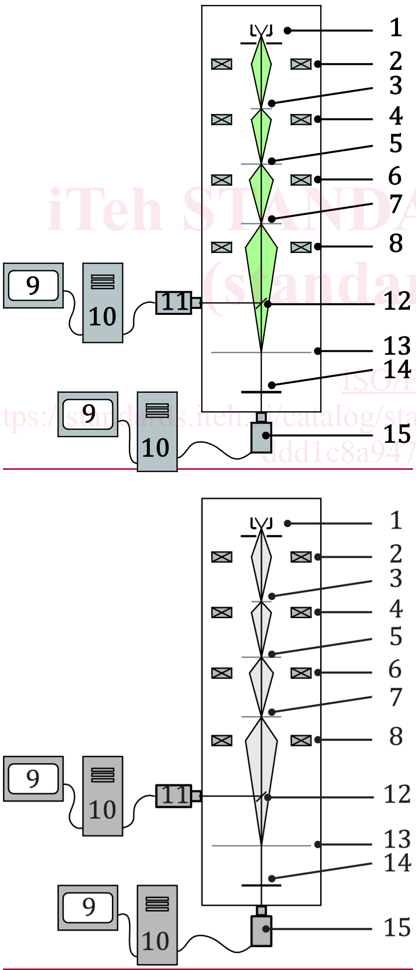
3.36
zone axis
crystallographic direction, designated [*uvw*], defined by the intersection of a number of crystal planes (h_1, k_1, l_1... ...h_i, k_i, l_i) such that all of the planes satisfy the so-called Weiss zone law; $hu + kv + lw = 0$

4 Image magnification

4.1 Definition of the image magnification

The image magnification (or scaling factor) of the TEM is defined by the ratio of the linear dimension of the specific structure on the detected image to the corresponding linear dimension of the specific structure in the specimen. There are three main kinds of image detectors: photographic film, imaging plate, and image sensor, such as CCD array or CMOS sensor built in the digital camera.

In general, the value of image magnification detected on an image sensor is different from the value of image magnification detected on the photographic film or imaging plate under the same electron optical conditions for TEM imaging, because the image-detecting positions are different from each other (see Figure 1).



Key

1 electron gun
2 condenser lens
3 specimen
4 objective lens
5 first magnified image
6 intermediate lens
7 second magnified image
8 projector lens
9 monitor
10 computer
11 digital camera (image sensor) 1
12 screen/monitor
13 viewing screen
14 photographic film/imaging plate
15 digital camera (image sensor) 2

Figure 1 — Detector position in TEM system

4.2 Expressing magnification

The magnification of an image recorded on the photographic film or the imaging plate, or detected by the image sensor, is given by a number representing the number of times, and the number is accompanied by the symbol × (e.g. 10 000×, 10k×, 1 000 000×, 1M× or ×10 000, ×10k, ×1 000 000, ×1M, where 10 000, 10k, 1 000 000 and 1M are magnitude numbers). Alternatively, introducing a scale bar having a length corresponding to unit length on the specimen can be used to represent the magnification. The digitized image should also indicate a magnification by detailing the number of pixels per unit distance of the raw data file.

NOTE The horizontal field width (HFW) is another way to define the scaling on a magnified image.

5 Reference materials

5.1 General

For calibrating the magnification of an image, wherever possible, choose a CRM that is produced in accordance with ISO 17034 and certified in accordance with ISO Guide 35.

When a suitable CRM is not available, an RM produced in accordance with ISO 17034 may be used.

5.2 Requirements for CRM/RM

Ensure that the chosen CRM/RM

— is stable with respect to vacuum and repeated electron-beam exposure,

— is aligned to a low-index zone axis along the electron optical axis, if the specimen region is a single crystal,

— provides a good contrast and clear interface for the periodic structure in the TEM image,

— can be cleaned to remove contamination without causing mechanical/electrical damage or distortion,

— has a smooth surface on both sides and identical thickness for a super-lattice structure, at least within the area used for the calibration process, and

— has an associated valid calibration certificate.

NOTE Single crystal specimens of pure elements used for calibration do not need a calibration reference certificate.