Confocal Microscope (Zeiss LSM 510)	**Category:** **A. Particle Synthesis and/or** **B. Particle Labelling and/or** **C. Particle Characterisation in and ex-situ and/or** **D. In-vitro toxicity studies** **Institute: CIC biomaGUNE** **Location: Paseo Miramon 182 C** **20009 San Sebastian** **Gipuzkoa, Spain** **Contact Details of Technology Expert:** **Name, Dr.I.Llarena, Dr.Bogdam Sczchupak** **Phone,** **Fax,** **E-mail illarena@cicbiomagune.es;** **boguslaw@cicbiomagune.es**

Short technology description/Overview:

A confocal microscope creates sharp images of a specimen that would otherwise appear blurred when viewed with a conventional widefield microscope. This is achieved by placing a pinhole in front of the detector to exclude light from the specimen that is not from the microscope's focal plane. The resulting image has less haze and better contrast than that of a conventional microscope and represents a thin cross-section of the specimen. The sample can be imaged at varying depths or planes in the z-direction producing a stack of 2-D images (optical sectioning) which can be combined to render or create a 3-D image. In a laser scanning confocal microscope (LSCM), the laser scans across the sample in order to build up the image of the specimen. It is conventional to combine LSCM with fluorescence lifetime imaging and fluorescence correlation spectroscopy techniques. In fluorescence lifetime imaging microscopy (FLIM), the contrast is provided by the decay time of the excited fluorophores. The fluorescence lifetime depends on the local environment, and can be used to image changes in local parameters (e.g. pH, viscosity, refractive index) or to probe interactions with other molecules (e.g. collision, energy transfer). Fluorescence correlation spectroscopy (FCS) is an optical method for investigating kinetic processes and molecular interactions by statistical analysis of the fluctuations of fluorescence intensity arising from the Brownian motion of fluorescent molecules into and out of a small laser-illuminated volume. FCS has been successfully applied to measure the aggregation of particles, binding constants, conformational changes, and association of biomolecules.

Main Features (Equipment Capabilities):

- 7 excitation wavelengths (405nm, 458nm, 477nm, 488nm, 514nm, 543nm and 633nm) and a wide range of emission filters
- PMT and APD detectors
- Wide assortment of high quality objectives
- Z sectioning and 3D reconstruction
- Time series for following cellular events
- Emission fingerprinting
- Spectral analysis

- FRAP (Fluorescence Recovery after Photobleaching) experiments
- Co-localization of multiple proteins and/or dyes
- FCS (Fluorescence Correlation Spectroscopy) ability with ConfoCor 3 module
- FLIM (Fluorescence Lifetime Imaging Microscopy) ability with TCSPC module, 405 nm pulsed laser diode, and multidimensional data analysis software

Typical Samples & Images:

- The Multifluorescence Images & Intensity Measurement
- The scanning mode (1D, 2D, 3D) & 3D reconstruction
- The Timelapse Images & Physiology Analysis

Any further Information: