

The New Class in Fixed-Stage Microscopy



We make it visible.

# **Accessing Potentials**

Electrophysiological applications have very special requirements - regardless of whether cell cultures, living tissue sections, organs or entire animals are being investigated. These requirements also affect the microscopes that are used for these complex applications. In many discussions with researchers and scientists, optimized workflows were identified and have decisively determined the development of a new research stand at Carl Zeiss. The result is a fixed-stage microscope which is tailored to the requirements of its demanding users in every detail: with completely new flexibility in its configuration options; with freedom from disturbance - mechanical and electrical; with diverse contrasting options; with a new optical concept; with an exceptionally large amount of space for positioning micropipettes; and with the option of even being able to operate/adjust many microscope settings in the ongoing experiment without disturbance. The microscope: Axio Examiner from Carl Zeiss - the fixed-stage stand, which also meets the expanded high expectations in research.

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# Configuring Flexibility

Economy is one of the important requirements – also in exacting research dimensions. Anyone who is looking for an efficient solution in every aspect opts for Axio Examiner and configures this fixed-stage system to fit those exact requirements.

# Four upper bodys, two lower bodys: seven custom-tailored configurations

Axio Examiner offers you exceptional flexibility in configuring your fixed-stage microscope from four upper bodys, two lower bodys and a large number of different components and motorization options to ideally meet your individual requirements. It is both efficient and economical. Beginning with a pure transmitted-light stand to the most demanding systems for Laser Scanning Microscopy. With Axio Examiner.A1 a cost-effective entry-level solution is at your disposal, optically designed to the standard objective parfocal length of 45 mm, with options for a

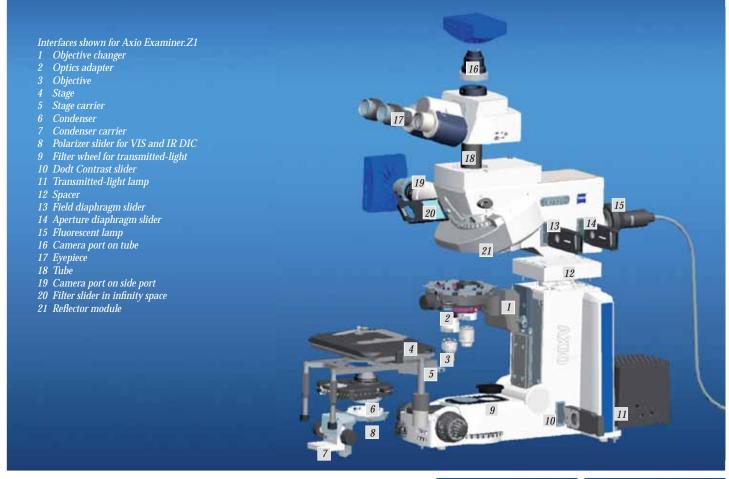
manual or motorized lower body. Axio Examiner.D1 and Axio Examiner. Z1 microscope versions have the innovative reflector turret of Axio Examiner line and a special optical concept. This new optical design allows improved accessibility to the specimen area for multi-electrode monitoring. When selecting a lower body for Axio Examiner.D1 you can again select between a manual or a motorized version. High-end Axio Examiner.Z1 microscope comprises a special upper body with side port coupling in the infinity space. The lower body of this version is always motorized.





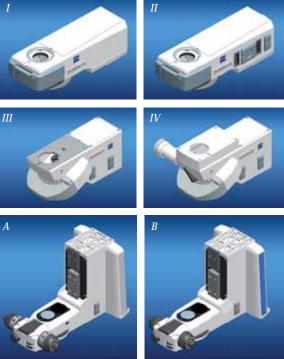


Configuration examples: 1) Axio Examiner.A1 with manual lower body 2) Axio Examiner.D1 with manual lower body and zoom tube 3) Axio Examiner.Z1 with motorized lower body



#### Flexibilized components

Axio Examiner has numerous interfaces, which allow very flexible microscope configuration to the individual requirements. Smart details which are particularly advantageous in multi-user environments. For example, the objective changer can be easily removed. According to requirements, one can select among a 1x holder, a 2x or a 4x turret. Designed as completely independent components, the stage and condenser carriers are also separately useable and completely removable. Interesting e.g. in intravital microscopy: for large specimens the condenser and stage carriers can be removed so the entire space can be used as specimen space. And for even larger specimens a spacer can be inserted at the interface between upper and lower bodies (see also p. 8).



- I. Upper body A1 transmitted-light
- II. Upper body A1 fluorescence
- III. Upper body D1 manual, configurable for transmitted-light or for transmitted-light and fluorescence
- IV. Upper body ZI can be optionally motorized, configurable for transmitted-light or for transmitted-light and fluorescence
- A. Lower body manual
- B. Lower body motorized

# **Designing Access**

Experimental workstations for upright electrophysiology typically involve complex set-ups for demanding micromanipulation, with many critical requirements. Among the important ones is the access to the specimen space. However, the possibility of appropriately adjusting the microscope during an ongoing experiment – without disturbing the sensitive experimental set-up – is also essential.





#### Expanded room in the specimen space

Never before has microscope design been conceived to ensure unimpeded work as it was for Axio Examiner. The result is extraordinary! The reflector turret is a special development. Positioned at a slant and integrated in an unusual mounting position, the optical axis passes through the front position. In this manner a large space is created for specimens and tools. This also applies to the 2x and 4x objective turrets. Each of them has also been constructed such that the optical axis passes through the front objective in its working position. This facilitates the positioning of pipettes decisively – not only from the sides, but also from the front. To change lenses, the objective is initially lifted more than 20 mm upwards and then rotated away to the rear at a safe distance from the pipettes.

#### Flexibly expand the free space

Axio Examiner's specimen space can be flexibly expanded according to your needs. Downwards by removing both the stage and the condenser as well as their carriers independently of each other. Upwards by using a 30 mm spacer. Depending on the instrument configuration, you can achieve a free working distance of more than 100 mm.

- Designed for optimum accessibility: the optical axis passes through the front positions of the reflector turret and the objective turret. The specimen space can be flexibly extended by removing the stage carrier and/or the condenser carrier. A 30 mm spacer between the upper and lower bodys creates additional space for intravital imaging.
- 2) Axio Examiner provides ample space for positioning patch pipettes.

#### The control elements made easy to operate

Microscope settings need to be changed in an ongoing experiment, which means that the relevant control elements must be easily accessible. For the architecture of Axio Examiner this means that all relevant control elements are positioned at the front. This applies to focus drive, stage drive and condenser drive, the light field diaphragm, the transmitted-light and reflected-light shutters, the switchover from visible to infrared light as well as to the intensity adjustment of transmitted-light illumination. And this also applies particularly to the important contrasting techniques. From simple oblique illumination DIC (Differential Interference Contrast) to the completely new integrated Dodt Gradient Contrast, all techniques can be operated from the front of the stand. This design has tangible advantages in practical applications, for all required settings can be easily adjusted without touching the sensitive set-up at all. In case you choose a motorized stand: all motorized functions are controllable via the keyrings, the docking station with touchscreen and the software AxioVision.





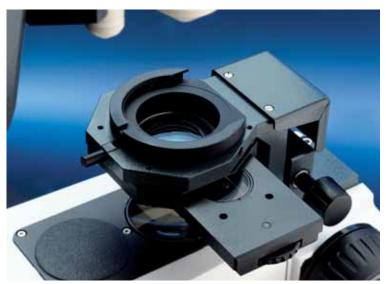
Whether one considers the manual entry-level version (above) or the motorized highend one (below), Axio Examiner can be optimally operated in every configuration.



The control elements for focus drive, stage drive and condenser drive, for the selection of visible or infrared light, for adjusting the light field diaphragm, the regulation of light intensity and for all contrast techniques in transmitted-light are positioned at the very front of the stand. On the motorized stand (right) all the motors can be controlled with the keyrings.

# **Enhancing Contrasts**

Tailor-made for demanding electrophysiological applications, a wide spectrum of contrasting options have been implemented in the fixed-stage Axio Examiner microscope. Optimally integrated, selectively applicable, and simply perfect for adaption to the respective specimen.



The polarizer slider has two positions, one for visible and one for infrared light. The contrast adjustment according to Sénarmont is performed by turning a knob to the right or left.



New standards even in simple contrasting: the segment diaphragm integrated in the condenser for oblique illumination can be turned through 360° and thus optimally adapted to the specimen structures orientation.

#### All important techniques, all integrated

In addition to resolution, contrast is the most important requirement for making the relevant structures in typical electrophysiological samples visible. Depending on the age of the experimental animals, tissue type, thickness of the sections, etc., the optical conditions can be very different. Consequently, it is extremely important to keep different contrasting options available on a microscope. It is ideal if the best technique can be selected for each specimen to ensure optimum visibility of the fine structures with minimal change to the work area. Now, having all techniques concurrently in one stand in Axio Examiner is a decisive developmental step in fixed-stage microscopy. Simple oblique illumination, Differential Interference Contrast (DIC) as well as the completely integrated Dodt Gradient Contrast can be used on Axio Examiner easily and without additional effort. You now have the flexibility of using the technique that provides the optimum visibility for the specific requirements of your specimen.

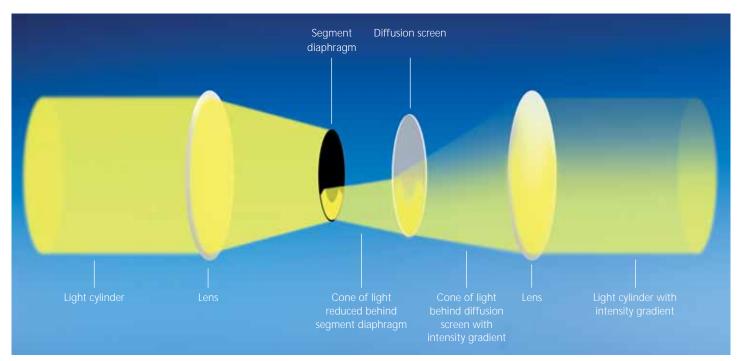
#### Oblique illumination

Even with the simplest contrasting technique, Axio Examiner sets new standards. Oblique illumination is already integrated in every condenser with the contrast turret and can thus be used without additional effort and extra costs. The segment diaphragm for oblique illumination on Axio Examiner is very easily rotatable through 360°. In this manner, the shadow projection can be adapted to the specimen's different structures, based on their orientation. In the process, depending on the specimen, the quality of the resulting image is already so convincing that oblique illumination offers a real alternative to the more complex DIC or Dodt Contrast.

#### **Dodt Gradient Contrast**

The patented gradient contrast according to Dodt offers several advantages in the examination of thick brain sections, in which the light is greatly diffused - both compared to contrast techniques with polarized light (DIC) and compared to simple oblique illuminations. As a result of the special segment diaphragm, the diffused light fraction in the sample is definitively reduced such that the maximum numerical aperture of the condenser can be completely exploited. Additionally, the combination of segment diaphragm with a diffusion screen ensures that no illumination fractions which are important for image formation are completely suppressed. In this manner, imaging artifacts are avoided. This means the following for your results: improved resolution, better contrast and thus improved structure recognition in deeper tissue layers. Since this technique does not require any polarizing

components, it is also more reasonably priced than Differential Interference Contrast. As the Dodt Contrast is only part of the transmitted-light illumination beam path it can easily be combined with fluorescence applications. Carl Zeiss has now developed an adaptation especially for Axio Examiner, which integrates this powerful technique directly in a special slider. The Dodt Contrast with 360° segment rotation can now for the first time ever be completely operated from the front of the stand – in an ideal position for adjustment without interfering with the electrodes and perfusion chambers. The 360° rotation allows its position to be optimized relative to the orientation of desired structures, something not possible with DIC without stage rotation, an option impractical with electrodes in position.



The principle of the Dodt Gradient Contrast: the arrangement of special segment diaphragm and diffusion screen reduces the diffuse light in the specimen, but does not result in complete suppression of illumination fractions. Consequently, imaging artifacts can be avoided and excellent results achieved on thick, extremely light-diffusing samples.

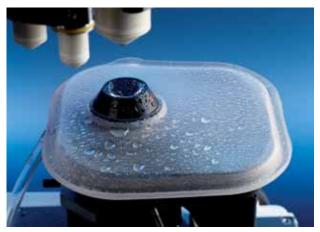
# **Enhancing Contrasts**



The Dodt Contrast slider is positioned in a conjugate plane to the aperture diaphragm...



... but the operation is performed at the very front of the stand.



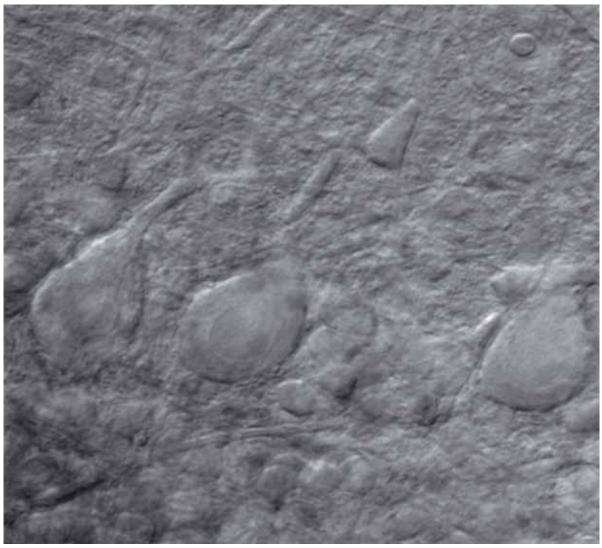
Aquastop: reliable protection of the optics and mechanism against overflowing salt solutions.

#### **Differential Interference Contrast (DIC)**

Naturally, the proven DIC technique, which provides unexcelled image results for thinner samples, remains an available contrasting technique of Axio Examiner. In order to make this technique as simple as possible to operate in electrophysiological experiments, it was implemented in Axio Examiner in the Sénarmont variant. The contrast setting is not performed on the DIC slider above the objective, but on the polarizer below the condenser. An intelligent solution to move the control elements out of the critical specimen space.

#### Oblique illumination, Dodt or DIC: VIS or IR

Infrared light is less strongly scattered than visible light in tissue and is thus particularly appropriate for high-contrast imaging in thick tissue imaging. All of these techniques are optionally available for visible or infrared light. This applies to oblique illumination and Dodt Contrast, in which changing from VIS to IR and back occurs simply by switching over the selection filter. And this naturally also applies to the Differential Interference Contrast (DIC). To be able to switch between visible and infrared light in this case, an impressive solution was developed for Axio Examiner. The polarizer slider under the condenser has two positions and can thus accommodate both the VIS and IR polarizers. In each case the desired polarizer must simply be shifted into the beam path. The analyzers in the reflector turret have been designed as Push&Click modules and are also correspondingly easy to change.



Purkinje cells in the cerebellum of a mouse, IR Dodt Gradient Contrast. F. Kirchhoff, Max-Planck-Institute for experimental medicine, Göttingen, Germany

#### Condensers from long-distance to water immersion

Unique in Axio Examiner: in addition to the 0.8 long working distance condenser and the 0.9 standard condenser, Carl Zeiss offers you a 1.2 W condenser. This water immersion condenser has a particularly high numerical aperture and offers the decisive advantage that the immersion has been exactly matched to the refractive index of the specimen. This is particularly important for multiphoton application with transmitted-light NDDs (Non-Descanned Detectors) in Laser Scanning Microscopy (see also p. 20).

#### Maximum safety

All condensers on Axio Examiner have stationary front optics and a nearly completely closed housing. Consequently, they are already very well protected against penetrating liquids. In addition a newly developed Aquastop protects their sensitive mechanism against overflowing salt solutions and other media. The special condenser Aquastop casing has a collection channel and a tube to drain the liquid – an effective protection against the high secondary costs which penetration of highly corrosive media can cause.

# Maximizing Visibility

The more difficult the specimens, the more demanding the experimental set-ups, the greater the demands on the optical performance of the fixed-stage system are. In Axio Examiner a large number of innovative details ensure maximum visibility at the limits of the achievable.

#### Newly developed optical concept

With objectives from 45 mm parfocal length, the proven ICS optics from Carl Zeiss offers excellent quality even in the entry-level Axio Examiner.A1 stands. In order to adapt the optical performance even better to the special requirements of fixed-stage microscopy, Carl Zeiss systematically modified its renowned optical concept for demanding transmitted-light and reflected-light applications with Axio Examiner.D1 and Axio Examiner.Z1 microscope versions. Part of the optical concept for these microscopes is an objective parfocal length of 75 mm. This innovation makes it possible to achieve consistent results over an even larger focus range. With excellent image quality, greater flexibility in z and improved accessibility in the specimen space. In the process, it is still possible to also use the standard objectives with 45 mm parfocal length in Axio Examiner. D1 and Axio Examiner. Z1 microscope versions. A 30 mm adapter which adapts the 45 mm objectives to the new optical system was developed for this purpose. The following applies to all microscopes in Axio Examiner line: due to a specially developed illumination concept with parallel beams, the illumination in transmitted-light remains optimum - regardless of how large the distance between condenser and light field diaphragm.

#### Objectively superior

Experiments involving manipulation of the specimen being observed place extreme demands on the objective geometry. For a high numerical aperture, a long working distance is required in order to be able to manipulate capillary pipettes under the objective. This is an extremely challenging demand which must be resolved by an optimum compromise. In order to be able to still capture the last photons, Carl Zeiss has redesigned the water immersion objectives with regard to correction and numerical aperture to meet the ideal balance of affordability versus optical performance requirements.

#### W N-ACHROPLAN

These cost-effective water immersion objectives have been especially designed for electrophysiological applications in physiological sodium chloride solutions, which approximate the cell milieu more closely than distilled water. They are ideal for many standard applications and attractive due to their particularly favorable price-performance ratio.

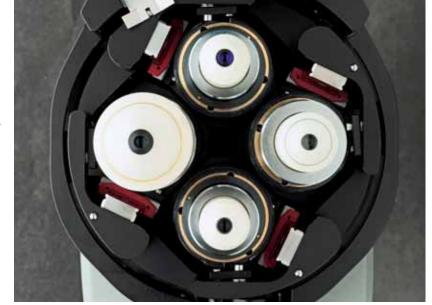
The new water immersion objectives W N-ACHROPLAN convince with an excellent price-performance ratio.



W Plan-APOCHROMAT: the high-end water immersion objectives for highest requirements.



High-tech in a very small space: the 4x objective turret. The optical axis passes through the front position, objectives that are not in use swivel to the rear out of the specimen area.



#### W Plan-APOCHROMAT

This objective series with the most sophisticated chromatic and spherical correction offers outstanding performance. In addition to apochromatic correction in the visible range, it has been corrected for the near infrared range. Consequently, these are the objectives of choice particularly for multiphoton applications. With their excellent correction and their extraordinarily high numerical apertures, these objectives are appropriate not only for confocal applications, but all applications concerned with capturing as many photons as possible from different wavelength ranges, e.g. for all Multichannel Fluorescence applications.

# Functionality in the smallest space: the objective changer

Completely redesigned to provide maximum space for electrophysiological experiments, the objective changers are available in different versions. You can use them according to requirements and simply exchange them as needed. As alternatives there are 1x holders as well as 2x and 4x turrets, which are advantageous in multi-user environments with many different applications. The objectives are shifted upwards with a perpendicular free travel range of 22 mm. With the 2x and 4x turrets the objectives then swivel toward the rear out of the working space in the smallest possible radius.

#### Variability in magnification change

Freedom from disturbance during an experiment was one of the main objectives in the development of Axio Examiner. This also included creating possibilities to change the magnification independently of the objective and with adequate distance from the specimen. Altogether there are three solutions available to change the magnification independently of the objective, all of which include "soft stop" detents to minimize mechanical distruption to the patch:

- Push&Click optovar modules in the reflector turret of Axio Examiner. These apply both to the eyepiece image and the camera image with factors of 1.25x, 1.6x and 2.5x
- A second option is the 4 position magnification changer turret. It is optically adjusted for parfocal magnification change and with the available factors of 0.5x, 1x, 1.6x, 2.5x, 4x and 5x it provides sufficient alternatives for varying the magnification across a wide range. In the process the position of the objective remains unchanged.
- Newly developed for Axio Examiner and in a league of its own: the zoom tube provides you with infinitely variable subsequent magnification options for two camera ports with identical image positions. It zooms from 0.5x up to 4x, i.e. by a factor of 8, without changing objectives. The upper port is adjustable in x, y and z so that the images on both ports can be exactly aligned to each other. The light can be distributed to the cameras via a 1x slider for the Push&Click modules. This allows an IR-DIC channel and a fluorescence one or two different fluorescence channels to be simultaneously imaged.

# **Motorizing Performance**

Motorization is a critical topic in electrophysiology because electrical disturbances must be absolutely avoided. However, for complex experiments some motorized functions cannot be dispensed with. How they are implemented is decisive.

#### Flexible motorization options

The motorized lower body of Axio Examiner provides an integrated high-precision z-drive, as well as options for a motorized transmitted-light shutter and as a special feature, the option of a motorized condenser focus. The lower body can be freely combined with all upper bodys. The motorizable upper body Z1 can optionally be equipped with a motorized reflector turret and a fast shutter.

#### Disturbances eliminated

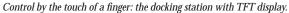
All motorized components have been designed to function with extremely low vibration, thus mechanical disturbances have been eliminated to the greatest possible extent. To also avoid electrical disturbance effects, all motors automatically switch off as soon as they have reached their target position. In addition, active grounding can be implemented by pushing a button. This ensures that any potentials which might still be present can completely leak off. Electrical disturbances are thus reliably eliminated

#### The motorized z-drive

High resolution and excellent reproducibility – Axio Examiner's motorized z-drive is unexcelled in its performance. It allows extremely small step widths of 25 nm and a reproducibility of ± 50 nm. Such a precise motor is particularly important when defined positions have to be repeatedly reached. It is therefore custom-tailored to demanding Z-stack imaging including Laser Scanning Microscopy.

#### The motorized condenser focus

The motorized condenser focus is unique in Axio Examiner and specially developed for the highest performance requirements of the fixed-stage microscope. As a result of its synchronous focusing, it automatically ensures the optimum illumination conditions for every focusing position of the objective. Additionally, it can be used in combination with corresponding control curves for the correction of different immersion media. This is a measurable gain in efficiency particularly for transmitted-light applications with NDDs in Laser Scanning Microscopy – a special application in which the condenser is not used for illumination





Functional motorization without risk: all motors automatically switch off and in addition, they can be actively grounded so any potantials which might still be present can leak off.

but to collect photons. Exactly in this case is it particularly important to avoid mismatches and to capture as many photons and as much information as possible.

#### The Remote Control option

Axio Examiner's motorized functions are easily reached and operated on the stand via the keyrings on the focus knobs. Or if the microscope cannot be touched, the same results can be obtained by using the optional docking station's touchscreen. This consequent merging of control and monitoring in one compact unit is unique to Carl Zeiss products. Many processes can be operated via the extremely clear menu guide, which is separate from the PC. The activation of individual functions or complete preconfigured settings of operational procedures is achieved by nothing more than a brief touch with a finger. A further option for Remote Control is offered by AxioVision Software, which supports all of Axio Examiner's motorized functions completely and can be integrated into the experimental workflow. In this manner, complex multichannel experiments can also be conducted over time or as Z-stack while simultaneously experiencing optimum specimen protection. The Remote Control option is relevant wherever interventions in complex experimental set-ups must be avoided.



Remote Control: all motorized functions can be operated via the docking station and the AxioVision Software.

# Redimensioning Sensitivity

Axio Examiner is the fixed-stage platform for the Laser Scanning Systems LSM 710 and LSM 710 NLO. With these innovations Carl Zeiss again sets new standards in confocal microscopy. An entire bundle of epic-making technical innovations gives research completely new impulses in multiple fluorescent-labelled, also live, specimens.





LSM 710 and LSM 710 NLO: unrivaled sensitivity in Laser Scanning Microscopy.

#### LSM 710 for highest sensitivity and flexibility

The newly developed LSM 710 laser scanning microscope achieves very high sensitivity with low dark noise and optimal suppression of the excitation light. The use of extremely different fluorescent dyes and the performance of multimodal experiments are just as easily possible as the imaging of preparations on strongly reflecting surfaces.

The new illumination and detection design of the system provides absolute freedom in selection. The basis for this is a filter free spectral detection unit, whose adjustment is infinitely variable across the entire wavelength range. Innovative analysis techniques such as integrated image correlation spectroscopy allow extraction of quantitative information at molecular concentrations and dynamics directly from the confocal images recorded.

The PTC laser concept of the LSM 710 is virtually revolutionary. It is completely independent of a laser module and allows a flexible combination as well as easy upgrade with different lasers from near UV into the infrared range. The innovative TwinGate main dichroic concept supports up to 50 laser line combinations. It impresses with individually exchangeable filters and convinces with its unexcelled suppression of the excitation laser light with brilliant, high-contrast images. The new QUASAR Detector in 2-, 3- or 34-channel version is more sensitive and more flexible than all previous spectral detector technologies and provides up to 3 nm spectral resolution. A spectral recycling loop increases the detection efficiency of the spectrally split emission light up to nearly 100 percent.

Automatic software tools for system calibration and control of the central performance parameters ensure that the LSM 710 is always operating in its optimum performance range. An additional system highlight is the Smart Setup function of the ZEN 2008 software, a tool for dyecentered device configuration. As a result, achieving an image becomes more rapid and simpler than ever before.

The LSM 710 NLO at Axio Examiner with up to 5 NDDs in both reflected-light and transmitted-light as well as the GaAsP detector integrated in the objective carrier



# **Redimensioning Sensitivity**

# LSM 710 NLO – with Axio Examiner unique for intravital multiphoton microscopy

Axio Examiner was jointly developed with the LSM 710 NLO and designed especially for demanding multiphoton imaging. The basis for this technique for three-dimensional imaging of complex biological systems are extremely sensitive Non-Descanned Detectors (NDDs). In Axio Examiner.Z1 these NDDs can be postioned close to the object on the side port coupling in the infinity space in reflected-light and additionally in transmitted-light on a coupler directly under the condenser. In addition to their new electronics, these NDDs are characterized by their uniquely high light conductance to the detectors and within the individual modules. A special variant, the GaAsP NDD detector offers an additional twofold higher sensitivity. The module combines objective holder and detector and conducts the emission signal immediately behind the objective onto an extremely light-sensitive detector.

Up to five standard NDDs can be used and simultaneously operated both in reflected-light and transmitted-light. As is the case with NDD equipment, the filter configuration can also be adapted by the customer. Additional advantages are the optimized transmission of the scanning module for the entire wavelength range up to 1100 nm and the free combinability of the

multiphoton laser with other laser lines, including lasers with emission in near UV. In this context, individually operable, motorized collimators also ensure the superimposition of excitation levels for combined use of different wavelengths. The complete integration of the system control in the LSM software ZEN 2008 ensures efficient working and maximum specimen protection. With the LSM 710 NLO Carl Zeiss has made an innovative system for multiphoton microscopy with femtosecond technology available to basic and biomedical research. The particular strengths of the LSM 710 NLO lies in the recording of high-resolution images of fluorescing structures in living animal and in thick tissue specimens. Similarly, long-term observations of developmental processes as well as functional imaging in connection with locally defined photomanipulation of fluorescing structures can be optimally performed. In the process the high signal-to-noise ratio ensures brilliant imaging of cellular and subcellular details also in deep tissue layers.

#### The 1.2 water immersion condenser

Especially for multiphoton applications in Laser Scanning Microscopy Carl Zeiss offers a 1.2 water immersion condenser and thus a unique performance characteristic, which is particularly advantageous when the condenser is used as imaging optics instead of illumination optics. The idea is that in multiphoton applications fluorescence signals are collected by the condenser. If the condenser has been calibrated for the same medium in which the specimen is located, the maximum photon occurrence will be captured at exactly assigned levels and mismatch will be avoided. In laser scanning applications, this is a decisive criterion for the efficiency (see also Condensers from long-distance to water immersion p. 13).

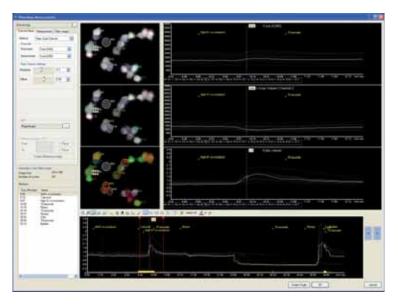


The highly sensitive GaAsP detector, which is integrated directly into the objective carrier provides up to twofold higher sensitivity.

- 1 Dichroic
- 2 Collection optics
- 3 Mirror
- 4 Focussing optics
- 5 GaAsP detector

# Integrating Software Highlights

AxioVision is the microscopy software for practical solutions in Digital Imaging. Developed in close cooperation with users, even in the basic version this software provides a great deal of functionality. AxioVision can be modularly expanded at any time for extremely different requirements in electrophysiology, e.g. by adding the modules Physiology and Dual Camera.



#### Physiology

Developed for simple yet powerful evaluation of the changes in fluorescence intensities in cells, Physiology augments your time series recordings with quantitative measurement functions. Intensity changes can be measured online or offline either directly or ratiometrically. Physiology offers a great number of functions for this:

- Recording of time series in streaming technique with maximum velocity.
- Ratio measurement of ion concentration such as calcium or pH
- Use of single channel dyes (such as Fluo-4) or two channel dyes (such as Fura-2 or Indo-1)
- FRET measurements according to established or one's own formulas
- Free drawing in up to 100 Regions of Interest (ROI)

- Online display of the measured values as diagrams allows exact control of the experimental course
- Influencing recording process by setting markers
- Flexible TTL control of external components
- Cutting out interesting image sequences with the cutting editor
- Conversion into the AxioVision ZVI image format
- Offline evaluation of any arbitrary ZVI time series images
- Display of ratio values as color scale with dimensioning

Prerequisite for the use of Physiology are the modules Fast Image Acquisition, Time Lapse, Multichannel Fluorescence as well as an appropriate light source (e. g. High Speed Colibri LED or Sutter Lambda DG4).

#### **Dual Camera**

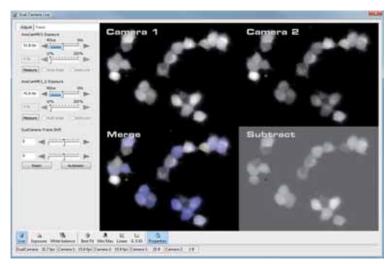
Whenever the speed requirements do not allow switching components (e.g. filter wheels), Dual Camera allows the perfect synchronous operation of two structurally identical cameras by means of patented synchronization technology. In this manner, on the one hand, the recording speed can be doubled, and, on the other hand, the synchronous recording of two channels is guaranteed to have absolute temporal synchronization. In this manner, time series of even extremely rapid processes without image offset can be recorded in two channels and as Z-stacks. Dual Camera also extends the module Physiology by functions for Emission Ratio Imaging as well as FRET imaging. Dual Camera requires at least the modules Multichannel, Time Lapse and Fast Image Acquisition.

# **Opening Dual Camera Options**

The requirements for high-performance camera technique are as different as the applications in electrophysiology and neurobiology. Correspondingly diverse is the selection of cameras and camera interfaces on new Axio Examiner fixed-stage microscope. The spectrum extends from the use of high-resolution color cameras over extremely fast fluorescence cameras up to the use of two cameras simultaneously.



The sideport slider in the infinity space: in combination with a phototube one of the available options for a dual camera system.



Complete control with the AxioVision module Dual Camera: live images from 2 simultaneously recording cameras, merged image and subtracted image.

#### Versatile camera interfaces

Flexibility in camera interfaces: Axio Examiner offers variable options for the use of digital microscope cameras.

- Different phototubes are available as standards.
- Side port coupling in infinity space provides an additional option for camera interface in the stand variant
   Axio Examiner.Z1. In this manner, and combined with
   a phototube a dual camera system can easily be implemented.
- A special dual camera adapter for the phototube is available for the realization of a high-performance dual camera system. Emission filter and color splitter can be interchanged by the customer such that you can adapt the equipment exactly to your requirements.
- The zoom tube provides two camera ports with identical image positions. This is ideal for combining images made with different techniques, such as DIC and fluorescence, for quantitative analyses.



Axio Examiner in two of the three possible dual camera configurations.

#### The blue microscope cameras

Carl Zeiss offers a broad spectrum of digital cameras in different performance classes. The monochrome cameras are ideal for fluorescence specimens and provide optimum resolution and highest sensitivity or highest recording speed for recording very rapid processes. The color cameras stand for the best color reproduction and highest resolution up to 12 megapixel without the compromises of color interpolation. All cameras are Peltier-cooled and provide for rapid synchronization with other components such as shutter or light source. The rapid live image with automatic exposure time adjustment and the complete integration in the Carl Zeiss systems approach characterizes all the cameras in the AxioCam family.



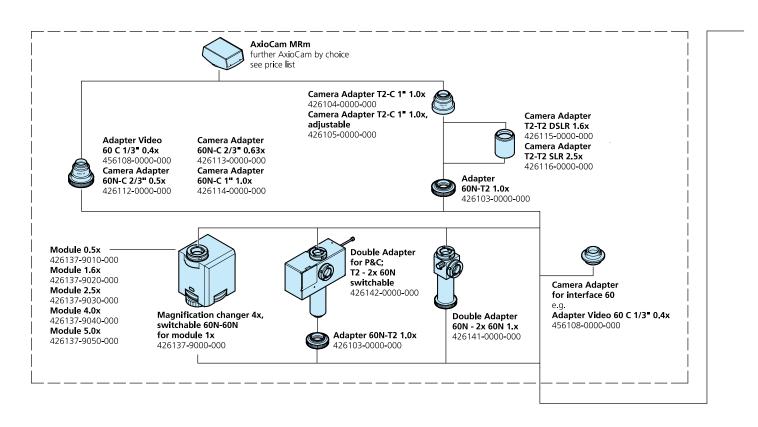
 $Simply\ convenient:\ the\ use\ of\ the\ Dual\ Camera\ option\ in\ the\ zoom\ tube.$ 

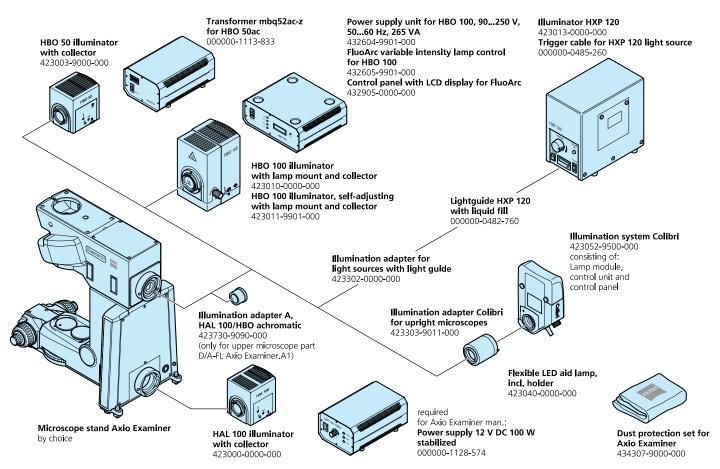
# Recognizing Advantages

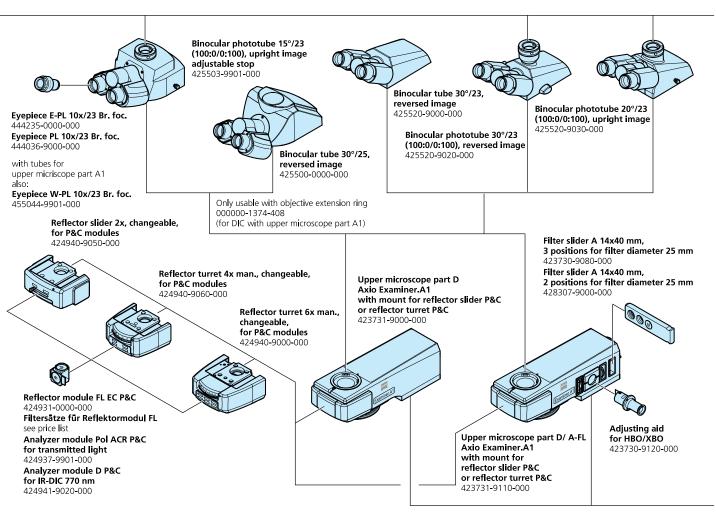
- Flexible stand concept with two lower bodies, four upper bodies and numerous interfaces
- Excellent access to specimen space from the sides and from the front
- Special reflector turret with optical axis passing through the front position
- Specimen space, depending on the configuration, extendable to more than 100 mm
- Stage carrier and condenser carrier independently removable
- All relevant control elements can be operated from the stand front: focusing drive, stage drive, condenser drive, light field diaphragm, Differential Interference Contrast (DIC), Dodt Gradient Contrast, oblique illumination, intensity setting of transmitted-light, VIS-IR switch, transmitted-light shutter, reflected-light shutter, reflector turret, etc.
- Optical concept optimized for fixed-stage conditions with objective parfocal length of 75 mm (all 45 mm objectives can be used with the optics adapter)
- Interchangeable objective turret with 1, 2, or 4 positions, optionally coded (2x and 4x)
- Free objective lift of 22 mm, focusing distance 15 mm
- Cost-effective W N-ACHROPLAN objective series for all standard applications
- High-end W Plan-APOCHROMAT objective series for highest requirements in Multichannel Fluorescence imaging, in Laser Scanning and in Multiphoton Microscopy
- Fluorescence beam path has been completely recalculated
- All relevant transmitted-light contrast techniques for VIS and IR can be integrated together in one stand: oblique illumination, Dodt Gradient Contrast, Differential Inference Contrast (DIC)
- Oblique illumination, which is freely rotatable through 360°, is already integrated in all condensers with modulator disk at no additional cost.
- Dodt Gradient Contrast available as integrated solution for the very first time, freely rotatable through 360°
- Simple switching between DIC and IR-DIC, contrast adjustment according to Sénarmont
- Condensers 0.8, 0.9 and 1.2 W

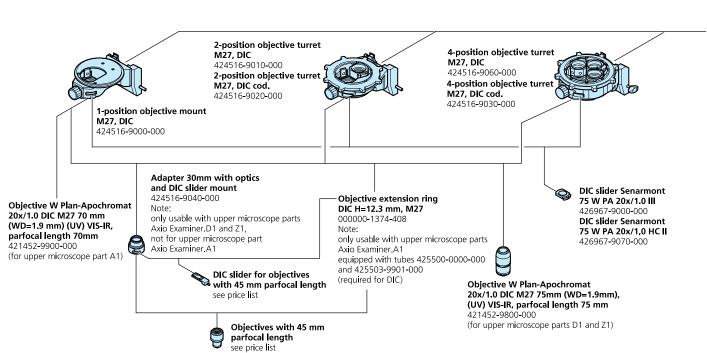
- Precisely fitting Aquastop for all condensers with modulator disk to protect against overflowing liquids
- Dual Camera options via side port coupling, zoom tube, Dual Camera adapter
- Magnification change options via optovar modules, magnification changer, zoom tube.
- Motorization options for z-drive, transmitted-light shutter, condenser drive, reflector turret, reflectedlight shutter.
- Optional motorization of the condenser drive for synchronous focusing with the objective, calibration curves adaptable for different immersion media. Consequently, continuous, optimum illumination conditions and substantial plus in efficiency for transmitted-light NDD (Non-Descanned Detection)
- All motors disconnect from the power supply after reaching their target position
- Motors switch off for additional active grounding to allow possibly present potentials to completely leak off.
- Operation of the motor functions via the keyrings on the stand, via the docking station with touchscreen, via the AxioVision software
- New AxioVision modules Physiology and Dual Camera for quantitative analyses in Live Cell Imaging.
- Motorized stand variant for LSM 710 and LSM 710 NLO
- Object-near coupling for NDDs (Non-Descanned Detectors) in reflected-light and transmitted-light
- Water immersion condenser 1.2 W for highest efficiency, not only in illumination, but particularly for non-descanned detection in transmitted-light
- Special objective carrier with integrated highly sensitive GaAsP detector for up to 2x higher sensitivity

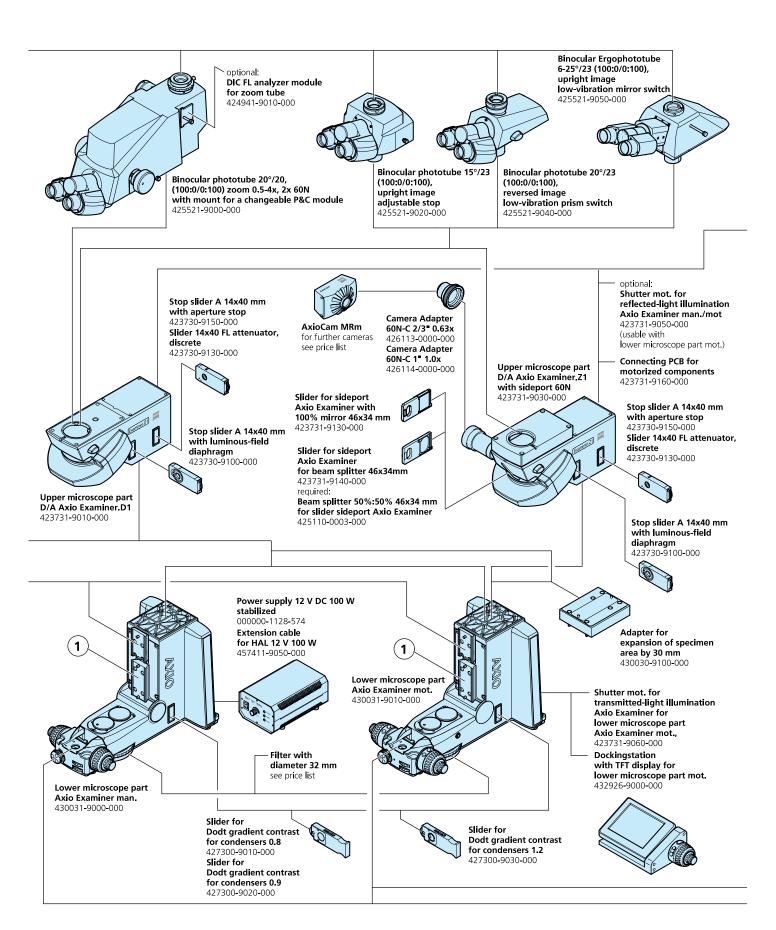




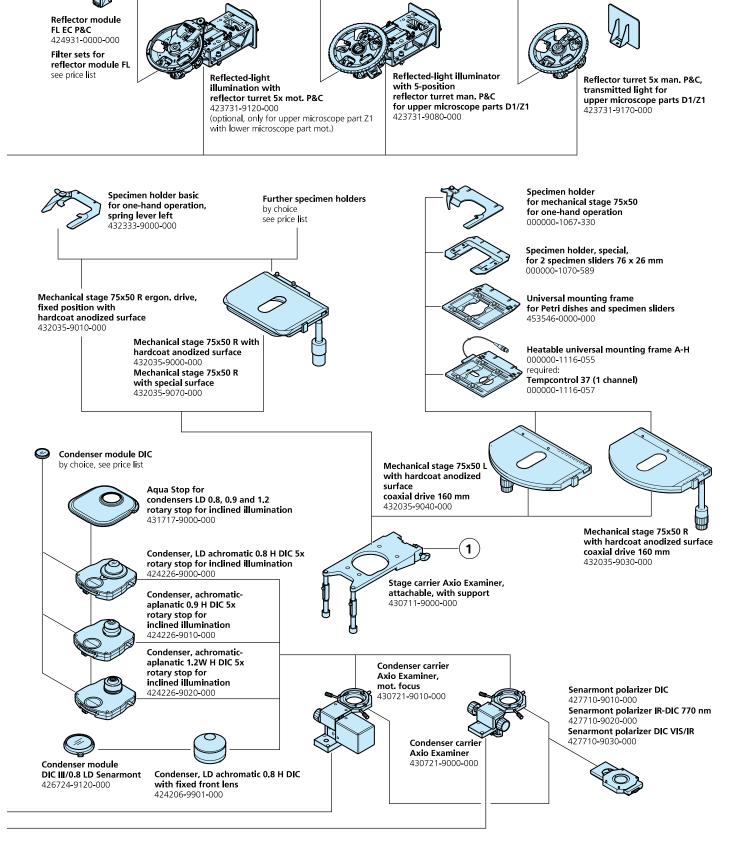




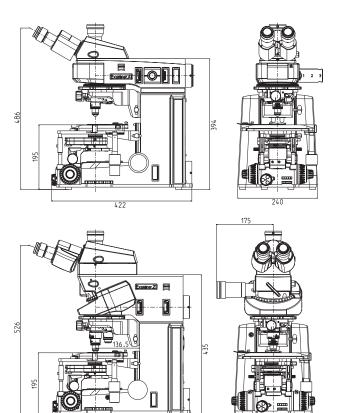


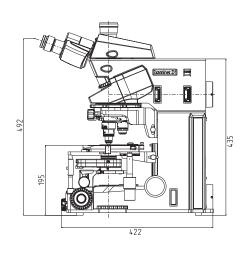


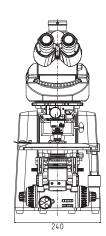
# System Overview



# **Reviewed Facts**







Axio Examiner.A1 (top left) Axio Examiner.D1 (top right) Axio Examiner.Z1 (below)

Stand	Lower body	Upper body	Objective
Axio Examiner.A1	manual	manual, Axio Examiner.A1 only for transmitted- light (Fluorescence not retrofittable!)	45 mm or 70 mm
Axio Examiner.A1	manual	manual, Axio Examiner.A1 for transmitted-light and fluorescence	45 mm or 70 mm
Axio Examiner.A1	motorized, z-drive motorized, transmitted-light shutter optional motorized, condenser drive optional motorized	manual, Axio Examiner.A1 only for transmitted- light (Fluorescence not retrofittable!)	45 mm or 70 mm
Axio Examiner.A1	motorized, z-drive motorized, transmitted-light shutter optional motorized, condenser drive optional motorized	manual, Axio Examiner.A1 for transmitted-light and fluorescence	45 mm or 70 mm
Axio Examiner.D1	manual	manual, Axio Examiner.D1, optional for transmitted-light or transmitted-light and fluorescence	45 mm + 30 mm optics adapter and 75 mm
Axio Examiner.D1	motorized, z-drive motorized, transmitted-light shutter optional motorized, condenser drive optional motorized	manual, Axio Examiner.D1, optional for transmitted-light or transmitted-light and fluorescence	45 mm + 30 mm optics adapter and 75 mm
Axio Examiner.Z1	motorized, z-drive motorized, transmitted-light shutter optional motorized, condenser drive optional motorized	motorizable, Axio Examiner.Z1, with side port, optional for transmitted-light or transmitted-light and fluorescence, reflector turret optional coded or motorized, reflected-light shutter optional motorized	45 mm + 30 mm optics adapter and 75 mm



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