# INTERNATIONAL STANDARD

ISO 18221

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# Microscopes — Microscopes with digital imaging displays — Information provided to the user regarding imaging performance

Microscopes — Microscopes avec écrans d'affichage numérique — Informations fournies à l'utilisateur concernant la performance

# iTeh STAffichogARD PREVIEW (standards.iteh.ai)

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### **Foreword**

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The committee responsible for this document is ISO/TC 172, *Optics and photonics*, Subcommittee SC 5, *Microscopes and endoscopes*.

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# Microscopes — Microscopes with digital imaging displays — Information provided to the user regarding imaging performance

#### 1 Scope

This International Standard specifies the minimum information to be provided to the user by the manufacturers of microscopes with digital displays regarding imaging performance.

It further specifies terms and definitions for describing the optical performance of the digital imaging path of microscopy systems including the observation of the image on digital displays.

NOTE Terms and definitions for the direct visual observation with eyepieces are specified in ISO 8039 and ISO 10934-1.

#### 2 Normative references

The following documents, in whole or in part, are normatively referenced in this document and are indispensable for its application. For dated references, only the edition cited applies. For undated references, the latest edition of the referenced document (including any amendments) applies.

ISO 10934-1, Optics and optical instruments — Vocabulary for increscopy — Part 1: Light microscopy

ISO 10934-2, Optics and optical instruments — Vocabulary for microscopy — Part 2: Advanced techniques in light microscopy https://standards.iteh.ai/catalog/standards/sist/1f8ed262-2174-489e-9866-e2d6a25b2251/iso-18221-2016

#### 3 Terms and definitions

For the purposes of this document, the terms and definitions given in ISO 10934-1, ISO 10934-2 and the following apply.

NOTE Terms shown in italics within a definition or a note are defined elsewhere within this International Standard or in ISO 10934.

#### 3 1

#### digital microscopy system

instrument consisting of an *objective*, an *image sensor* (3.2) and a *digital display* (3.3) to make visible minute details that are not seen with unaided eye

Note 1 to entry: The system might include means for image enhancement, image analysis, archiving, etc.

#### 3.2

#### image sensor

device to transform an optical image into a digital image

#### 3.3

#### digital display

output device for the visual presentation of a digital image and other information

#### 3.4

#### resolving power limit

maximum spatial frequency distinguishable in object space, expressed in lp/mm

Note 1 to entry: The resolving power actually achieved will always be lower than the resolving power limit due to reduced contrast, moiré patterns, etc. In addition, it depends on sensor type, object structure, illumination, etc.

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#### 3.5

#### pixel pitch

distance between the centres of adjacent pixels (picture elements) in an image sensor (3.2) or a digital display(3.3)

#### 3.6

#### line pair

one line and one space between the lines of a spatial grid with equal line thickness and space width between lines

#### 3.7

#### viewing distance

distance between the digital display (3.3) and the observer's eye

#### Symbols and abbreviated terms 4

 $c_1$ coefficient related to the operation mode of the image sensor

 $d_{AXSYS}$ depth of field for observation of the image presented on the digital display

 $d_{VIEW}$ actual viewing distance

dview usef useful range of viewing distance

height of the object field limited by the image area in the display h<sub>LIM DIS</sub>

height of the object field limited by the sensor h<sub>LIM</sub> SEN

height of the object field of the microscopy system PRFVIFW hsys

wavelength λ

display lateral magnification standards.iteh.ai)  $M_{\rm DIS}$ 

visual display magnification M<sub>DIS VIS</sub>

useful range of the perceived visual display magnification 2174-489e-9866-M<sub>DIS</sub> VIS USEF

lateral magnification at the image projected onto the sensor M<sub>TOT PROI</sub>

total visual magnification of the virtual microscope image formed by the eyepiece M<sub>TOT VIS</sub>

NA numerical aperture

number of image pixels in the display in *x*-direction  $N_{X \text{ DIS}}$ 

 $N_{X \text{ SEN}}$ number of active sensor pixels in x-direction

number of image pixels in the display in y-direction  $N_{v \text{ DIS}}$ 

number of active sensor pixels in y-direction  $N_{V}$  SEN

n refractive index of the object or of the immersion media

pixel pitch of the digital display  $pp_{DIS}$ pixel pitch of the image sensor ppsen

display resolving power limit in object space  $RL_{DIS}$  $RL_{OPT}$ optical resolving power limit in object space RLSEN sensor resolving power limit in object space  $RL_{SYS}$ system resolving power limit in object space

width of the object field limited by the image area in the display WLIM DIS

width of the object field limited by the sensor WLIM SEN width of the object field of the microscopy system WSYS

## Magnification

#### Optical magnification, $M_{\text{TOT PROI}}$

 $M_{\rm TOT\ PROI}$  is the lateral magnification at the image projected onto the image sensor. It should be expressed in proportional form, e.g. 10:1.

#### **Display magnification**, M<sub>DIS</sub>

 $M_{\rm DIS}$  is the lateral magnification at the digital image presented on the digital display or monitor. It should be expressed in proportional form, e.g. 100:1.

The display magnification, M<sub>DIS</sub>, is the ratio of a given distance in the image presented on the monitor to the corresponding distance in the object.

#### Visual display magnification, $M_{DIS \ VIS}$

 $M_{\rm DIS~VIS}$  is the display lateral magnification by observing the digital image presented on the digital display. It should be expressed in numerical form with the multiplication sign, e.g. 50x.

The visual display magnification is given by:

$$M_{\text{DIS VIS}} = \frac{M_{\text{DIS}} \cdot 250}{d_{\text{VIEW}}}$$
 (1)

ere

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where

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 $M_{DIS\,VIS}$  is the visual display magnification;

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is the display lateral magnification;/sist/1f8ed262-2174-489e-9866-**MDIS** 

is the reference viewing distance in mm; 250

dview is the actual viewing distance in mm.

NOTE The value of  $M_{\text{DIS VIS}}$  is comparable with the value of  $M_{\text{TOT VIS}}$  of light microscopes with eyepieces.

# Resolving power limit

#### Optical resolving power limit, $RL_{OPT}$

 $RL_{OPT}$  is the lateral resolving power limit in object space that is limited by the optical system. It should be expressed in line pairs per millimetre, e.g. 200 lp/mm. It is given by:

$$RL_{\rm OPT} = \frac{2 \cdot NA \cdot 10^6}{\lambda} \tag{2}$$

where

 $RL_{OPT}$  is the optical resolving power limit in object space in line pairs per millimetre;

NA is the effective numerical aperture at object side of the optical system projecting the image onto the sensor;

λ is the wavelength in nm.

NOTE 1 The formula for  $RL_{OPT}$  does not apply for super-resolution. NOTE 2 In consideration of consistency with the definition of resolving limit of electric equipment, the optical resolving limit defined in this International Standard is based on the cut-off frequency of an optical system.

# **6.2** Sensor resolving power limit, $RL_{SEN}$

 $RL_{SEN}$  is the lateral resolving power limit in object space that is limited by the image sensor. It should be expressed in line pairs per millimetre, e.g. 100 lp/mm. It is given by:

$$RL_{\text{SEN}} = \frac{500 \cdot M_{\text{TOT PROJ}}}{c_1 \cdot pp_{\text{SEN}}} \tag{3}$$

where

*RL*<sub>SEN</sub> is the sensor resolving power limit in object space in line pairs per millimetre;

 $M_{\rm TOT\,PROI}$  is the lateral magnification at the image projected onto the sensor;

 $c_1$  is a coefficient related to the operation mode of the image sensor;

 $pp_{SEN}$  is the pixel pitch of the image sensor in  $\mu$ m.

EXAMPLES for  $c_1$ 

- 1 corresponds to a standard or full frame mode;
- 2 corresponds to a 2 × 2 to binning mode; 2 corresponds to a 2 × 2 to binning mode;
- 3 corresponds to a 3 × 3 binning mode; and ards.iteh.ai)
- 0,5 corresponds to a pixel shift mode with a lateral movement of the image sensor of 0,5 *pp*<sub>SEN</sub>.

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# 6.3 Display resolving power limit, R2-d625b2251/iso-18221-2016

 $RL_{\rm DIS}$  is the lateral resolving power limit in object space that is limited by the digital display. It should be expressed in line pairs per millimetre, e.g. 50 lp/mm. It is given by:

$$RL_{\rm DIS} = \frac{M_{\rm DIS}}{2 \cdot pp_{\rm DIS}} \tag{4}$$

where

RL<sub>DIS</sub> is the display resolving power limit in object space in line pairs per millimetre;

 $M_{\rm DIS}$  is the display lateral magnification;

*pp*<sub>DIS</sub> is the pixel pitch of the digital display in mm.

# **6.4** System resolving power limit, $RL_{SYS}$

 $RL_{SYS}$  is the lateral resolving power limit in object space of the microscopy system. It should be expressed in line pairs per millimetre, e.g. 50 lp/mm. Its value corresponds to the smallest value of either the optical resolving power limit or the sensor resolving power limit or the display resolving power limit, for example:

$$RL_{SYS} = \min(RL_{OPT}; RL_{SEN}; RL_{DIS})$$
(5)

where

*RL*<sub>SYS</sub> is the system resolving power limit in object space in line pairs per millimetre;

 $RL_{OPT}$  is the optical resolving power limit in line pairs per millimetre;

*RL*<sub>SEN</sub> is the sensor resolving power limit in object space in line pairs per millimetre;

*RL*<sub>DIS</sub> is the display resolving power limit in object space in line pairs per millimetre.

NOTE If  $RL_{SYS}$  is smaller than  $RL_{OPT}$ , the image quality might be limited by moiré effects, especially when inspecting periodically structured specimens.

# 7 Useful range of magnification

#### 7.1 Useful range of viewing distance

Within the useful range of viewing distance, resolved details are seen by the observer's eye under an angle between 2,3' and 4,6' (minutes of arc). Its value is given by:

$$d_{\text{VIEW USEF}} = \frac{60}{(2,3..4,6)} \cdot \frac{180}{\pi} \cdot \frac{M_{\text{DIS}}}{RL_{\text{SYS}}}$$
(7)

where

 $d_{\text{VIEW USEF}}$  is the useful range of viewing distance in mm;

2,3 .. 4,6 is the range of angles in minutes of arc that allows to resolve 1 line pair by the observer's eye;

 $M_{\rm DIS}$  is the display lateral magnification: 16

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*RL*<sub>SYS</sub> is the system resolving power limit in object space in line pairs per millimetre.

#### 7.2 Useful range of visual magnification

The perceived visual magnification is within the useful range of magnification if the actual viewing distance is in its useful range. The value of the useful range of magnification is given by:

$$M_{\text{DIS VIS USEF}} = \frac{(2,3..4,6)}{60} \cdot \frac{\pi}{180} \cdot 250 \cdot RL_{\text{SYS}} = \frac{RL_{\text{SYS}}}{(3..6)}$$
(8)

where

 $M_{\rm DIS\,VIS\,USEF}$  is the useful range of the perceived visual display magnification;

2,3 .. 4,6 is the range of angles in minutes of arc that allows to resolve 1 line pair by the observer's eye;

is the reference viewing distance in mm;

*RL*<sub>SYS</sub> is the system resolving power limit in object space in line pairs per millimetre.

NOTE 1  $M_{\rm DIS~VIS~USEF}$  is comparable with the useful range of magnification for visual observation of light microscopes with eyepieces which is usually taken to lie between 500 and 1 000 times the numerical aperture of the objective.

NOTE 2 When the actual value of the perceived visual display magnification is less than the lower limit of  $M_{DIS\ VIS\ USEF}$ , then the resolving power limit of the digital microscopy system cannot be fully utilized.