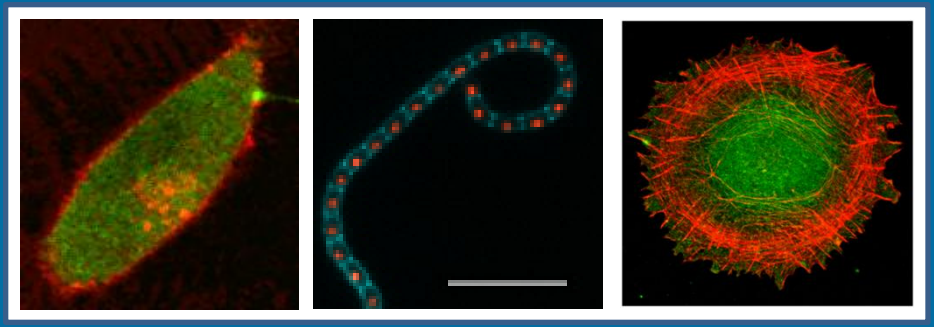
Uniwersytet Wrocławski

BIO-IMAGing in research **IN**novation and **E**ducation



UNIVERSITY OF WROCŁAW

Faculty of Biotechnology

Tamka 2,

Przybyszewskiego 63/77,

WROCŁAW, POLAND

Uniwersytet Wrocławski

Legend

I. **Confocal system** (LSM510 META - Zeiss) - Laboratory of Cytobiochemistry
II. **FACS - flow cytometer** (Becton Dickinson FACSCalibur™) - Laboratory of Protein Biotechnology
III. **Fluorescence microscopy system** (AxioObserver" Z1 Cell Observer - Zeiss) - Laboratory of Molecular Microbiology
IV. **Confocal system** (FlowView 500 - Olympus) - Laboratory of Cell Pathology

Confocal system

Laboratory of Cytobiochemistry

Prof. Dr. hab. Aleksander Sikorski

- **The system is open to all interested users.**

LSM510 META (Zeiss) upgraded with FLIM/F(L)CS (TCSPC) Facility (PicoQuant)

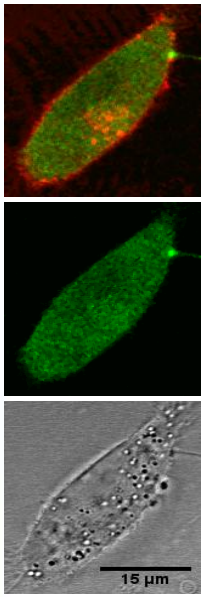
- **Features:**

- **META detector (412-748 nm)**
- **live cell imaging facility**
- **objectives: oil and water immersed (40x, 63x), dry (10x, 20x)**
- **two SPADs (possible FCCS measurements)**
- **filters for FLIM/F(L)CS**
- **laser lines [nm]: cw: 405, 454, 488, 514, 561, 633;**
  pulsed diode lasers: 470 (FWHM ~58ps), 635 (FWHM: ~59ps)
  repetition frequency 31,25kHz – 80 MHz.

- **Softwares:**

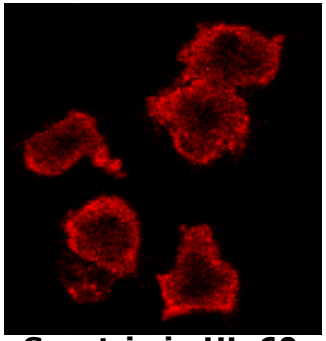
- **ZEN - support microscope operation and image analysis: FRAP, FRET, deconvolution, 3D projections, linear unmixing, colocalization, time lapse imaging,**
- **SymPhoTime – operation and analysis of time-resolved imaging/measurements: F(L)CS fitting to four models implemented: pure diffusions, triplet, conformational and protonation.**

Confocal system

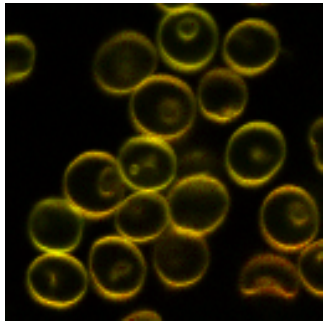


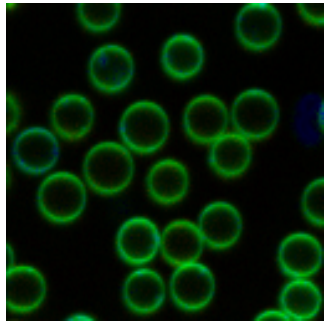
➢ Research focus and imaging techniques:

- imaging studies of fixed specimen,
- determination of fluorescent lipid diffusion in liposomes by FRAP,
- FLIM in lipid rafts imaging
- determination of interaction between eGFP-tagged proteins and fluorescently labeled,
- plasma membrane by acceptor bleaching FRET and FLIM-FRET,
- spectrine engagement in immunological synapse formation,
- determination of PKB activity in living cells with FRET based probe.

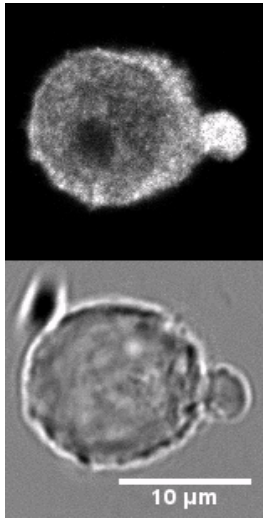


Spectrin in HL-60 during apoptosis





Di-4-ANEPPDHQ in human RBC (time-resolved image)



FACS - flow cytometer

Laboratory of Protein Biotechnology

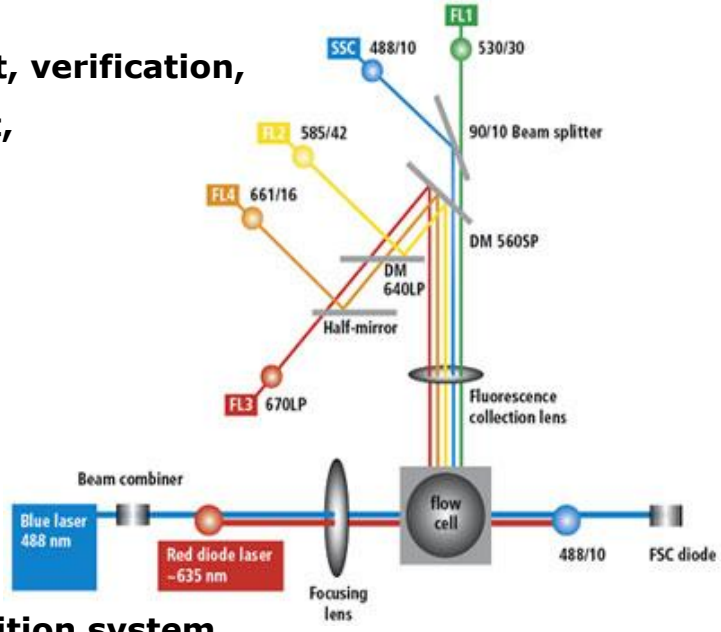
Dr hab. Ewa Marcinkowska

Becton Dickinson FACSCalibur™

➢ **The BD FACSCalibur flow cytometer was purchased by Wroclaw Research Centre EIT+ as an equipment necessary to complete the projects covered by the programme BioMed (access restricted).**

➢ features:

- **a multiparameter system**
- **system for routine cell analysis, assay development, verification, and identification of cellular populations of interest,**
- **dual-laser technology,**
- **4-colour analysis capabilities.**



➢ planned equipment base

- **separate equipment for efficient and precise cell sorting in sterile conditions,**
- **separate for detection of up to 18 colors simultaneously with 4 lasers and digital data acquisition system.**

FACS - flow cytometer

➢ Research focus and imaging techniques:

Projects:

• "Identification of molecular markers of diseases related to nuclear receptors dysfunction and implementation in diagnostics."

• "Synthesis and characterization of bionanomaterials for specific medical applications."

• "Employment of ferrum and haem assimilation by *Porphyromonas gingivalis* bacteria in the prevention and treatment of periodontopathy."

Measurements :

• of cell surface antigens expression,

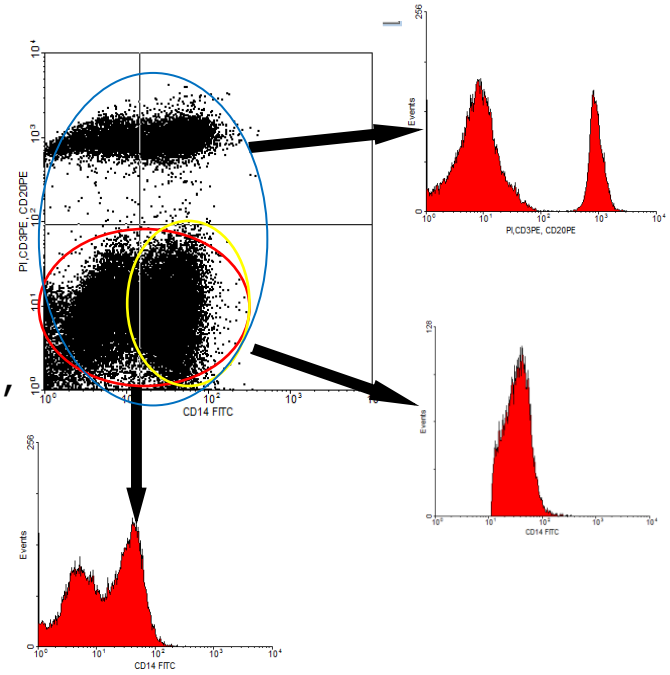
• of expression levels of intracellular proteins,

• of cytokine levels in blood serum

• of transfection efficiency using fluorescent proteins,

• detection of apoptosis using various techniques,

• cell cycle analysis using measurements of DNA content.

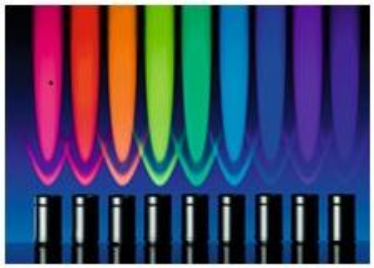


Fluorescence Microscopy Systems

Laboratory of Molecular Microbiology

Prof. Dr. hab. Jolanta Zakrzewska-Czerwińska

The microscope was purchased by Wroclaw Research Centre EIT+ as an equipment necessary to complete the projects covered by the programme BioMed (access restricted).



AxioObserver" Z1 Cell Observer (Zeiss)

➢ Features:

- **fully automated,**
- **BF, Fluorescence, Phase Contrast and DIC,**
- **filter sets (DAPI (49) , TexasRed (43 HE), GFP (38 HE), Cy 5 (50))**
- **COLIBRI system,**
- **dry and oil objectives (10x, 40x, 100x),**
- **combined with Definite focus (eliminate Z drift),**
- **modules multichannel, Z-Stack, time lapse, autofocus, mark & find, mosaiX,**
- **temperature, CO_2 control,**
- **AxioVision - image analysis software for acquisition and image processing.**

➢ planned equipment base

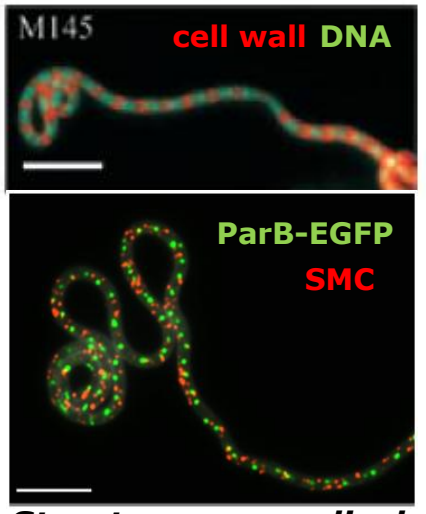
- **more advanced software for image analysis, system for FRAP.**

Fluorescence Microscopy System

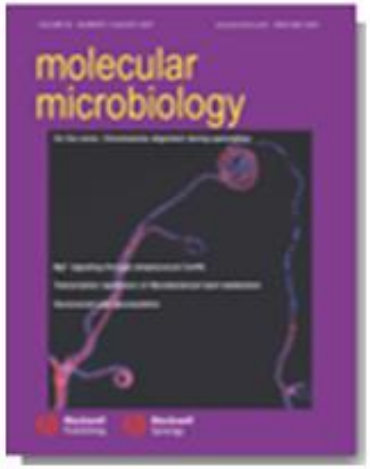
➢ Research focus and imaging techniques:

Projects:

- Chromosome replication, condensation and segragation in *Streptomyces* and *Mycobacterium*,
- Development of *Streptomyces venezuele,*
- Genome mining for drug discovery regulation of chromosome replication in *Streptomyces.*

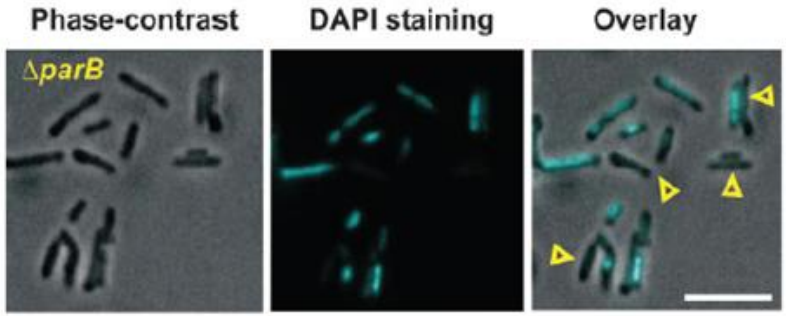


Streptomyces coelicolor



Techniques:

- localization of proteins and chromosomal regions (gfp, cherry fusion protein)
- observation of migration of proteins and chromosomes in **real time**
- immunostaing.



Mycobacterium smegmatis

Uniwersytet Wrocławski

Laboratory of Protein Engineering

Dr hab. Artur Krężel

- **Innovative imaging techniques**

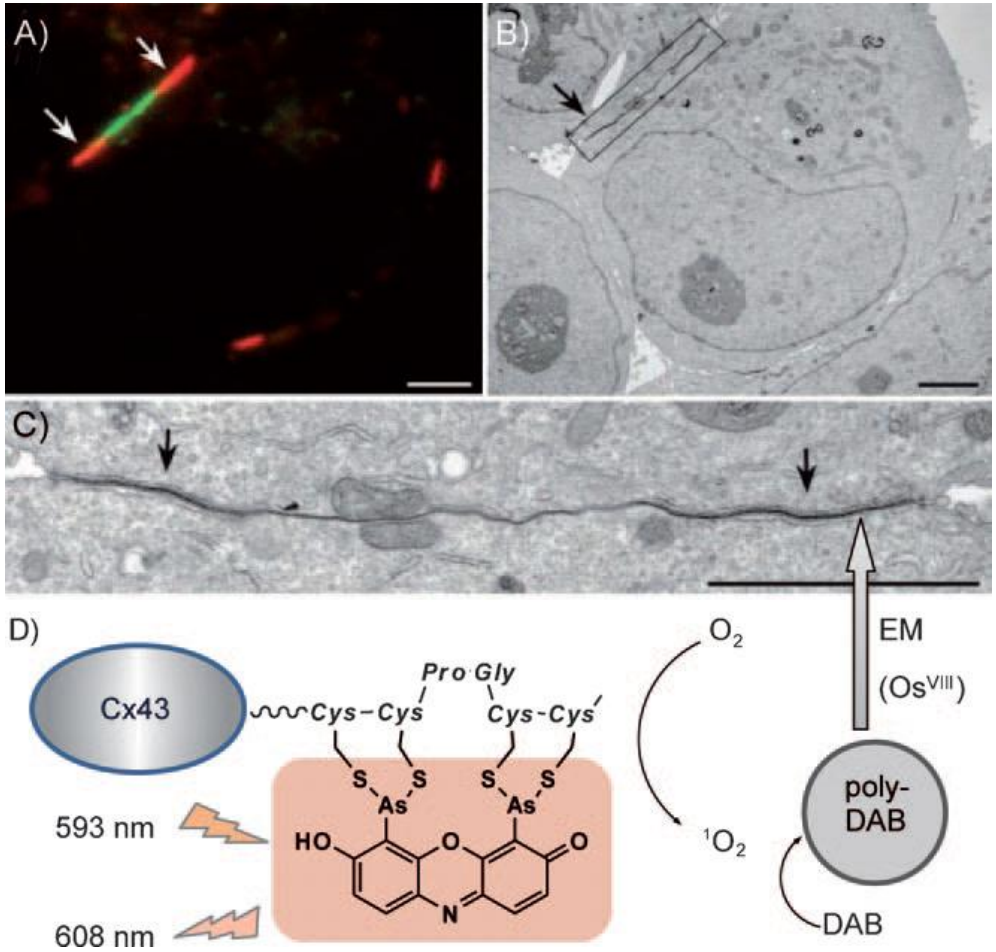
Biarsenical fluorescent tags

Two components:

•very short (6- to 12-residue), tetracysteine sequence (CCXXCC), placed at a protein terminus;

• nonluminescent biarsenical probe;

Protein contains the tetracysteine motif becomes highly fluorescent upon labeling with a nonluminescent biarsenical probe, and forms very stable covalent complexes.



Science **2002, 296, 503 –507**

Confocal system

Laboratory Of Cell Pathology

Prof. Dr. hab. Maria Malicka-Błaszkiewicz

- **The system is open to all interested users.**

- **Equipment:**

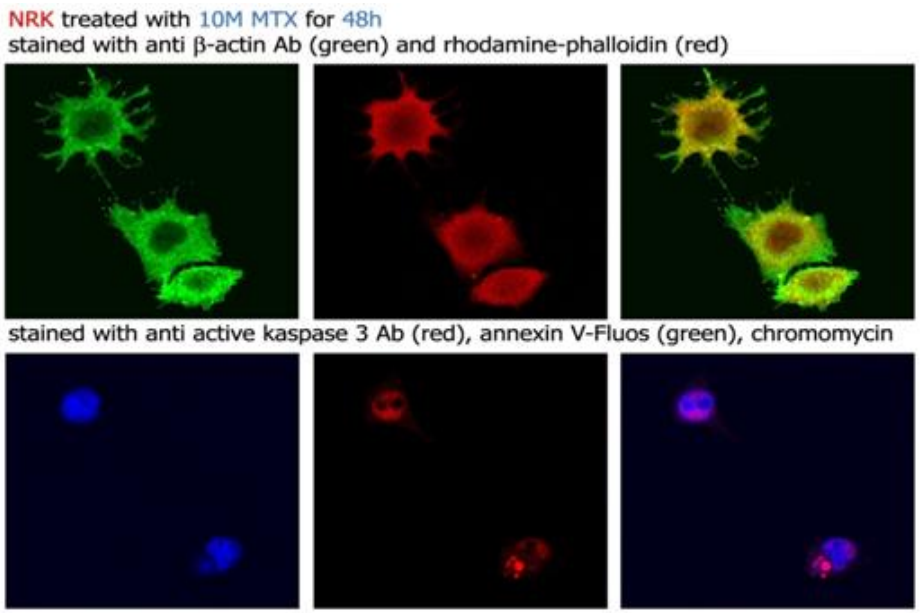
Confocal fluorescence microscope FlowView 500 Olympus

- **Research interest**

• The role of actin and actin binding proteins (ABPs) in the cell pathology and clinic;

• Drug induced apoptosis as a therapeutic tool;

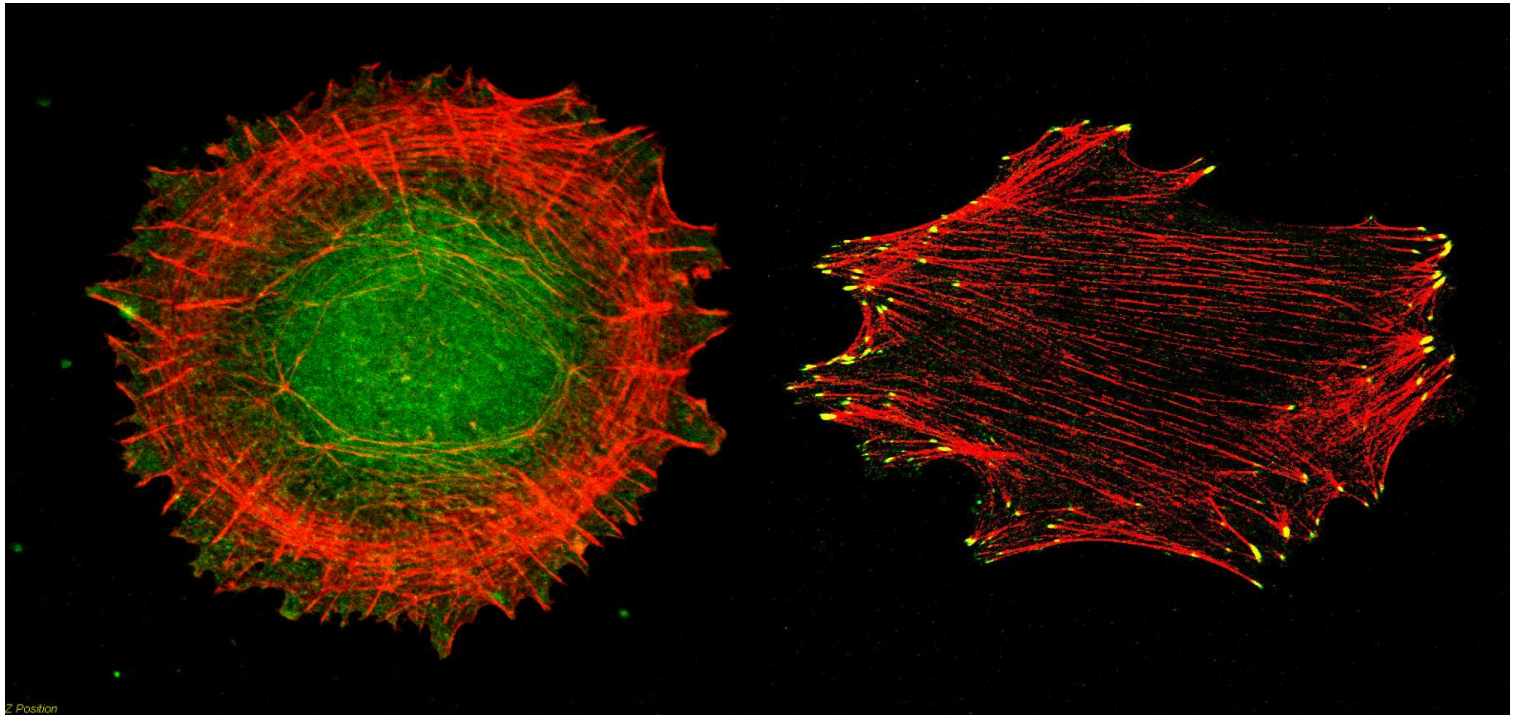
• Cell culture as a model in cytotoxicity assay and biomedical studies.



Uniwersytet Wrocławski

Confocal system

A 375 melanoma cell **Fibroblast**



green – gelsolin **green – vinculin**

red – rhodamine- phalloidin