

IC²S-Objectives for Biomedical Applications



**Confocal Laser Scanning Microscopy
Multiphoton Laser Scanning Microscopy
Fluorescence Correlation Spectroscopy**



We make it visible.

Whether your research subject is in cell biology, developmental biology, neurobiology or physiology, Carl Zeiss offers you a wide range of objectives to fit the special properties of your specimen and the LSM.

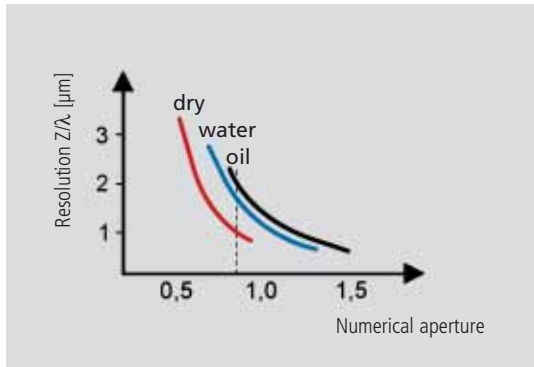
This brochure presents a selection of the best ZEISS objectives for LSM use, sorted by type of usage and correction properties. It is intended as a help for choosing the right objectives for your LSM, in order to guarantee the best possible image results.

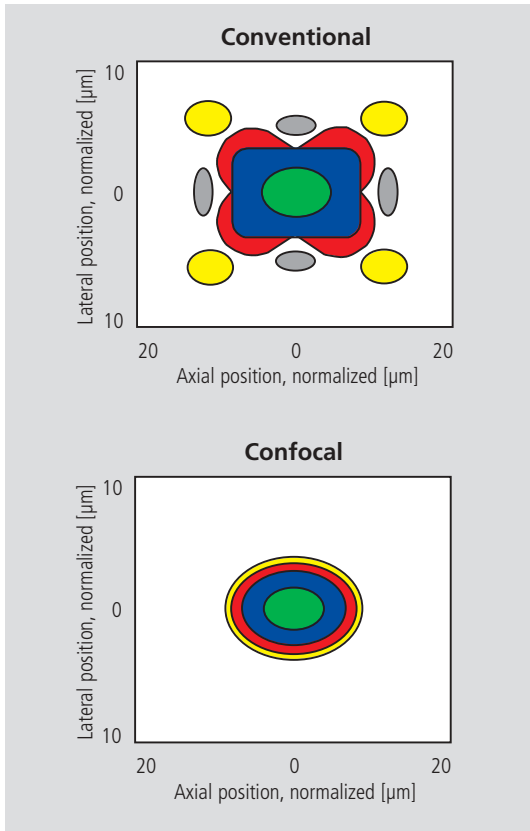


Why Special Objectives for Confocal Microscopy?

In confocal microscopy, the requirements for objective design and quality are much higher than in conventional light microscopy. Due to the ability to obtain optical sections in Z and to collect high resolution data of one point in the specimen at various wavelengths simultaneously, confocal microscope objectives need a perfect correction of longitudinal chromatic and spherical errors over the full wavelength range.

Half width of axial resolution in relationship to the numerical aperture of objective types. Relatively seen, the axial resolution at a given numerical aperture is highest in dry objectives, whereas the maximal possible resolution is achieved in oil immersion objectives. A good compromise of both properties is available in water immersion objectives.





Maximum quality of point spread functions (PSF) in conventional (wide field) microscopy and in confocal microscopy. The almost ideal PSF in confocal microscopy is only available, of course, if perfectly corrected ("diffraction limited") objectives are used.

Recommendation of objective classes and biological specimen properties.

| Specimen | | Matching objectives regarding coverslip, media correction and working distances | | | |
|--|-----------------------|---|------------------------------------|---|-----------------------------------|
| Subject | Optical property | Dry | Water immersion | Oil immersion | Water dipping |
| Cell Biology Microbiology | covered, thin | EC Plan-Neofluar,, Fluar, Plan-Neofluar LD | C-Apochromat, LCI Plan-Neofluar | Plan-Apochromat, EC Plan-Neofluar, Fluar | |
| Developmental Biology | covered, in dish etc. | Plan-Neofluar LD 40x & others | C-Apochromat | LCI Plan-Neofluar 25x & others | |
| Zoology, Botany Neurobiology | covered, thick | Plan-Apochromat 20x & others | C-Apochromat | Achroplan 50x & others | |
| Physiology, Micromanipulation | uncovered | Fluar, EC Plan-Neofluar 2.5–10x range | | | W Achroplan, W Plan-Apochromat |

Confocal Specialists

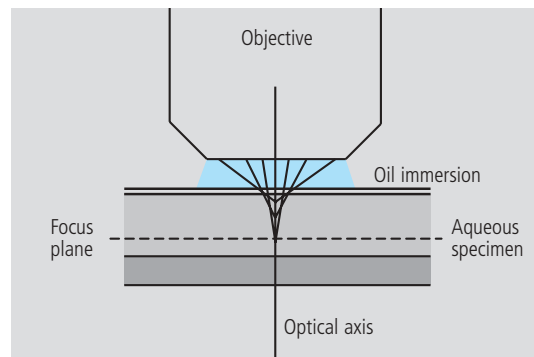
Cell Biology
Microbiology

Developmental
Biology

Zoology, Botany
Neurobiology

The C-Apochromat objectives (C for confocal) have been developed to match the requirements of diffraction limited optics. The C-Apochromat series water immersion objectives are corrected for an extended range of six wavelengths instead of three or four in conventional Apochromats, starting at 360 nm up to 1000 nm (e.g. for the C-Apochromat 63x UV-VIS-IR), and therefore they work especially well for extended Z-scans in biological tissue and for spectral imaging with the META detector. Magnifications of C-Apochromats range from 10x overview to 40x and 63x with adjustable coverslip and temperature correction.

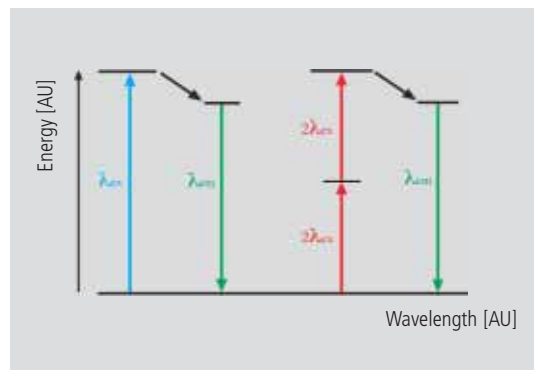
For multiphoton applications and time lapse studies in living specimens, the LD C-Apochromat and C Achroplan NIR are available. These objectives with very high transmission in the near infrared offer an outstanding working distance even for deepest penetration of tissues.



Spherical aberration resulting from different refractive indices of sample and immersion media. This aberration is the greater, the deeper the focus goes into the sample.

| Objective Type / Magnification / n.A. | W.D. (mm) | Thread | Order Number |
|--|-----------|--------|-----------------|
| C-Apochromat 10x/0,45 W | 1,8 | M27 | 421747-9900-000 |
| C Achroplan 40x/0,80 W NIR | 1,7 | W 0,8" | 000000-1080-378 |
| LD C-Apochromat 40x/1,1 W Korr UV-VIS-IR | 0,62 | M27 | 421867-9970-000 |
| C-Apochromat 40x/1,2 W Korr UV-VIS-IR | 0,28 | M27 | 421767-9970-000 |
| C-Apochromat 63x/1,2 W Korr UV-VIS-IR | 0,28 | M27 | 421787-9970-000 |

Fluorescence excitation and resulting emission. Left: excitation with 1 photon; right: cumulated effect of 2 photons at the doubled wavelength.



Top Grade Optics

Developmental
Biology

Zoology, Botany
Neurobiology

Using water or glycerol immersion is the closest match to the refractive index of biological tissue and popular embedding media, and reduces spherical aberration even at very deep focus settings. That's why Carl Zeiss offers the LCI Plan-Apochromat objectives with outstanding correction and working distance for live cell imaging applications.

| Objective Type / Magnification / n.A. | W.D. (mm) | Thread | Order number |
|--|-----------|--------|-----------------|
| LD LCI Plan-Apochromat 25x/0,8 Imm Korr DIC | 0,60 | M27 | 420852-9870-000 |
| LCI Plan-Apochromat ¹ 150x/1,35 Glyc Korr DIC | 0,18 | M27 | 420792-9970-000 |

¹ in prep for summer 2007

Resolution Experts

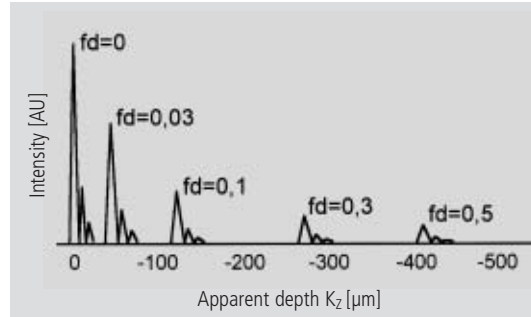
Cell Biology,
Microbiology

Zoology, Botany
Neurobiology

The range of LCI Plan-Apochromats can be complemented for conventional thin specimens with multiple fluorescence preferentially in the VIS range (420 – 670 nm) by the Plan-Apochromat dry and oil immersion objectives of highest numerical apertures and perfect image flatness. Our legendary Plan-Apochromat 63x/1.40 Oil and Plan-Apochromat 20x/0.80 offer exceptional properties

| Objective Type / Magnification / n.A. | W.D. (mm) | Thread | Order Number |
|---|-----------|--------|-----------------|
| Plan-Apochromat 10x/0,45 | 2,0 | M27 | 420640-9900-000 |
| Plan-Apochromat 20x/0,80 | 0,55 | M27 | 420650-9901-000 |
| Plan-Apochromat 40x/1,30 Oil DIC VIS-IR | 0,21 | M27 | 420762-9800-000 |
| Plan-Apochromat ² 63x/1,40 Oil DIC | 0,19 | M27 | 420782-9900-000 |
| Plan-Apochromat 100x/1,40 Oil DIC | 0,17 | M27 | 420792-9900-000 |
| α Plan-Apochromat 100x/1,46 Oil Iris VIS-IR | 0,10 | M27 | 420796-9800-000 |

² heat insulated „i“ version available



Loss of axial resolution resulting from focussing an oil immersion objective into an aqueous sample (FD=focus distance). The deeper the focus goes into the sample, the smaller becomes the intensity maximum.



regarding fluorescence efficiency and free working distance. The new Plan-Apochromat 40x/1,30 Oil and Plan-Apochromat 100x/1,46 Oil even offer an improved performance up to the IR range.

Universal Solutions

Cell Biology
Microbiology

Developmental
Biology

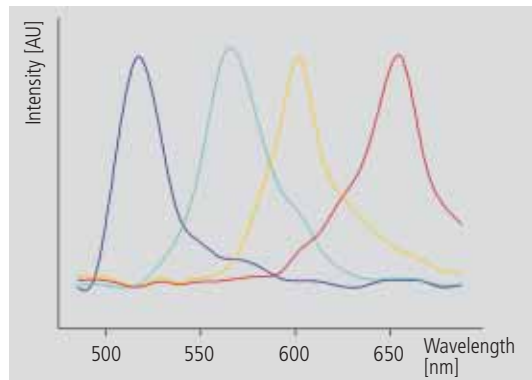
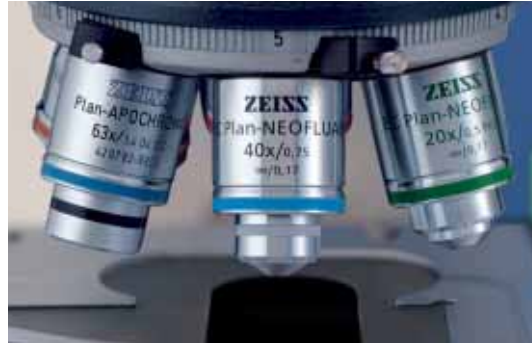
In addition to objectives dedicated to high end confocal microscopy, a universal objective solution is often required to match the properties of various specimens and protocols. The EC Plan-Neofluar objectives are perfectly designed for general fluorescence microscopy. The universal EC Plan-Neofluar dry and oil immersion objectives are corrected for an extended range from 435 nm up to 670 nm, sufficient even for 4-color fluorescence, with a correction level matching or even surpassing yesterday's finite system Apochromats.

Transmission even starts in the UV range at 340 nm. As a truly versatile solution, various types are available including the LCI* Plan-Neofluar 25 x and 63 x Imm., which can be used with water and glycerol to match any refractive media index.

| Objective Type / Magnification / n.A. | W.D. (mm) | Thread | Order Number |
|--|-----------|--------|-----------------|
| EC Plan-Neofluar 20x/0,50 | 2,0 | M27 | 420350-9900-000 |
| LD Plan-Neofluar 40x/0,60 Korr | 2,9 | M27 | 421360-9970-000 |
| EC Plan-Neofluar 40x/0,75 | 0,71 | M27 | 420360-9900-000 |
| EC Plan-Neofluar 40x/1,30 Oil DIC | 0,21 | M27 | 420462-9900-000 |
| Plan-Neofluar Imm 16x/0,50 Imm | 0,17 | W 0,8" | 440530-0000-000 |
| LCI Plan-Neofluar ² 25x/0,8 Imm Korr DIC | 0,21 | M27 | 420852-9972-000 |
| LCI Plan-Neofluar ² 63x/1,30 Imm Korr DIC | 0,17 | M27 | 420882-9970-000 |

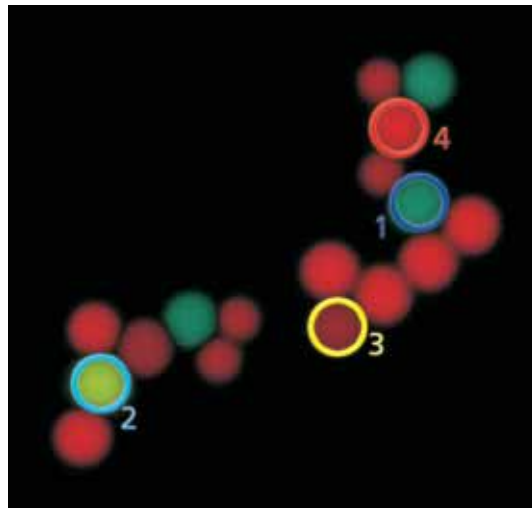
² heat insulated „i“ version available

* Live Cell Imaging



Spectral signature of a fluorescence emission. Four different fluorochromes alone can be matched into the VIS range of 450 – 670 nm.

Lambda- (wavelength-) coded view of multi-color fluorescence beads.



Transmission Experts

Cell Biology
Microbiology

Developmental
Biology

To offer ideal prerequisites for physiological measurements, photon collecting and TIRF microscopy, the Fluar series of dry and oil immersion objectives are available as the fluorescence specialists in the ZEISS objective range.



| Objective Type / Magnification / n.A. | W.D. (mm) | Thread | Order Number |
|---------------------------------------|-----------|--------|-----------------|
| Fluar 2,5x/0,12 | 6,3 | M27 | 420120-9900-000 |
| Fluar 40x/1,30 Oil | 0,16 | M27 | 420260-9900-000 |
| α Plan-Fluar 100x/1,45 Oil | 0,11 | M27 | 421190-9900-000 |
| Ultrafluar 40x/0,60 Glyc | 0,36 | W 0,8" | 440015-9901-000 |
| Achroplan 10x/0,25 | 4,8 | W 0,8" | 440030-0000-000 |
| Achroplan 50x/0,90 Oil | 0,29 | W 0,8" | 440057-0000-000 |

Extended transmission from true UV 340 nm to 670 nm and special high numerical apertures are the hallmarks of the Fluars. The complementary Achroplan objectives offer a great combination of high transmission up to IR wavelengths and long working distances, with a good correction for dual labelled samples in the visual range.

Dipping Objectives

Developmental
Biology

Physiology,
Micromanipulation

For physiological experiments with fixed stage microscopes, the unique ZEISS W Achroplan and W Plan-Apochromat objectives are available. They combine long working distances in water with special inert coatings and excellent fluorescence transmission. The W Achroplan objectives are also



| Objective Type / Magnification / n.A. | W.D. (mm) | Thread | Order Number |
|---------------------------------------|-----------|--------|-----------------|
| W Achroplan 40x/0,80 W | 3,6 | W 0,8" | 440090-9901-000 |
| W Achroplan 40x/0,80 W IR | 3,6 | W 0,8" | 440095-0000-000 |
| W Achroplan 63x/0,90 W IR | 2,2 | W 0,8" | 440065-9901-000 |
| W Achroplan 63x/0,95 W | 2,2 | W 0,8" | 440067-9901-000 |
| W Achroplan 100x/1,0 W | 0,97 | W 0,8" | 440087-0000-000 |
| W Plan-Apochromat 63x/1,0 VIS-IR | 2,1 | W 0,8" | 441470-9900-000 |
| W Plan-Apochromat 20x/1,0 DIC VIS-IR | 2,0 | M27-70 | 421452-9900-000 |

available in versions corrected for the IR range up to 1100 nm. The W Plan-Apochromat objectives are corrected for both, visual range and IR. Hence they are ideally suitable for IR-microscopy and for multiphoton excitation. The new W Plan-Apochromat 20x/1,0 VIS-IR offers an outstanding numerical aperture, which makes it both the ideal one objective solution and the ideal multiphoton light collector.

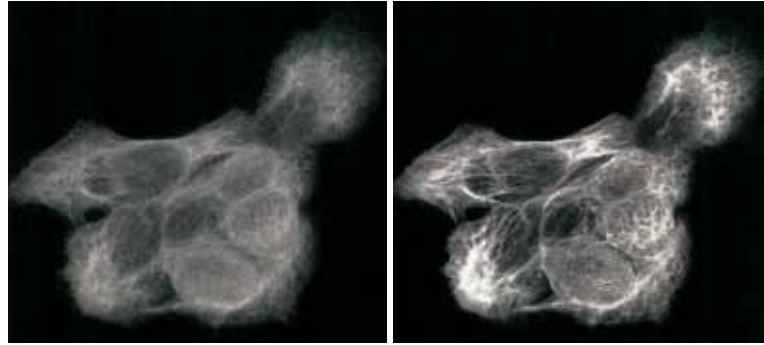
Accessories for Confocal Objectives

Developmental
Biology

Zoology, Botany
Neurobiology

To match even advanced requirements in confocal imaging, Carl Zeiss offers a very unique range of accessories to complement the LSM dedicated range of objectives.

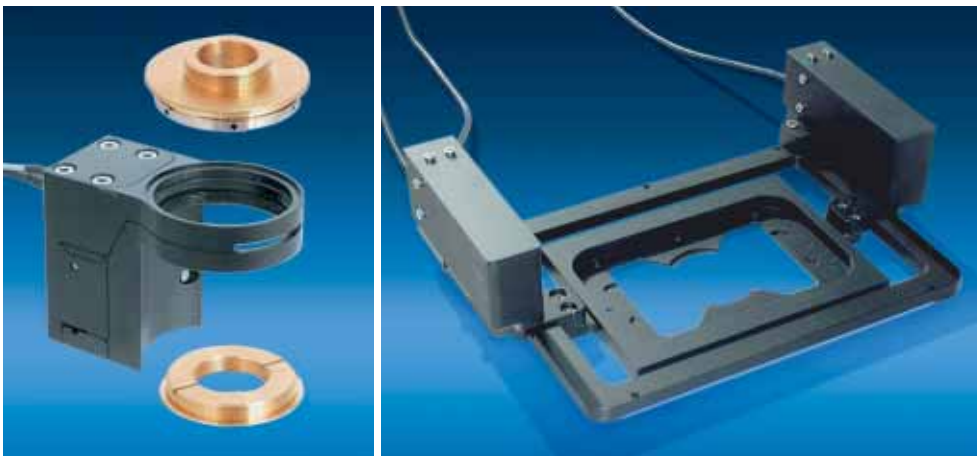
The immersion medium Immersol W with a refractive index of $n_e = 1.33$ exactly matches the C-Apochromat and LCI Plan-Neofluar multiimmersion range of objectives for demanding applications. While offering an ultimate solution for the effective reduction of spherical aberrations, it still maintains the physical advantages of immersion oils regarding evaporation and run-off behavior. Chromatic properties and temperature stability are nearly perfect, and handling even in inverted configurations is as easy as you ever wanted it to be.



Resulting image quality in an aqueous sample in case of the use of an immersion medium with incorrect refractive index (unsharp) and correct refractive index (sharp).

Immersol W matches the handling properties of conventional oil and the water-like refractive index of thick biological specimens.

In case an extremely fine Z-focus capacity is required without moving the specimen, the ZEISS piezo focus attachment, especially for physiological applications, adds ultrafine objective focusing to every microscope stand.



Piezo devices for faster and more precise objective and sample focusing.

Imaging Methods and Objective Types

| Objective Type | Magnification / n.A. | W.D. (mm) | Coverslip | Immersion | Thread | Order Number | UV-capable | NIR-capable | Color-Correction |
|-----------------|----------------------|-----------|-----------|-----------|--------|-----------------|------------|-------------|------------------|
| Fluar | 2,5x/0,12 | 6,3 | 0,17 | -- | M27 | 420120-9900-000 | ★★★★★ | ★★★★ | ★★ |
| Achroplan | 10x/0,25 | 4,8 | 0,17 | -- | W 0,8" | 440030-0000-000 | ★★ | ★★★ | ★★★ |
| Plan-Apochromat | 10x/0,45 | 2,0 | 0,17 | -- | M27 | 420640-9900-000 | ★★★ | ★★★★ | ★★★★★ |

| | | | | | | | | | |
|-------------------|---------------------------|------|-------|-----|--------|-----------------|-------|-------|-------|
| EC Plan-Neofluar | 20x/0,50 | 2,0 | 0,17 | -- | M27 | 420350-9900-000 | ★★★★ | ★★★ | ★★★★ |
| Plan-Apochromat | 20x/0,80 | 0,55 | 0,17 | -- | M27 | 420650-9901-000 | ★★★ | ★★★★★ | ★★★★★ |
| LD Plan-Neofluar | 40x/0,60 Korr | 2,9 | 0-1,5 | -- | M27 | 421360-9970-000 | ★★★★ | ★★★ | ★★★★ |
| EC Plan-Neofluar | 40x/0,75 | 0,71 | 0,17 | -- | M27 | 420360-9900-000 | ★★★★ | ★★★ | ★★★★ |
| Fluar | 40x/1,30 Oil | 0,16 | 0,17 | Oil | M27 | 420260-9900-000 | ★★★★★ | ★★★★ | ★★ |
| EC Plan-Neofluar | 40x/1,30 Oil DIC | 0,21 | 0,17 | Oil | M27 | 420462-9900-000 | ★★★★ | ★★★★ | ★★★★ |
| Plan-Apochromat | 40x/1,30 Oil DIC VIS-IR | 0,21 | 0,17 | Oil | M27 | 420762-9800-000 | ★★★ | ★★★★ | ★★★★★ |
| Achroplan | 50x/0,90 Oil | 0,29 | 0,17 | Oil | W 0,8" | 440057-0000-000 | ★★ | ★★★ | ★★★ |
| Plan-Apochromat | 63x/1,40 Oil DIC | 0,19 | 0,17 | Oil | M27 | 420782-9900-000 | ★★★ | ★★★★ | ★★★★★ |
| Plan-Apochromat | 100x/1,40 Oil DIC | 0,17 | 0,17 | Oil | M27 | 420792-9900-000 | ★★★ | ★★★★ | ★★★★★ |
| α Plan-Fluar | 100x/1,45 Oil | 0,11 | 0,17 | Oil | M27 | 421190-9900-000 | ★★★★★ | ★★★ | ★★★★ |
| α Plan-Apochromat | 100x/1,46 Oil Iris VIS-IR | 0,10 | 0,17 | Oil | M27 | 420796-9800-000 | ★★★★★ | ★★★★ | ★★★★★ |

| | | | | | | | | | |
|----------------------------------|--------------------------|------|-----------|------------|--------|-----------------|-------|-------|-------|
| C-Apochromat | 10x/0,45 W | 1,8 | 0,17 | W | M27 | 421747-9900-000 | ★★★★ | ★★★★★ | ★★★★★ |
| Plan-Neofluar Imm | 16x/0,50 Imm | 0,17 | 0,17 | Oil/Glyc/W | W 0,8" | 440530-0000-000 | ★★★★ | ★★★ | ★★★★ |
| LCI Plan-Neofluar | 25x/0,8 Imm Korr DIC | 0,21 | 0-0,17 | Oil/Glyc/W | M27 | 420852-9972-000 | ★★★ | ★★★★ | ★★★★ |
| LD LCI Plan-Apochromat | 25x/0,8 Imm Korr DIC | 0,60 | 0-0,17 | Oil/Glyc/W | M27 | 420852-9870-000 | ★★★ | ★★★★★ | ★★★★★ |
| Ultrafluor | 40x/0,60 Glyc | 0,36 | 0,2 | Glyc | W 0,8" | 440015-9901-000 | ★★★★★ | ★★★★ | ★ |
| C Achroplan | 40x/0,80 W NIR | 1,7 | 0,17 | W | W 0,8" | 000000-1080-378 | ★★★ | ★★★★ | ★★★ |
| LD C-Apochromat | 40x/1,1 W Korr UV-VIS-IR | 0,62 | 0,14-0,19 | W | M27 | 421867-9970-000 | ★★★★ | ★★★★★ | ★★★★★ |
| C-Apochromat | 40x/1,2 W Korr UV-VIS-IR | 0,28 | 0,14-0,19 | W | M27 | 421767-9970-000 | ★★★★ | ★★★★★ | ★★★★★ |
| C-Apochromat | 63x/1,2 W Korr UV-VIS-IR | 0,28 | 0,14-0,19 | W | M27 | 421787-9970-000 | ★★★★ | ★★★★★ | ★★★★★ |
| LCI Plan-Neofluar | 63x/1,30 Imm Korr DIC | 0,17 | 0,15-0,19 | Glyc/W | M27 | 420882-9970-000 | ★★★ | ★★★★ | ★★★★ |
| LCI Plan-Apochromat ¹ | 150x/1,35 Glyc Korr DIC | 0,18 | 0,14-0,18 | Glyc | M27 | 420792-9970-000 | ★★★★ | ★★★★★ | ★★★★★ |

| | | | | | | | | | |
|-------------------|--------------------|------|---|---|---------|-----------------|------|-------|-------|
| W Plan-Apochromat | 20x/1,0 DIC VIS-IR | 2,0 | 0 | W | M27 -70 | 421452-9900-000 | ★★★★ | ★★★★★ | ★★★★★ |
| W Achroplan | 40x/0,80 W | 3,6 | 0 | W | W 0,8" | 440090-9901-000 | ★★★ | ★★★★ | ★★★ |
| W Achroplan | 40x/0,80 W IR | 3,6 | 0 | W | W 0,8" | 440095-0000-000 | ★ | ★★★★ | ★★★ |
| W Achroplan | 63x/0,90 W IR | 2,2 | 0 | W | W 0,8" | 440065-9901-000 | ★ | ★★★★ | ★★★ |
| W Achroplan | 63x/0,95 W | 2,2 | 0 | W | W 0,8" | 440067-9901-000 | ★★★ | ★★★ | ★★★ |
| W Plan-Apochromat | 63x/1,0 VIS-IR | 2,1 | 0 | W | W 0,8" | 441470-9900-000 | ★★★ | ★★★★★ | ★★★★★ |
| W Achroplan | 100x/1,0 W | 0,97 | 0 | W | W 0,8" | 440087-0000-000 | ★★★ | ★★★ | ★★★ |

★, ★★ = standard performance

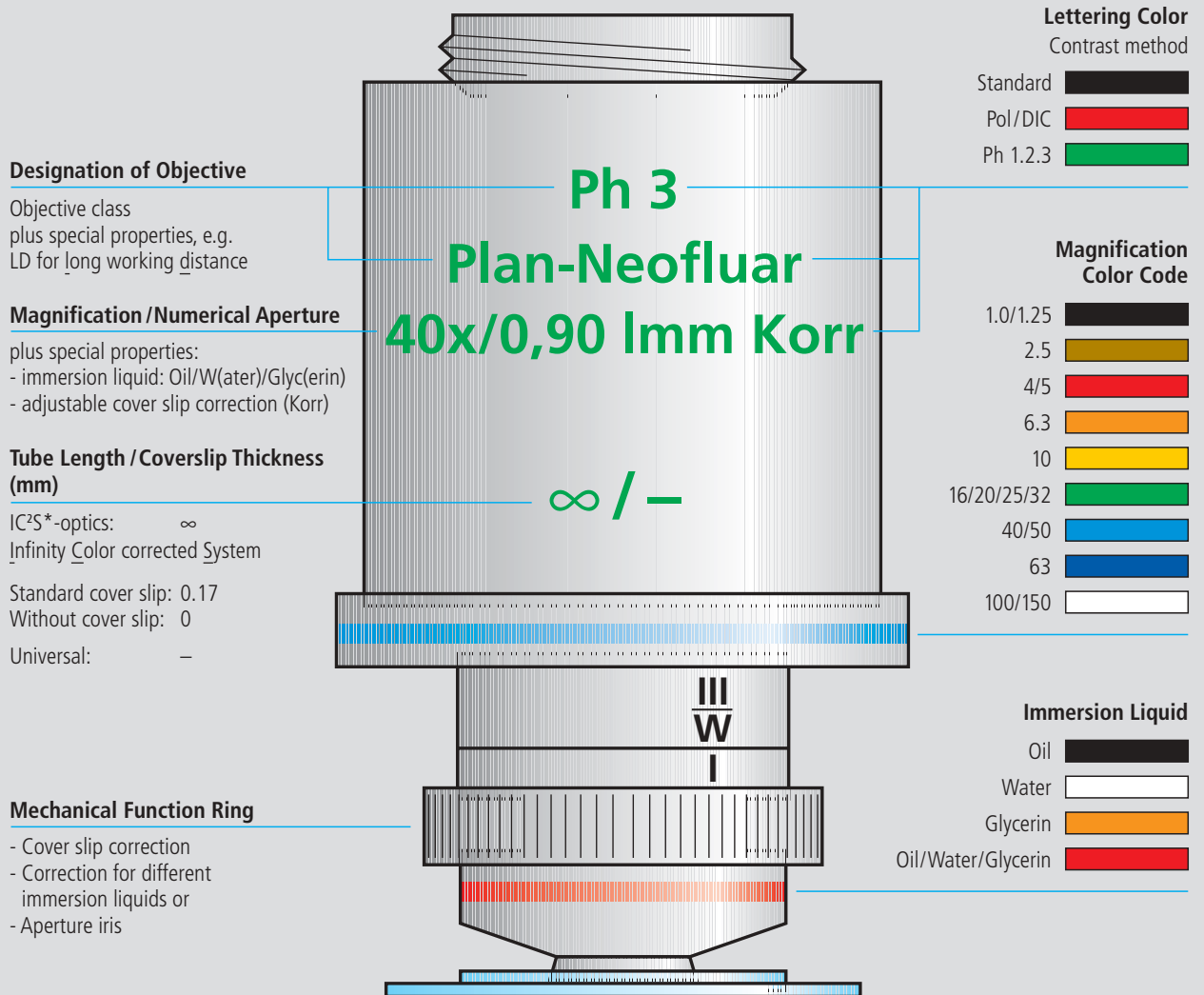
★★★, ★★★★ = good performance

★★★★★, ★★★★★ = excellent performance

¹ in prep for summer 2007

Markings on Objectives

(example)

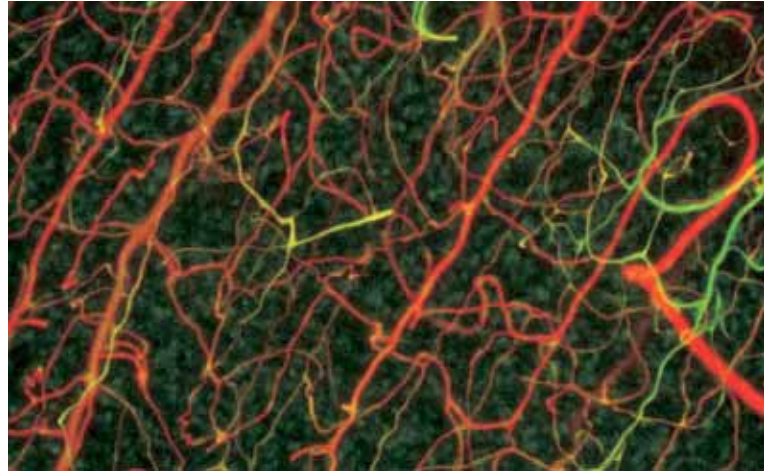


*The IC²S beam path – innovation for higher quality

Newly designed for Axio Imager, this is the result of the systematic optimization of the proven ZEISS ICS infinity optics. Its outstanding benefits include high image contrast, perfect homogeneity and unprecedented resolution plus integrated light traps for impressive performance. IC²S - Infinity Contrast & Color Corrected System.

Selected Objectives for Laser Scanning Microscopes

Carl Zeiss offers a large range of IC²S-objectives especially suiting the needs of confocal microscopy. The objectives selected for this brochure offer some of the best optical correction and transmission properties on the market. Low longitudinal chromatic aberration, low chromatic magnification difference, corrected spherical aberration and transmission values of 85 – 90% in the main wavelength range (70% at the borders given) combined with excellent image flatness create the best possible results. All this in combination with comfortable handling properties – that's Carl Zeiss, the pioneer of modern optics.



Blood vessels in brain tissue

Patents:

LSM 5 PASCAL and LSM 5 EXCITER

German Patents: 19702752C2, 69131176T2
US Patents: 5127730, 6037583, 6167173,
6462345, 6563632, 6665068,
6848825

LSM 510

German Patents: 19702752C2, 19702753C2,
19758744C2, 19758745C2,
19758746C2, 19758748C2,
19829981C2, 69131176T2
US Patents: 5127730, 6037583, 6167173,
6278555, 6462345, 6486458,
6563632, 6631226, 6848825,
6941247

LSM 510 META

German Patents: 10033180B4, 10038526B4,
19915137C2,
US Patents: 6403332, 6750036, 6858852,
6891613, 6958811, 7009699

LSM 510 NLO and LSM 510 META NLO

German Patents: 19919091C2, 69032621T,
69034117T
US Patents: 5034613, 6344653, 6403332,
6521899, 7119898



For further information, please contact:

Carl Zeiss MicroImaging GmbH

07740 Jena, Germany
Phone: +49 3641 64 3400
Fax: +49 3641 64 3144
E-mail: micro@zeiss.de

www.zeiss.de/micro