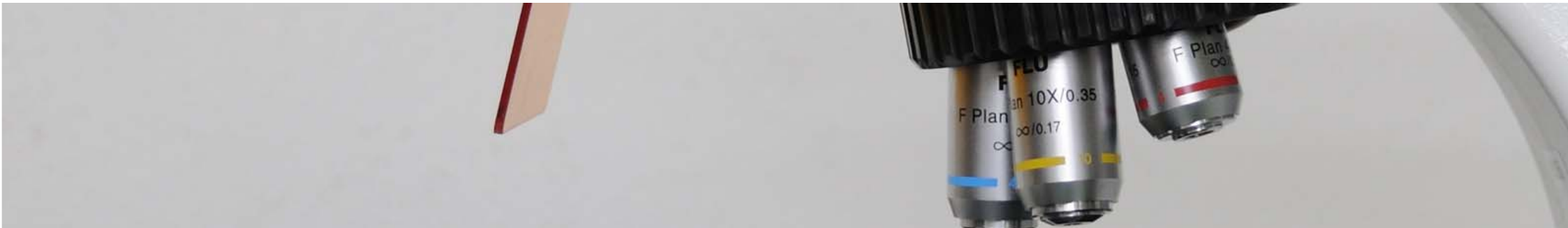
epi-fluorescence microscope

epi-fluorescence microscope

Certain substances by virtue of its chemicals structure are able to give out light of a determined wavelength after absorbing light of lower wavelength. This property is called fluorescence and it is characteristic of certain molecules as for example collagen, elastin, lignin or chlorophyll and a series of chemical compounds, the fluorochrome (FITC, DAPI, TRITC) with large and varied applications; among them two routine techniques in research and diagnosis laboratories, for carrying out them it is indispensable to use a fluorescence microscope:

- Immunofluorescence (IF): it consists of detecting and marking certain molecules of interest to biopsies or histology cuts through the utilization of specific antibodies conjugated with a fixed fluorochrome, this allows the diagnosis of certain diseases.
- Hybridization in place with fluorescence (FIPH): it consists of the hybridization of certain DNA fragments thanks to waves marked with fluorochromes, allowing mutations detection and genetic alterations therefore it is useful for antenatal diagnosis and for detection and diagnosis of certain tumors.



theoretical foundations

Epi-fluorescence o fluorescence of reflected light is basis on the incidence of a light beam of a determined wavelength. This sample absorbs energy from incident light and at the same time it emits light at a greater wavelength.

To allow this phenomenon is requiered the utilization of a filters system (cubes) with the following components:

- Excitation filter (XF): selects the light of the incident wavelength.
- Dichroic mirror (DM): reflects the light of certain wavelength while it allows to pass the light of greater wavelengths. Therefore, it reflects excitation light and it reaches the sample while it allows to pass emited light of the fluorescence substance.
- Emission filter or barrier (BA): selects fluorescence wavelength emited by the fluorochrome and it allows to reach to the eyepiece.

Following graphic shows an operating sketch of the epi-fluorescence microscope. Light comes from source (mercury lamp), it goes through a first filter which selects wavelength, this wavelength is able to excite the fluorochrome. This light is reflected in a dicronic mirror and it has an effect on the sample, exciting the fluorochrome which emits photons of a greater wavelenght than the incident. Emited light by the sample is not reflected but it goes through the dicronic mirror and it reaches a second filter which selects emission wavelength of fluorochrome, allowing to reach the eyepieces.

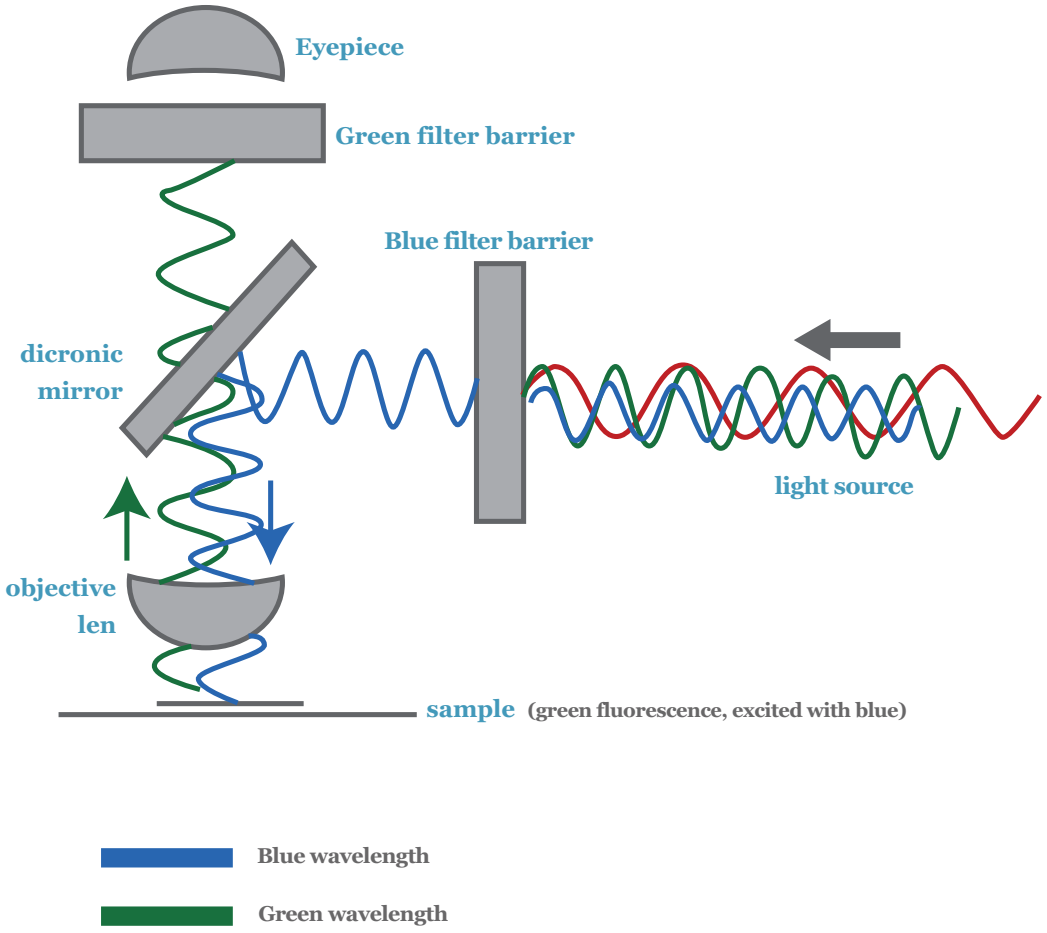
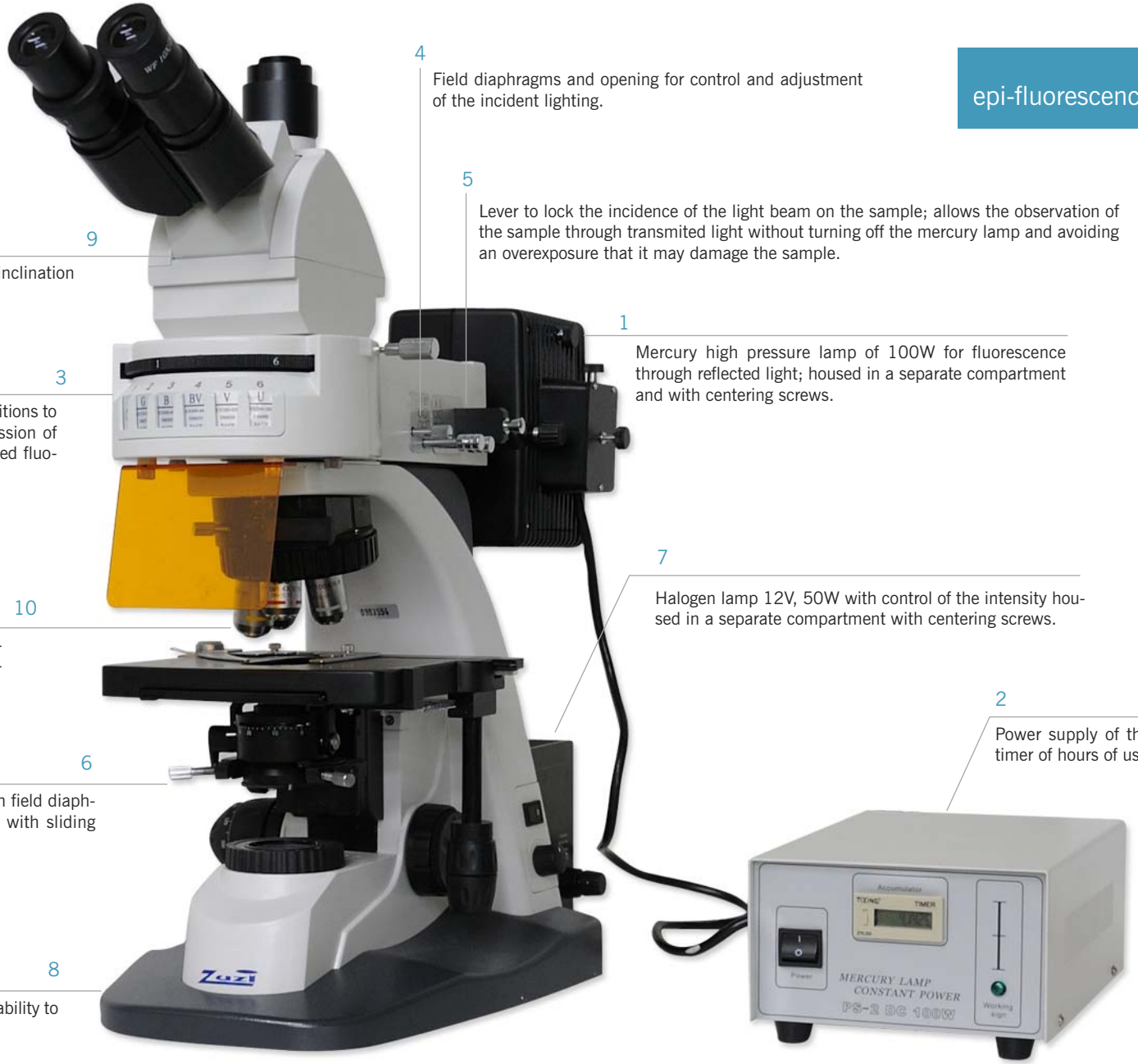


table filters

Turret position	Excitation filter (EX)	Emission filter (BA)	Dicronic mirror (DM)	Applicable fluorochrome
V (verde)	510-560	590	575	TRITC Phycoerythrin Ethidium bromide
B (azul)	450-490	520	505	FITC GFP Cy2
BV (azul/violeta)	400-440	470	455	Orange acridine Yellow acridine Alexa fluorine 488
V (violeta)	380-420	450	430	---
U (ultravioleta)	330-380	420	400	DAPI Alexa fluorine 350

epi-fluorescence microscope



4

Field diaphragms and opening for control and adjustment of the incident lighting.

5

Lever to lock the incidence of the light beam on the sample; allows the observation of the sample through transmited light without turning off the mercury lamp and avoiding an overexposure that it may damage the sample.

9

Head with adjustable inclination 0-40º.

1

Mercury high pressure lamp of 100W for fluorescence through reflected light; housed in a separate compartment and with centering screws.

3

Turret with different interchangeable positions to select excitation filter of cube and emission of wavelength more appropriate for the used fluorochrome (see filters table).

7

Halogen lamp 12V, 50W with control of the intensity housed in a separate compartment with centering screws.

10

Plane achromatic objetives with infinity corrected optical, specially designed for fluorescence applications.

2

Power supply of the mercury lamp with timer of hours of use.

6

Köehler system for transmited lighting with field diaphragms and opening and Abbe condenser with sliding lens of the optical way.

8

Broad and solid base that provides great stability to the equipment.

technical features

Code	50158358
Head	Trinocular, adjustable inclination 0-40º; adjustment of interpupil distance (50-75mm) and diopter correction
Optics	Infinity corrected
Eyepieces	WF10x/20
Nosepiece	Sixdruple
Objetives	Plane apo-chromatic of fluorescence
	4x, A.N.: 0.15
	10x, A.N.: 0.35
	40x (R), A.N.: 0.75
	100x (R), A.N.: 0.90
Stage	Two larger mechanical stage (180x160mm); displacement (80x50mm)
Condenser	Abbe (AN:1.25), with condenser sliding lens
Lighting	Köehler type with field diaphragms and opening
Light source	
Epi-fluorescence	Mercury high pression lamp 220V, 100W
Transmitted	Halogen lamp 12V, 50W

feature objectives table

Objetive	4x	10x	40x	100x
Numerical opening	0.15	0.35	0.75	0.90
Mechanical tube length	∞	∞	∞	∞
Thickness cover	0.17	0.17	0.17	0.17
Colour .	Red	Yellow	Blue	White
System	Dry	Dry	Dry	Dry
Work distance	21.6	2.9	0.6	0.62
Diameter visual field	6	2.4	0.60	0.24
Resolution	2.24	0.96	0.45	0.37

AUXILAB S.L.

Material de laboratorio
Laboratory supplies

Polígono Morea Norte
Calle D Nº 6 Beriain [Navarra]
C.P. 31191
Tel: +34 948 310 513
www.auxilab.es