MAGUS

CHOOSING A MICROSCOPE CAMERA

A digital microscope camera is a professional device that displays the sample image on a computer screen or monitor. The camera is used to take photos and video, save information for archiving and visual presentation, or for online streaming.

When choosing how to travel, we first have in mind the tasks it is intended to solve, and only then do we decide on the size of the vehicle: In one case, an airplane is a perfect fit, in another case, we choose a tanker, and in a third case, a bicycle is the best option. It is the same with a digital camera: First, you need to understand what tasks the camera needs to accomplish, and then relate them to the techniques used in the microscope. The typical tasks that the camera can be used for: documenting or only online presentation of the observation results; photo or video shooting; image processing or only output onto a screen. The techniques employed in the microscope: manipulation under the microscope or only observation; using low magnification or a 100x objective; using the brightfield or darkfield, fluorescence or phase-contrast techniques; observation of moving or stationary objects.

Camera descriptions include the concepts of sensor, pixel, and resolution. **The camera matrix, or sensor**, is a chip that consists of light-sensitive elements (photodiodes). In a nutshell, this is how it works: The microscope objective forms an image (light), photodiodes receive light, form an electrical signal, and then convert it into a digital one. One diode is one **pixel. The sensor resolution** is determined by the number of photodiodes: The more pixels there are, the higher the resolution. Manufacturers offer microscope cameras with resolutions ranging from 0.3MP to 40MP. The customer chooses a camera with a higher resolution hoping to get better image quality, but it does not work that way.

In this article, we will explore what camera features you should pay attention to in order to get the sharpest and most detailed image possible.

HOW THE SENSOR RESOLUTION AND SIZE RELATE TO THE MICROSCOPE OPTICS

The camera is selected to match a specific microscope. Therefore, when choosing a camera, you should take into account the optical properties of the microscope and particularly the objective because only the objective contributes to the object image formation for the camera.

The key characteristic of an objective is the resolution limit. The resolution limit is the shortest distance between two points or lines at which these points or lines will be distinguishable and not merge into one.

The resolution limit of a 100x objective with an aperture of 1.25 is 0.20–0.35μm, and the resolution limit of a 4x objective with an aperture of 0.1 is 2.77–3.38μm.

Therefore, when choosing the camera resolution, you should refer to the objective which you are more likely to use during observations. The effective resolution of the camera also depends on the sensor size and the magnification of the optical adapter.

When observations are made with 100x objectives, 1.7MP to 5MP cameras will be up to the task. More pixels will not have any additional impact on the image quality. When low-magnification objectives are used, on the contrary, the extra pixels will allow for more detail. Therefore, for a 4x objective and for a stereomicroscope, cameras of 5MP and above are suitable. The camera resolution is also directly related to the sensor size: The larger the sensor size is, the more the resolution will be effective.

For example, if you mount an 18MP camera with a 1/2.3" sensor size on a microscope when working with a 100x objective, the image quality will degrade. The quality degradation will occur due to the lowered signal-to-noise ratio and operation at increased sensitivity settings. A 3MP camera with a 1/1.2" sensor size, on the other hand, will produce sharp images with a 100x objective and a 40x objective.

Below is a table where the average 555nm wavelength and optical adapters that fit a particular sensor size were used to calculate the camera resolution. The familiar Abbe formula $d = \lambda/2NA$, where λ is the wavelength and NA is the numerical aperture of the objective, was used to determine the resolution limit of the objective. The product of the objective magnification and its resolution limit gives the image of the point to be read out by a camera. The camera distinguishes two points if the distance between them is at least 0.4 of the Airy disk diameter. The table is calculated based on these inputs.

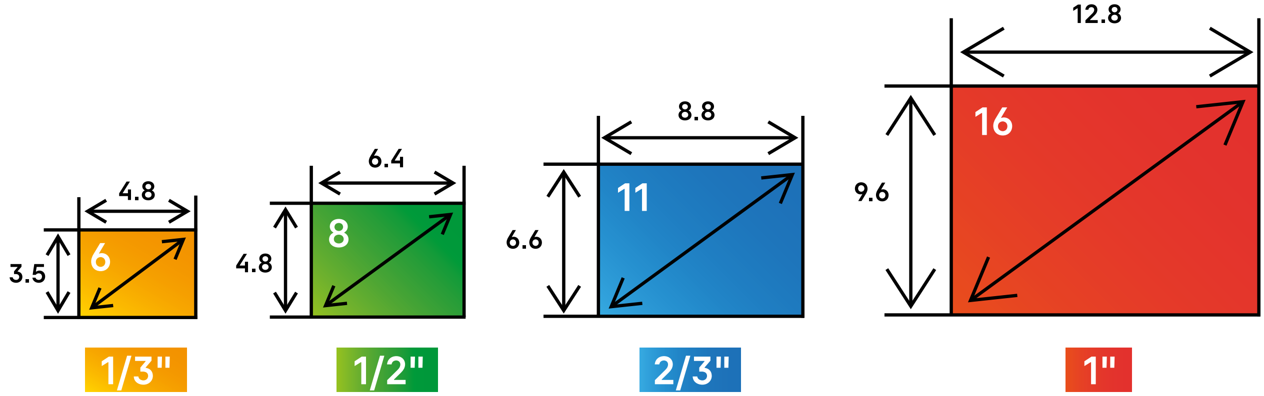
sensor size/ adapter objective magnification/aperture	1/2.5" 0.5x	1/2.3" 0.5x	1/2" 0.5x	1/1.8" 0.5x	2/3" 0.75x	1/1.2" 0.75x	1" 1x	1.1" 1x	4/3" 1x
4x/0.10	5.04	5.85	6.02	8.03	5.46	6.40	5.91	7.36	11.68
10x/0.25	5.04	5.85	6.02	8.03	5.46	6.40	5.91	7.36	11.68
20x/0.40	3.23	3.74	3.86	5.14	3.50	4.10	3.78	4.71	7.48
40x/0.65	2.13	2.47	2.55	3.39	2.31	2.71	2.50	3.11	4.94
60x/0.80	1.43	1.66	1.71	2.28	1.55	1.82	1.68	2.09	3.32
100x/1.25	1.26	1.46	1.51	2.01	1.37	1.60	1.48	1.84	2.92

The optics errors and the nature of the sample structure are not considered in the accurate theoretical calculation of the camera resolution, and so it is recommended to choose a higher camera resolution than what is calculated in theory.

Let us explore some more parameters to consider when choosing a microscope camera.

SENSOR SIZE

The sensor size is the diagonal length expressed in inches. Microscope cameras use the following typical sensor sizes: 1/4", 1/3", 1/2.8", 1/2.5", 1/2.3", 1/2", 1/1.8", 2/3", 1/1.2", 1", 1.1", 4/3". The actual measurement unit is not an inch, but rather the Vidicon inch, which is 2/3 of a full-size imperial inch. Sensor side lengths are indicated in millimeters.



A larger sensor offers the following benefits:

- Less noise and, therefore, a cleaner image: This is important when using the darkfield or fluorescence techniques.
- More light and, therefore, more shades and color depth, which is important for the polarized light microscopy and when observing monochrome samples.
- Larger field of view: If you choose the correct C-mount adapter, the field of view of the camera image approaches the field of view in the eyepieces.
- The larger the sensor is, the more larger photodiodes will fit, which means that the two tasks of increasing the light sensitivity and resolution are met.

The 1/2.5" sensor accommodates 5.1mn pixels measuring 2.2x2.2µm, while the 4/3" sensor accommodates 21mn pixels measuring 3.3x3.3 µm.

A sensor of 1/3"–1/2.5" is suitable for an educational microscope, and 1/2.3" and more is for a laboratory microscope. However, for the professional fluorescence microscopy, it is best to choose a sensor of at least 1/1.2".

PHYSICAL PIXEL SIZE

The pixel size corresponds to the length of the photodiode sides in microns. In microscope cameras, the pixel size varies from 1.25x1.25μm to 9x9μm.

The larger the pixel is, the more light it will absorb, which means less image noise and higher light sensitivity of the camera. Let us compare two cameras with the same 1/1.2" sensor size. The MAGUS CDF50 camera features a 5.8x5.8μm pixel sensor. The MAGUS CDF30 camera features a 2.9x2.9μm pixel sensor. The light sensitivity of the first camera is 1.5 times higher than that of the second one.

When choosing between cameras with the same sensor size but a different resolution, keep in mind that the camera with the larger physical pixel size, i.e. lower resolution, has higher light sensitivity.

The 3x3μm pixel sensor is suitable for the brightfield technique with 40x, 60x, and 100x objectives. For the darkfield microscopy with these objectives, it is recommended to choose a pixel size of at least 3.5x3.5μm. For shooting in the fluorescence microscopy, sensors with a pixel size of 5x5μm or larger will be a perfect fit.

HOW TO CHOOSE A CAMERA ADAPTER

A camera adapter is a mechanical device for mounting a camera on a microscope. Adapters vary in design and magnification. The adapter design is determined based on the microscope, on which the camera is mounted. The adapter magnification is selected to match the camera.

The market offers two types of adapters: original adapters for mounting a camera into the microscope's camera port and universal eyepiece adapters. One end of both types of adapters is fitted with a C-mount thread onto which a standard camera is screwed. The other end is mounted into the microscope and varies in design and appearance. The design of the original C-mount adapter matches the design and optical properties of the camera port of the specific microscope model. Original adapters are supplied with MAGUS trinocular microscopes.

The other type is universal eyepiece C-mount adapters mounted into the eyepiece tube with a 23.2mm, 30mm, or 30.5mm inside diameter instead of the regular eyepiece. Such universal C-mount adapters are not supplied with microscopes or MAGUS camera sets, and they are purchased separately if there is a need to mount a camera into the binocular microscope.

Original adapter

Universal eyepiece adapter

The adapter magnification is selected based on the camera sensor size and the purpose of the camera. The typical requirement for adapter optics is to show as large a field of view as possible with no distortion. The following adapters are used to comply with this requirement:

— 0.37x or 0.35x for 1/4"–1/3" sensors

— 0.5x for 1/3"–1/1.8" sensors

— 0.63x for 1/2"–2/3" sensors

— 0.75x for 1/1.8"–1" sensors

— 1x for 1/1.2" sensors and larger.

Unconventional tasks can be solved by additional adapters:

1. If the task is to cut the field of view and get the highest possible magnification on the screen, you will need an adapter with a higher magnification – 0.75x or 1x.

Example photo from CBF10 camera (18MP, 1/2.3'' (6.14x4.61mm)) on MAGUS STEREO 9T stereomicroscope at 0.7x objective magnification with the 1x adapter.

2. The 0.37x or 0.5x reduction adapter will be up to the task of obtaining the largest possible field of view.

Example photo from CBF10 camera (18MP, 1/2.3'' (6.14x4.61mm)) on MAGUS STEREO 9T stereomicroscope at 0.7x objective magnification using the 0.37x adapter.

You should keep in mind:

— If we fit the circle of the objective field of view into the monitor rectangle in this way, the edges of the adapter will appear in the image.

— The 0.37x adapter, when using 4x and 10x objectives, reduces the image resolution of cameras with a pixel size of 6μm and above.

3. If the task is to show both the maximum field of view and the highest possible resolution, you will need to install on the adapter a 1x camera with a large sensor diagonal and high resolution.

Example photo from CBF70 camera (21MP, 4/3'' (17.4x13.0mm)) on MAGUS STEREO 9T stereomicroscope at 0.7x objective magnification using the 1x adapter.

The photos are compressed when placed in an article, and so the difference in the image quality between the second and third examples is not apparent. In the original photos, the difference in the image quality is evident.

CAMERA: COLOR AND MONOCHROME

The cameras are available in monochrome and color versions.

A sensor is initially monochrome – black and white. A pixel can only accumulate light and does not distinguish between colors. A monochrome sensor responds equally to light according to its own sensitivity.

A color sensor is produced from a monochrome one. Four color filters are applied on each pixel of a monochrome sensor: 1 red, 1 blue, and 2 green. Four microlenses are applied on top of the filters to collect light. The sensor pixels show the intensity of these colors due to color filters. At the same time, part of the light is reflected from the filters, which reduces the total sensitivity of the pixel.

The same sensor, made in black-and-white and color versions, differs in specifications: The light sensitivity and frame rate of the monochrome sensor are on average 2–4 times higher. Therefore, a monochrome image will be brighter and more contrast at the same light level.

Let us compare two cameras with the same sensor of 1.7MP, 1.1" size, and 9x9μm pixel in terms of the light sensitivity and frame rate.

	Light sensitivity	Frequency
MAGUS CLM50 color camera	4910mV	33fps
MAGUS CLM70 monochrome camera	8100mV	94fps

However, both cameras are priced nearly the same.

A color camera is suitable when color is important for highlighting or classifying an object being observed.

A monochrome camera is suitable for applications that utilize low-light contrast microscopy, such as darkfield or fluorescence techniques.

FRAME RATE

The frame rate, or FPS (frames per second), is the number of frames that the camera captures per second. The frame rates of microscope cameras vary from 2 to 125 frames per second. The claim that the human eye does not perceive frame rates higher than 24FPS is a myth that originated in the realm of cinematography. The motion picture industry adopted 24 frames for cost efficiency reasons, as this was the minimum frame rate that produced realistic video and maintained acceptable sound during playback. Increasing the speed would increase the financial cost.

The actual threshold of the human visual perceptibility is 1000FPS. In everyday life, humans do not live at the limits of vision, on average seeing and processing up to 150 frames per second and only reaching 250FPS with regular training. These are gamers, fighter pilots, and car racers.

Choose the camera with a high frame rate for studying live objects under a microscope, manipulation under a stereomicroscope, and screen presentation in a student classroom. For shooting stationary objects, a camera with a lower FPS will do, as the frame rate is only important for adjusting the focus of the microscope.

A 5FPS camera is not suitable for the convenient setup of the microscope, nor for capturing moving objects or manipulating under the microscope. Therefore, cameras with such FPS are not included in the lineup of MAGUS professional cameras.

The MAGUS CBF50 camera with 53FPS frame rate will make it easy to adjust the focus even with a 100x objective.

If you perform manipulations under a stereomicroscope, the MAGUS CHD20 camera with 60FPS frame rate will be up to the task.

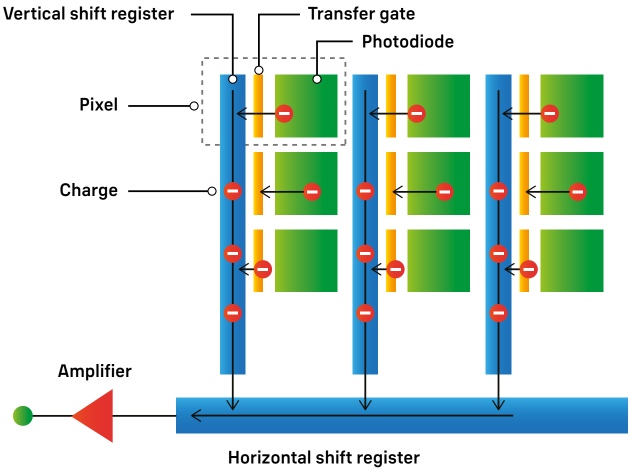
A 20FPS camera will ensure a convenient setup of the microscope at low magnification, while 50FPS will be required at 100x objective magnification.

A camera with 60FPS and above will allow performing manipulations under a stereomicroscope, shooting fast-paced processes or moving objects.

TYPE OF SENSOR

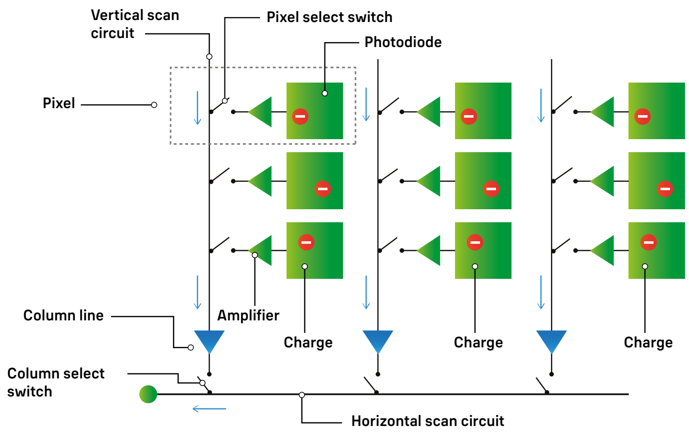
Microscope cameras employ two types of sensors: CCD and CMOS. The difference between the sensors lies in the technique of converting the signal that comes from the pixel.

The CCD (Charge Coupled Device) sensor operates as follows: The charge between sensitive elements is sequentially transferred from pixel to pixel with a minimum of noise. The charges are shifted along the sensor in rows from top to bottom. Before leaving the sensor, the charge is amplified, and the analog signal is forwarded to a dedicated analog-to-digital converter. The digital data is converted into bytes that represent an image string. The sensor is slow and energy consuming, but it creates less interference and, therefore, produces less output noise. The complex electronic circuitry adds to the cost of a camera with a CCD sensor.



CCD sensor architecture

A CMOS (Complementary Metal Oxide Semiconductor) sensor operates differently: The information processing unit is placed next to each pixel. The sensor is fast and less energy-consuming, but this technique adds noise.



CMOS sensor architecture

Technology is constantly improving. Producers pay more attention to CMOS sensors. Therefore, CMOS cameras are experiencing a major development. CMOS sensors have improved low-light performance due to SONY's backlight technology. This is how the lack of light sensitivity was corrected.

The image quality of CMOS sensor cameras is often as good as that of CCD cameras. CMOS cameras consume less power and run faster.

It costs less to manufacture these cameras, and so the price of CMOS cameras is lower.

Let us compare two monochrome fluorescence cameras with two-stage cooling based on the sensors from the same manufacturer (SONY):

Type	Resolution	Diagonal	Pixel	Light sensitivity	Speed
MAGUS CLM70	1.7MP	1.1"	9.0x9.0	8100mV	94FPS
CCD ICX825ALA	1.4MP	2/3"	6.45x6.45	2000mV	25FPS

CMOS camera features outperform those of a CCD camera, while the price of a CMOS camera is half that of a CCD camera.

Modern CMOS sensor-based cameras can be safely selected for laboratory microscopy, online streaming, and visual presentation in the educational process.

CCD sensor-based cameras still feature less digital noise. Such cameras are suitable for a research-grade microscope if speed is not a priority, there are no financial constraints, and no noise in the image is of crucial importance.

SHUTTER

This term originated from analog photography. The shutter on an analog camera is the curtain that allows light to reach the film and determines the exposure time. The shutter of a digital camera refers to the technique of reading the signal from the sensor's light-sensitive elements. The first type is a rolling shutter. The second one is a global shutter.

In a rolling shutter mode, the signal is read out line by line from top to bottom, i.e. each successive line shifts the exposure time from the previous one. The disadvantage is the distortion of fast-moving objects. In a global shutter mode, all pixels are read out at the same time. Global Shutter captures the entire image of moving subjects and shoots in low-light conditions. Its downsides compared to a rolling shutter: more impact on sensor warm-up, less dynamic range, more image noise, and more expensive.

A global shutter is recommended when purchasing a camera for fluorescent microscopy or for capturing fast-moving objects. In other cases, a rolling shutter will do.

INTERFACE

An interface is a connector used to transmit information from the camera. Microscope cameras are equipped with USB 2.0, USB 3.0, or HDMI interfaces, or more than one.

The USB 2.0 port is considered outdated, but most computers have it. Manufacturers continue to bring USB 2.0 cameras to market, but the model range is shrinking rather than growing.

The USB 3.0 port is a new interface whose data transfer speed and maximum current differs from the USB 2.0. Manufacturers implement new inventions in USB 3.0 cameras.

The HDMI port connects a camera directly to a monitor or TV without using a computer.

The first difference between HDMI and USB cameras is the control. USB cameras are controlled from a computer. Cameras output images on a computer screen, take photos, shoot video, measure, process images, and save files to a computer disk using a special program. The software is supplied with a camera. HDMI-cameras connect directly to a monitor, projector, or TV. The pictures are saved on an SD card. The cameras are controlled by a mouse that plugs into a special port on the camera. Mouse control excludes vibrations caused by direct user contact with the camera.

The second difference: HDMI connection speeds up the data transfer between the camera and monitor. In addition, the HDMI camera has a frame rate of 30-60 FPS. These two parameters together provide live video with no lag.

Optional USB 2.0 or USB 3.0 interfaces on HDMI cameras are used for photo and video processing applications, while optional Wi-Fi and WLAN interfaces transmit information over the network.

USB 2.0 cameras are the right choice when the goal is to save money, whether you are an amateur home movie maker or a student in a high school biology class. You should also choose a USB 2.0 camera if your classroom has outdated computers and there are no plans for upgrades. USB 3.0 cameras will not function with a USB 2.0 computer port because they require more amps or will lag during operation.

USB 3.0 cameras are recommended for professional laboratory work, research, and presentations at universities. It is not wise to save money in these cases. Let us compare two cameras. The 5MP USB 2.0 camera transmits information at 5-7 frames per second. This speed will not even allow you to comfortably adjust the focus of the microscope. The 6.3MP USB 3.0 camera transmits information at 59 frames per second. This speed will allow you to comfortably adjust the focus, find the desired area of the sample, and shoot live video.

HDMI cameras with high frame rates and high data rates are suitable for stereomicroscopic manipulations or online presentations. HDMI autofocus cameras with autofocus are a good choice for an inexperienced user and will help avoid hiccups in the presentation process.

CHOOSING A CAMERA

A camera is a device for specific purposes, and if you take a professional approach to choosing a camera, it is not as complicated as it seems at first glance. The information in this article will help in each individual case to professionally assess which parameter is not important and allows saving money, which remains a priority.

We invite our clients to visit our showrooms to ensure you make the right choice. You can bring your samples, compare the performance of several cameras, take our advice, and choose the perfect camera for your needs.

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