ABOUT RESOLUTION

The resolution of an image sensor describes the total number of pixel which can be used to detect an image. From the standpoint of the image sensor it is sufficient to count the number and describe it usually as product of the horizontal number of pixel times the vertical number of pixel which give the total number of pixel, for example:

$$no.\,pixel_{hor} \times \; no.\,pixel_{ver} = total\; no.\,pixel$$

Or take as an example
the sCMOS image sensor CIS2521:

$$(2560_{hor} \times 2160_{ver})\; pixel = 5.5\; Mpixel$$

That is the usual information found in technical data sheets and camera advertisements, but the question arises on “what is the impact or benefit for a camera user”?

1 Benefit For A Camera User

It can be assumed that an image sensor or a camera system with an image sensor generally is applied to detect images, and therefore the question is about the influence of the resolution on the image quality. First, if the resolution is higher, more information is obtained, and larger data files are a consequence. Second, the amount of information, which can be obtained by a camera, is inseparably connected to the applied imaging optics, which are characterized by their own optical resolution or ability to resolve details, therefore this has to be taken into account as well.

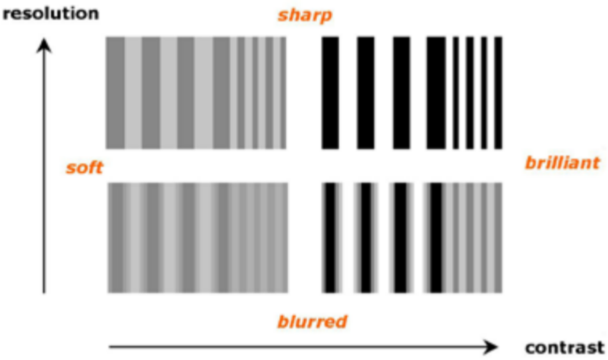

Figure 1
Illustration of the influence of resolution and contrast on image quality.

In figure 1 the influence of resolution and contrast on the image quality is illustrated. The higher the resolution of an optical system consisting of a camera and imaging optics, the sharper the images, while the contrast controls the range of soft or brilliant perception.

2 Image Sensor & Camera

Starting with the image sensor in a camera system, then usually the so called modulation transfer function (MTF) is used to describe the ability of the camera system to resolve fine structures. It is a variant of the optical transfer function[1] (OTF) which mathematically describes how the system handles the optical information or the contrast of the scene and transfers it onto the image sensor and then into a digital information in the computer. The resolution ability depends on one side on the number and size of the pixel.

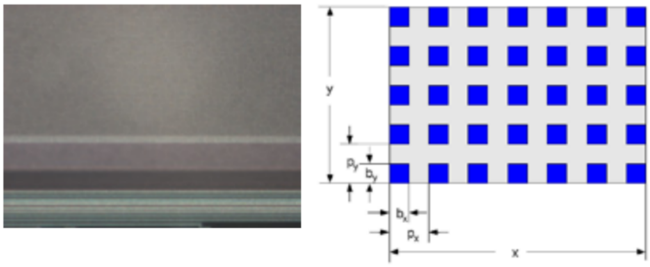

Figure 2
a) a macro image of an image sensor showing the pixels at the edge of the imaging area, b) illustration of an image sensor with characteristic geometrical parameters: x, y - horizontal, vertical dimensions, p_x, p_y - horizontal, vertical pixel pitch, b_x, b_y - horizontal, vertical pixel dimensions

The maximum spatial resolution is described as ability to separate patterns of black and white lines and it is given in line pairs per millimeter ([lp/mm]). As theoretical limit it is described in the literature and comprehensive that the maximum resolution is achieved if one black line is imaged on one pixel while one white line is imaged to the neighbor pixel.

Assuming square pixel with $b_x = b_y = b$ and $p_x = p_y = p$ (see fig. 2 pixel schematic) then the maximum possible axial R_{axial} and diagonal $R_{diagonal}$ resolution ability is just given by the dimensions of the pixel:

$$R_{axial} = \frac{1}{2 \cdot p} \qquad R_{diagonal} = \frac{1}{\sqrt{2} \cdot 2 \cdot p}$$

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[1] https://en.wikipedia.org/wiki/Optical_transfer_function

ABOUT RESOLUTION

In the following table there are the maximum resolution ability values for image sensors with different pixel sizes given.

Table 1: Maximum Theoretical MTF Data Of Image Sensors

item	image sensor/ lens type	pitch [µm]	R_{axial} [lp/mm]	$R_{diagonal}$ [lp/mm]
ICX285AL	CCD	6.45	77.5	54.8
MT9M413	CMOS	12	41.7	29.5
GSENSE5130	sCMOS	4.25	117.7	83.2

The contrast which is transferred through the optical system consisting of camera and imaging optics is defined as contrast or modulation M, which is defined with the intensity I [count] or [DN^2] in an image:

$$M = \frac{I_{max} - I_{min}}{I_{max} + I_{min}}$$

The modulation depends on the spatial frequencies, which means that M is a function of the resolution R: M = M(R). The quality of the imaging process is described by the modulation transfer function, MTF. So both parameters, the resolution and the contrast, define the quality of an image, as is illustrated in figure 1. Increasing resolution improves the sharpness of an image while increasing contrast increases the "brilliance".

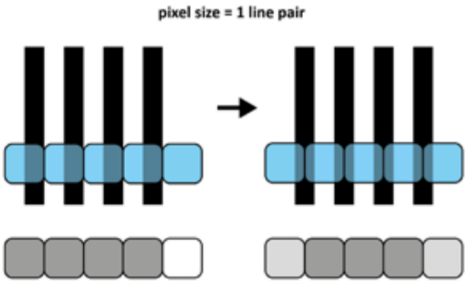

Figure 3
Illustration of a line pair structure which is imaged to one row of pixel with a pitch similar to the width of the line pair. Left: the structure is imaged in a way that each pixel "sees" a line pair. The pixel row below shows the resulting measured light signal of the corresponding pixel. Right: the structure is shifted compared to the pixel row. Again the pixel row below shows the resulting measured light signal of the corresponding pixel above.

But this is only the maximum possible MTF and requires the measuring pattern to be exactly adjusted, if the line pair pattern is shifted by half a pixel, nothing could be seen as shown in figure 4. This is illustrated by three different use cases. Let us assume the structure to be resolved is given by these black and white line pairs. Then figure 3 shows what happens, if the pixel of an image sensor has the same pitch like the width of one line pair.

In this case the structure could never be resolved, even if it is moved, the resulting light information (see fig.3 pixel rows below) is not able to give enough information about the structure. If now the theoretical maximum MTF is assumed, we come to the illustration in figure 4.

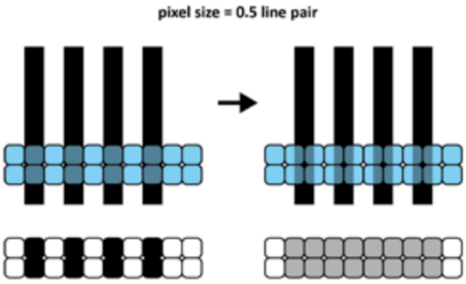

Figure 4
Illustration of a line pair structure which is imaged to one row of pixel with a pitch similar to half the width of the line pair. Left: the structure is imaged in a way that each pixel "sees" either a black or a white line. The pixel row below shows the resulting measured light signal of the corresponding pixel. Right: the structure is shifted compared to the pixel row, now the pixel see always half white and half black. Again the pixel row below shows the resulting measured light signal of the corresponding pixel above.

Only in case that the structure is imaged in a way, that each pixel "sees" either black or white, the maximum MTF can be reached. In case the structure is shifted by half a pixel all the information is gone, and nothing can be resolved. Therefore the maximum theoretical MTF value is a nice start, in case the user has to estimate some starting values for the imaging optics, which should be used with a camera system. A more practical case and condition is shown in figure 5.

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[2] DN = digital number, like count

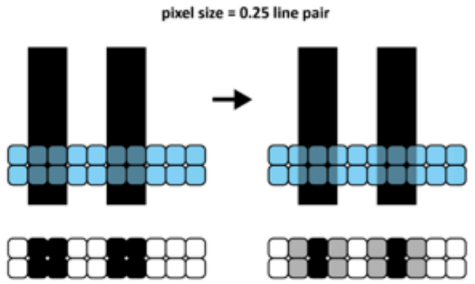

Figure 5
Illustration of a line pair structure, which is imaged to one row of pixel with a pitch similar to the quarter of the width of the line pair. Left: the structure is fully resolved by the pixel. The pixel row below shows the resulting measured light signal of the corresponding pixel. Right: the structure is shifted compared to the pixel row, still the structure can be resolved with a little bit less sharpness compared to the left image. Again the pixel row below shows the resulting measured light signal of the corresponding pixel above.

Now the pixel pitch corresponds to the quarter of the line pair width (see fig. 5). In this case the structure can be always resolved with more or less sharpness, even if the structure is not optimum positioned on the pixel row. Therefore in each imaging application for structures which have to be resolved it is important to match the selected imaging optics to the resolution and the pixel size to the image sensor in the camera system, to finally get best possible results.

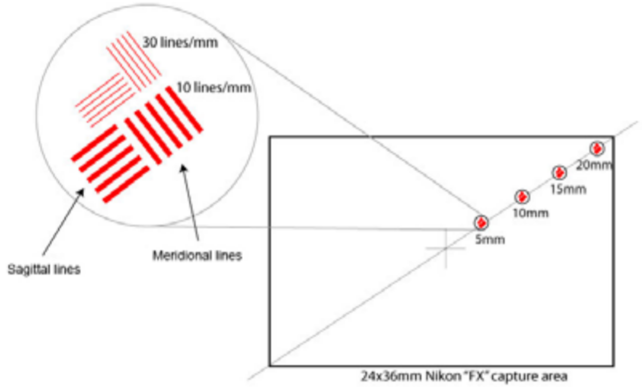

Figure 6
Illustration of the orientation of the test patterns for MTF measurements of camera lenses (taken from: https://www.nikonusa.com/en/learn-and-explore/a/products-and-innovation/what-is-a-lens-mtf-chart-how-do-i-read-it.html).

3 Imaging Optics – Camera Lens

Lens manufacturers like Zeiss, Nikon, Canon offer either simulated MTF curves of their lenses or measured data and also provide material how to understand and use these lens MTF charts or curves [3, 4, 5].

For example Nikon distinguishes two groups of data plotted on a MTF chart, they call them sagittal and meridional lines. Zeiss calls these lines sagittal and tangential, and it is about how a parallel line pair pattern is oriented compared to the image sensor.

In the definition of Nikon the "Sagittal lines" (the solid lines) represent the contrast measurements of pairs of lines that run parallel to a central diagonal line that passes through the middle of the lens from the bottom left hand corner to the top right hand corner. The "meridional lines" (see fig. 7, the dotted lines) represent line pairs also positioned along an imaginary line from the center of a lens to the edge but these line pairs are perpendicular to the diagonal line.

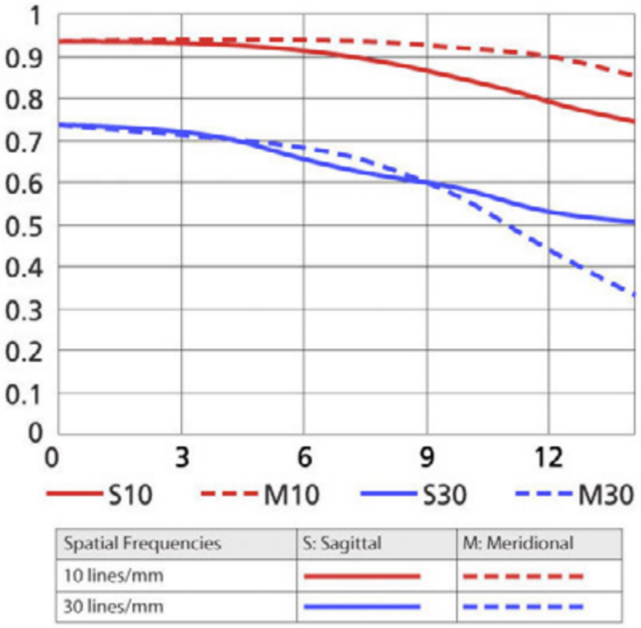

Figure 7
Nikon lens MTF chart example for different linepair resolutions: 10 lines/mm and 30 lines/mm (taken from: https://www.nikonusa.com/en/learn-and-explore/a/products-and-innovation/what-is-a-lens-mtf-chart-how-do-i-read-it.html).

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[3] https://www.nikonusa.com/en/learn-and-explore/a/products-and-innovation/what-is-a-lens-mtf-chart-how-do-i-read-it.html#
[4] https://photographylife.com/how-to-read-mtf-charts
[5] https://www.zeiss.com/content/dam/Photography/new/pdf/en/cln_archiv/cln31_en_web_special_mtf_02.pdf

The company Nikon shows two groups of test lines for each Sagittal and Meridional value: one set of line pairs at 10 lines per millimeter (resolution 100 µm) and a second set at 30 lines per millimeter (resolution 33 µm). The lower line pairs (10 lines/mm) will generally give better results than the more challenging fine resolution 30 lines/mm. In figure 7 in the graph the y-axis gives the contrast or modulation M value in relation to the distance from the center of the image circle (which would be the center of an image sensor as well).

More information can be found on the web if phrases like “how to read MTF curves” or “how to understand MTF charts” are retrieved.

4 Imaging Optics - Microscope Objective

In microscope applications the situation is a little bit more complex since there are typically not MTF charts available and a couple of lenses or objectives are involved until the image reaches the camera. But there are characteristic parameters and physical relationships that help to figure out what the best possible resolution is.

In microscopy there is the so-called “Rayleigh-Criterion” (see fig. 8) which describes the minimum distance between two objects which can be separated as a function of the numerical aperture (NA) of the objective and the spectral wavelength of the light that should be detected. In a simplified way it is given by:

$$d = \frac{0.61 \cdot \lambda}{NA}$$

(with distance d = width of line pair, λ wavelength and numerical aperture NA of the objective). The major parameters of each microscope objective are the magnification Mag_{obj} and the numerical aperture NA.

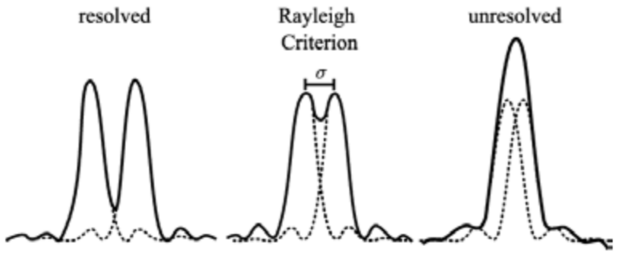

Figure 8:
Schematic to illustrate the Rayleigh criterion, if two point signals (dotted curves) which can be resolved (left graph, solid line is the impression of the optical system) approach each other, they reach a minimum distance, in which they still can be resolved (middle graph, Rayleigh criterion, solid line is the impression of the optical system). If the distance is further decreased, both signals cannot be resolved and they are perceived as one signal (right graph, unresolved, solid line is the impression of the optical system)[7].

Table 2: Parameters Of Microscope Objectives

Objective	Mag_{obj}	NA
CFI Plan Apochromat Lambda 4X Oil	x 4	0.2
CFI Plan Apochromat Lambda 40XC	x 40	0.95
CFI Plan Apochromat Lambda 60X Oil	x 60	1.4
Objective Fluar 5x/0.25	x 5	0.25
Objective Clr Plan-Neofluar 20x/1.0	x 20	1.0
Objective I Plan-Apochromat 63x/1.4 Oil	x 63	1.4

The total magnification of the object on the microscope stage is defined as magnification of the microscope objective multiplied by the magnification of the so called TV- or camera-adapter, which consists of a lens with c-mount and mount to the microscope which serves as “ocular” for the camera. Therefore the total magnification Mag to be considered is:

$$Mag = Mag_{obj} \cdot Mag_{CamAd}$$

From the chapter before it was concluded that the optimum pixel size or pitch should be equal to a quarter of the line pair width which corresponds to the minimum resolvable distance.

$$pixel\ pitch_{opt} = 0.25 \cdot width\ of\ line\ pair = 0.25 \cdot d$$

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[7] Schematic taken from: https://www.researchgate.net/publication/298070739_Tomographic_reconstruction_of_combined_tilt-_and_focal_series_in_scanning_transmission_electron_microscopy

ABOUT RESOLUTION

If now the Rayleigh criterion is inserted for d and the total magnification of the optical path in the microscope is included, the pixel $\text{pitch}_{\text{opt}}$ can be expressed as follows:

$$pitch_{opt} = \left(\frac{0.25 \cdot 0.61 \cdot \lambda}{NA}\right) \times \left(Mag_{obj} \cdot Mag_{CamAd}\right)$$

To illustrate the consequences, let's take an example: an objective with Mag_{obj} = 60 and NA = 1.4, the camera adapter has a $\text{Mag}_{\text{CamAd}}$ = 1.0 and blue-green fluorescence with λ = 514 nm should be observed:

$$pitch_{opt} = \left(\frac{0.25 \cdot 0.61 \cdot 0.514}{1.4}\right) \times (60.0 \cdot 1.0)[\mu m] = 3.4\ [\mu m]$$

This means a relatively small pixel pitch. Just in case an objective would be used with a smaller NA, for example like NA = 0.93, the resulting optimum pixel pitch would be 5.2 µm. The result is similar sensitive towards the correct chosen magnification of the camera adapter, if it is for example smaller like MagCamAd = 0.5, the optimum pixel pitch would be 1.7 µm. As well if we just apply the theoretical limit of 0.5 times the width of the line pair, it would result in 6.8 µm.

Or it is possible to take an existing pixel pitch, which is popular for emCCD and some new sCMOS image sensors like 11 µm and ask what the optimum magnification Mag_{obj} of an objective is, if we assume an NA around 1.

$$Mag_{obj\ opt} = \frac{pixel\ pitch \cdot NA}{0.25 \cdot 0.61 \cdot \lambda} \times \frac{1}{Mag_{CamAd}}$$

With a pixel pitch = 11 µm, NA = 1.0, MagCamAd = 0.7 and the same wavelength like before λ = 514 nm we would get:

$$Mag_{obj\ opt} = \frac{11 \cdot 1.0}{0.25 \cdot 0.61 \cdot 0.514} \times \frac{1}{0.7} = 200.5$$

This is well above the largest common magnifications of 150 for microscope objectives. The value could be optimized by a larger magnification of the camera adapter, but this would reduce the imaged area compared to the area as seen through the oculars.

It might be possible that the optimum value is not achieved. Nevertheless attention has to be taken on a proper selection of objective and camera when a camera should be used at a microscope in a specific application.

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