

# MICROSCOPE-SPECTROPHOTOMETER FOR CELLS IDENTIFICATION.

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**KEY WORDS:** cell, single cell absorption spectroscopy, cell identification, microspectrophotometer, multispectral imaging, image processing.

For single cell identifications was created UV-visible-NIR microscope-spectrometer. It was used to measure spectra of microscopic samples (individual cells). This system consists of microscope connected with spectrometers. It is possible to registrations spectrum of transmission (absorption) or reflectance for microscopic sample (individual cells). Systems can be used for cytology, histology and for living cells. It is possible to study static or dynamics.

Microscope connected with spectrometers by means of fiber optics. Fiber optics cables are connected to the optical axis of microscope. The absorption spectra are recorded from ultraviolet (UV) to near infrared (IR) (from 350nm to 1100 nm). It is possible observation of cellular absorption by ultraviolet, optical and infrared light. Range of field for spectral measurements – is from 0.004 mm to 1 mm. It is possible to measure spectra for cell or some components of cell.

System applications – individual cells (stem cells) identification and sorting.

The first step of the procedure is the calibration. This is performed by inserting dark shutter in the optical path, and by setting the dark-current amplifier to zero.

Next step – light source calibrations.

Next step – educations – spectrum registrations for a priory known type of cells. It is possible for identifications one or more type of cells. For each type of cells it is need to registration big amount of spectrum examples. After educations it was created single spectrum for each type of cells.

Next step – cell identifications. We produced spectrum registrations for unknown cell. This spectrum is compared with spectrum of educational cells. It was used pattern recognition methods for spectrum identifications. To further assist in the spectral classification of individual cells, multivariate analysis was also applied.

Next step – sorting. It is used handle sorting. Microscope have left and right double holder microinjection. It is possible to sort cell by four class. Then individual cell was identified it was safe in suit microinjectors.

It is possible to use systems as microfluorimeter for registration full fluorescence spectra of cell. In this case it is need to remove emission (barrier) filter. For fluorescence registration it is need to increase time of accumulations.

UV and IR spectrum registration is a modification on UV and IR microscopy. The near IR range of an absorption spectrum contains features relating to the vibration absorption of the molecular bounds within the cell being examined.

If we use XY stage motors – it is possible to create full 2-dimentional spectral imaging. In this case we will get 3-dimentional imaging – X, Y and Lambda. For image processing in this case it was used special algorithms for cell identifications. It is need to use special methods for multispectral image processing. It is need specials methods for segmentations and image processing.