nanoVista

NEWSLETTER #3

PHOTONICS ANTENNAS FOR BIOLOGY
Tools for biosensing and cellular imaging

OBJECTIVES

- The first objective of NanoVista is to pioneer the technological development of **novel photonic antenna** geometries (probes & 2D arrays) for both **ultrasensitive detection** at **high sample concentrations** in fluids and simultaneous spatial **nanometric resolution** and **sub-ms** time resolution in living cells.
- The second objective of the European project is to develop **high-throughput large-scale nanofabrication** of photonic antenna arrays fully compatible with biomolecule detection and live cell nanoimaging.
- The third objective of the consortium is to demonstrate the bionanophotonic technology for **ultra-sensitive detection of biomolecules for diagnostic** purposes and for **nanospectroscopy on living cells**.

NEWS

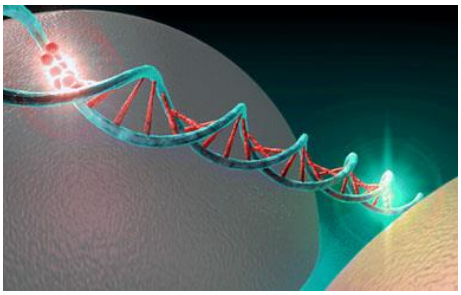
- COSINGO, Spanish technology SME, developed a compact, robust and highly sensitive instrument for biosensing and nanospectroscopy on living cells. A first demonstrator to perform fluorescence correlation spectroscopy (FCS)has been built and is under validation.

FCS accessory for inverted microscopes manufactured by COSINGO

- NanoVista collaboration between Institut Fresnel and ICFO in NanoLetters.

Förster resonance energy transfer (FRET) is commonly used as a nanoruler in biochemistry to monitor the short (nanometer) distance between donor and acceptor dyes. Yet FRET is equally sensitive to the mutual dipole orientation, which often complicates the distance analysis in biological samples. In the far field picture FRET is forbidden for perpendicularly oriented donor and acceptor dipoles. Luckily plasmonic nanoantennas do generate strongly inhomogeneous and localized fields. The non-zero near-field components in all directions open new energy transfer routes, which can overcome the limitations from the mutual dipole orientation and enhance the FRET efficiency.

In a collaboration between Institut Fresnel in Marseille, University of Montpellier and ICFO research groups led by ICREA Professors at ICFO Maria Garcia-Parajo and Niek van Hulst, nanoantennas with nanogaps have been optimized to bring about the increase in energy transfer efficiency for a DNA-based FRET system, with nearly perpendicular donor and acceptor dipoles, enabling an energy transfer which is simply forbidden in a homogeneous environment. The approach increases the applicability of single molecule FRET over diffraction-limited approaches, with the additional benefit of higher sensitivity and higher concentration range toward physiological levels. The work was published in Nano Letters, 16, 6222 (2016).



DNA-based donor-acceptor-pair in nanoantenna gap

PARTNERS

ICFO Institut de Ciències Fotòniques
Radboudumc university medical center
INSTITUT FRESNEL
EPFL ÉCOLE POLYTECHNIQUE FÉDÉRALE DE LAUSANNE
COSINGO Imagine Optic Spain S.L.
SEVENTH FRAMEWORK PROGRAMME

MORE INFORMATION: www.nanovista.eu

Project ref.: **288263**; Funded under: **Information and Communication Technologies**; Area: **ICT-2011.3.5**; Duration: **2011-2016**;

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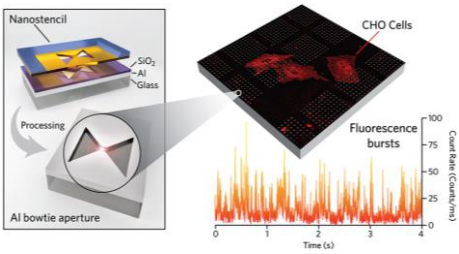
NEWS

▪ Large-scale arrays of photonic antennas for nanoscale dynamics in living cell membranes.

A study carried out in collaboration with researchers of the EPFL and ICFO has succeeded in fabricating hundreds of thousands of photonic antennas to measure for the first time the nanoscale dynamics of individual molecules in living cells. This work constitutes a major breakthrough in our ability to study biological processes in living cells at the truly nanometer scale and in real-time. The work, supported by EU project NanoVista, was published in Nano Letters, 15, 4176 (2015).

Photonic antennas amplify and confine optical fields at the nanoscale. As such, they break the sample concentration limit by overcoming diffraction, allowing the detection of individual molecules in solution at physiologically relevant concentrations (µM range). Unfortunately, extending this technology to live cell research is challenged by the difficulty of working with living cells and their inherent variability, requiring the development of large-scale antenna arrays while maintaining nanoscale control of their geometries. Now, EPFL researchers have developed a novel blurring-free stencil lithography-based patterning technique that relies on localized reactive ion etching to fabricate large arrays of nanoaperture-based antennas. The work demonstrates the reproducible fabrication of chips containing 12-antenna arrays for a total of over 400 000 antennas, with features controlled down to 20 nm.



Antenna arrays fully compatible with live cell research at the nanometer scale.

To validate their applicability on living cells, ICFO researchers used the antenna substrates as hotspots of localized illumination to excite fluorescently labeled lipids on living cell membranes. The high signal-to-background afforded by the antenna arrays allowed for the first time, the recording of single fluorescent bursts corresponding to the passage of freely diffusing individual lipids through hotspot excitation regions as small as 20 nm. Statistical analysis of burst length and intensity together with simulations demonstrate that the measured signals arise from the ultraconfined excitation region of the antennas.

The results of this joined collaboration establish that thru-stencil etching of metal nanostructures represents a cost-effective and scalable alternative for the fabrication of large arrays of photonic antennas fully compatible with life science applications. We foresee that these engineered substrates would become inexpensive, powerful tools to investigate the plasma membrane of living cells with nanoscale resolution at endogenous expression levels.

PROJECT MEETINGS

NanoVista comes to an end. It has been four years of fruitful research and collaborations. Partners of the consortium will meet to present results of the project and set the basis for future developments:

10th of January 2017 at EC headquarters (Brussels, Belgium), closing meeting with EC project officer and reviewers.

MORE INFORMATION: www.nanovista.eu

Project ref.: **288263**; Funded under: **Information and Communication Technologies**; Area: **ICT-2011.3.5**; Duration: **2011-2016**;

PUBLICATION HIGHLIGHT · NANOVISTA IN NATURE NANOTECHNOLOGY

Precision assembly of nanoparticles, top-down meets bottom-up.

EPFL researchers have developed a method to place and position hundreds of thousands of nanoparticles very precisely on a one centimeter square surface. This will open new doors in nanotechnologies.

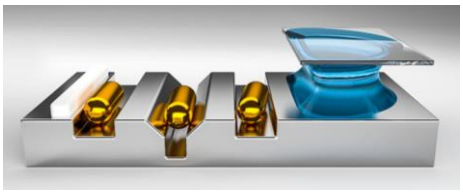
Whether it has to do with making pens or building space shuttles, the manufacturing process consists of creating components and then carefully assembling them. But when it comes to infinitely small structures, manipulating and assembling high-performance nanoparticles on a substrate is no mean feat.

Researchers in EPFL's Laboratory of Microsystems, which is headed by Jürgen Brugger, have come up with a way to position hundreds of thousands of nanoparticles very precisely on a one centimeter square surface. The nanoparticles were placed within one nanometer – versus 10 to 20 nanometers using conventional methods – and oriented within one degree.

Their work, which was published in Nature Nanotechnology, sets the stage for the development of nanometric devices such as optical detection equipment and biological sensors. "If we manage to place gold nanoparticles one nanometer apart, we could, for example, confine light to an extraordinary degree and detect or interact with individual molecules," said Valentin Flauraud, the lead author.

"Playing golf" with nanoparticles

For their study, the researchers used gold nanoparticles that were grown chemically in a liquid. "These nanoparticles exhibit better properties than those produced through evaporation or etching, but it is more difficult to manipulate them, because they are suspended in a liquid," said Flauraud.



A solvent drop is dragged over a patterned template and the particles assemble in the grooves

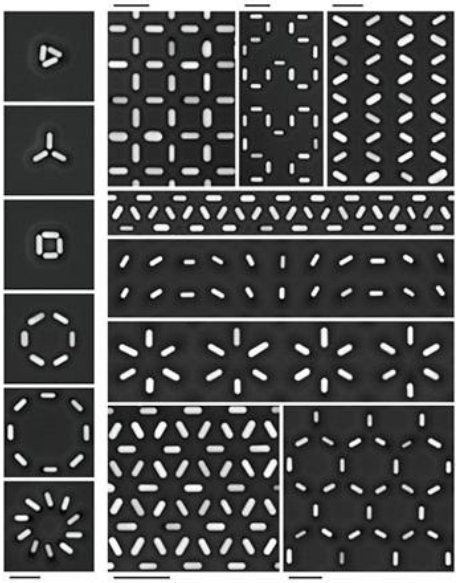
Their technique consists of taking a drop of liquid full of nanoparticles and heating it so that the nanoparticles cluster in a given spot. This drop is then dragged across to a substrate with nanometric barriers and holes.

When the nanoparticles encounter these obstacles, they detach from the liquid and are captured by the holes. "It's a little like playing miniature golf," said the researcher. Each trap is designed to orient a nanoparticle in a specific way. "The challenge was to figure out how the liquid, the particles and the substrate interact at the nanometric scale so we could trap the nanoparticles effectively," said Massimo Mastrangeli, the second author and now a researcher at the Max Planck Institute for Intelligent Systems in Stuttgart.

Writing out with nanoparticles

To show how well their method works, the researchers took on several challenges. First, they tested the optical properties of their system with a powerful transmission electron microscope in EPFL's Interdisciplinary Center for Electron Microscopy (CIME).

They then showed that their technique could be used to produce geometrically complex structures by writing out with nanoparticles. "All of this work is the result of strong synergies between the various technical platforms and the labs," said Professor Brugger. "It's an excellent example of how top-down and bottom-up methods can be combined, opening the door to numerous unexplored fields of nanotechnology."



Examples of two-dimensional patterns of Au nanorods fabricated by topographically templated capillary assembly (scalebars: 250nm)

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