FINAL ACTIVITY AND MANAGEMENT REPORT

To be completed by researchers who are:

-*Fellows benefiting from a Marie Curie Intra European Fellowship;*
-*Fellows benefiting from a Marie Curie Return and Reintegration Grant;*

To include as annexes:

- *list of participation in conferences;*
- *list of publications;*
- *list of patents .*
- *list of other outcomes considered useful to assess the work done during the fellowship;*

This report is to be completed and signed together by the fellow and the scientist in charge.

Type of Marie Curie action:	Intra-European Fellowship
Contract No.:	235323
Title of the Project[1]:	PhotoNanoFluidix
Duration of fellowship/appointment/training period (Start Date – End Date[1]):	01/05/2009 -30/04/2011

[1] Pre filled when applicable

1. PUBLIC OUTREACH: PUBLISHABLE SUMMARY OF WORK PERFORMED AND RESULTS

Include all main keywords referring to the objectives and results of this project, stressing ***the most important scientific achievement made.*** *Write clearly and concisely, and make it understandable for the general public.*

Particles in liquids are continuously buffeted about by random collisions with the surrounding bath of molecules. This means that a small particle in a fluid continuously tends to just wander away. However, fascinating things could be done if one could "trap" tiny objects, a thousand times smaller than the width of a human hair, in water. Researchers have developed several different ways to do this, all of these methods entail the application of external forces to the object to hold it in place; and the smaller the object the harder the task. In our work we demonstrated an entirely passive way to trap an object in solution that works just as well for small objects as for larger ones, as long as the object carries a certain amount of charge. This new technique should enable researchers in fields ranging from biology to materials science to address fundamental questions and develop new technologies.

The aim of the proposal centered on investigating geometry-induced effects on the dynamics nano-objects in confined spaces. Based on an improved understanding of the interactions we sought to optimize the self-assembly of discrete charged metal or dielectric nano-objects into arrays with lattice constants comparable to the wavelength of light and to explore the use of these structures in photonics applications.

The major goals of the project have been met and resulted in a publication in Nature in 2010. In this publication we described a geometry-induced electrostatic trap for confining, arranging and manipulating single nanometric objects in solution.

List of Keywords

Electrostatic trap, nanofluidics, directed assembly

Websites where additional information may be found

2. REPORT ON WORK PERFORMED AND RESULTS

Please report on the work performed and on the results of the research (maximum 4 pages), addressing the following points clearly and concisely:

a) Accomplishment of research objectives as presented in the original proposal.

b) New objectives established during the course of work and new lines of research.

The following structure should be used in the description of points a) and b) for each objective separately.

- *Objective of the research;*
- *Work performed (mentioning also unsuccessful approaches and unforeseen developments);*
- *Results and degree to which the objectives were met;*
- *List specific training received on scientific and technical aspects;*
- *Relevance for basic and applied science and for applications, including industrial links.*

Changes to original proposal: Note that the Commission has to be informed in advance of any changes to the original proposal. For point a) it is important that any deviations from the original proposal are clearly indicated.

In order to help illustrate the work carried out during the fellowship, please enclose copies of the most relevant publications and reports as well as abstracts of other publications and manuscripts. Note that this is in addition to the free-hand report requested above.

The objectives of the research were to extend preliminary investigations on the attractive interactions arising between colloidal objects and like-charged confining walls in low-ionic-strength solution, in order to:

- better understand the *origin of the force* and its dependence on crucial system parameters such as ionic strength, particle size and confinement dimensions.
- optimize the *self-assembly of discrete charged metal or dielectric nano-objects* into arrays (or arbitrary arrangements) with sub-100 nm lattice constants.
- use the developed nanoparticle self-assembly method to *study phenomena in nano-optics*, e.g., plasmonic coupling of resonantly excited metal nanoparticles, modification of fluorescence emission of single emitters diffusing in solution very close to discrete metal nano-objects, realization of novel ordered and disordered arrangements of nano-objects (e.g. dielectric particles like TiO2) for studying light scattering phenomena in two dimensions.

Objective 1 centered on achieving an improved understanding of unexpected geometry-induced effects on the dynamics of confined charged colloids and macromolecules. The research carried out under this project revealed that the phenomena described in Nanoletters 2007 (Section B.1.2.1) and that described in Section B1.2.2 (unpublished at the time) of the proposal were two separate effects. The former effect is still a topic of investigation from the theoretical perspective and has not yet been explained comprehensively. We focused on the latter of the two confinement-induced effects (Section B1.2.2) in this project, as it holds greater promise and flexibility for manipulating matter at the nanoscale of relevance in fields from single molecule biophysical research to materials science and photonics. We performed numerical calculations of the electrostatic potential using Poisson-Boltzmann (PB) theory in order to explain the observed trapping of single nano-objects. We were successfully able to explain the experimental observations and performed experiments outlined in Section B1.4.1 in order to characterize the phenomenon and check agreement with theoretical predictions. These experiments involved scattering interferometric imaging of trapped gold nanoparticles solutions of various ionic strengths and different channel geometries. Encouraged by the excellent agreement between the theory experiments we made further progress by demonstrating the trapping of single lipid vesicles and proposing conditions under which the effect could be harnessed to trap single nanometric objects as small as proteins. This is well beyond what was anticipated in the original proposal; the results were published in Nature 467, 692-695 (6 October 2010) (1), a copy of which is appended to this report.

A crucial achievement in the course of our studies on the mechanism of the particle trap was the three dimensional spatial tracking of individual trapped particles. This was not initially proposed in the grant proposal and was possible due to the unique expertise available in the host laboratory. Preliminary studies were done via a collaboration with Philipp Kukura, a postdoctoral researcher working with the scientist-in-charge. Subsequently, together with Nassir Mojarad, a PhD student in the host lab, we went on to perform in-depth spatial characterizations of single gold particles in traps of various geometries. We have found that comparing the measured behaviour of a particle in a trap with calculations could enable us to measure its charge and size. Since the method is inherently suited to massively parallel measurements it could be possible to characterize a colloidal suspension at a level of resolution impossible using current techniques based on light-scattering. We are currently in the process of submitting a manuscript on this work. The ability to characterize and study the properties of single objects based on their behaviour in a trap and the practical implications thereof, were not envisioned in the original proposal.

Experiments on trapping particles in a non-aqueous environment were planned initially as outlined in the original proposal. Organic solvents were purified and gold particles suspensions in organic solvents were prepared. However, as investigations of the phenomenon in the aqueous phase alone opened so many avenues of research, experiments on the effect of different dielectric media were not pursued.

In order to further refine the theory work, the researcher implemented a free energy calculation based on the numerical calculation of the PB electrostatic energy for the full three dimensional system, including the contribution of the object itself. The full calculation provides a framework for detailed comparisons of measured and predicted behaviour of the particle in the trap. Comprehensive experimental and theoretical work will be of vital importance in fostering the use of this methodology in areas from single molecule biophysics to photonics and a manuscript on the topic is currently under preparation. The significant progress made on the theoretical front has been a backbone of the project and were not envisioned at the time of writing the proposal.

The goals of *Objective 2* followed from the results obtained in Objective 1. We demonstrated the ability to self-assemble lattices of metal (gold) and dielectric (polystyrene) particles at lattice spacings comparable to and smaller than the wavelength of light. Some of the results are published as supporting material in (1). Calculations showed that assembly of lattices with spacings slightly larger than the diameter of the particle would face challenges owing to interparticle repulsions. The outcome of the investigations in Objective 2 are likely to have their strongest impact in the applied sciences, although studies on interparticle interactions in confined environments are also important from the point of view of basic sciences. Further detailed studies in the area of Objective 2 were superseded by interesting questions that arose in the context of Objective 3 discussed below.

Objective 3 focused on using nanoparticle self-assembly to study nano-optic phenomena and developing devices for plasmonic detection of single emitters using conventional fabrication techniques. Within the scope of this objective the researcher developed new methods not only to position particles in high-density arrays but also to control the orientation of asymmetric nanoparticles such as nanorods in an assembly. With the current level of interest in photonic structures based on nanowires and nanorods, the ability to control assemblies of nanorods is of significant scientific importance and the ability to do so within the framework of this technique was not anticipated in the original proposal. Therefore, although the researcher invested significant time and effort in fabricating plasmonic nanofluidic structures by traditional nanofabrication techniques and characterizing these structures for detection of single emitters, the focus of Objective 3 was directed towards the control and assembly of photonic structures based on nanorods. Samples of silver nanorods were obtained from a collaboration with the group of Carsten Soennichsen, Univ. of Mainz, Germany. We used metal nanorods as test objects as their plasmon resonance properties can be used effectively to probe particle orientation using polarized light. Since the method is based on electrostatic charge, the technique can be extended to semiconductor nanowires, polymer and dielectic rods as long as the particles carry some next charge. These experiments resulted in our demonstrating for the first time the ability to control the orientation of single nanorods in solution. A manuscript on this work is currently under preparation.

To summarize, the goals of Objectives 1 and 2 of the original proposal have been met. Work on Objective 3 is currently in progress.

New lines of research that were developed in the course of the project were as follows:

- High resolution 3d tracking of single trapped objects for improved comparisons with theory
- Numerical calculations of the system free energy as a predictive design tool for experiments
- Oriented trapping of asymmetric objects such as nanorods, and in general, the ability to trap nano-objects in specific orientations based on their shape
- Trapping of biological soft objects such as vesicles, macromolecular complexes and proteins

A *major new line of research* that was established in the course of the project evolved from recognizing that the current nanofluidic trapping methodology could be applied not only to assemble structures relevant for photonics but also to trap single charged macromolecular complexes and proteins for biophysical and bioanalytical studies. Trapping single proteins in solution has been a significant scientific challenge over the last decade. Collaborations have since been established with Prof. Ben Schuler, Institute of Biochemistry, University of Zurich on the trapping of single macromolecular complexes – GroEL and with Prof. Nenad Ban, Institute for Molecular Biology and Biophysics, ETH Zurich in the trapping of single ribosomes in solution. Initial experiment on the GroEL complex revealed issues related to its stability at the lower ionic strengths needed for proof-of-principle demonstrations. We are working together with the Schuler group to address these and related biochemical issues.

Work on both the trapping of single proteins and macromolecular complexes as well as nanorod control and assembly have yielded promising initial results and were the focus of the latter phase of the project.

In general, a major new line of research arose from a better fundamental understanding of the trap mechanism for a single object. As a result, the possible applications of controlling nano-objects using geometry-related effects in nanofluidic confinement saw an explosion in comparison with what was envisaged at the time of the proposal submission. The applications in biological/biophysical questions were not foreseen and arose exclusively from understanding the theoretical underpinnings of the phenomenon. Further unforeseen applications in basic and applied science have arisen from the ability to study in great detail the spatial behaviour of a single trapped object.

Relevance for applications: Given the broad applicability of techniques to position and control single colloidal objects in fluids, the researcher and scientist-in-charge filed an invention disclosure at ETH Zurich in May-June 2009. The patent application was not pursued further as the results were judged too preliminary for immediate commercial relevance. Nonetheless, we feel that the techniques developed in this project will find numerous applications upon into a mature technology.

Training received: A major training goal achieved within this project was that the researcher gained experience in an area of expertise of the host laboratory, namely *label-free imaging of matter at the nanoscale*.

The researcher also attended a class in *Experimental Methods in Quantum Optics* and received training in *presentation skills* via a workshop organized by the host laboratory.

MANAGEMENT REPORT

Please justify any deviations and/or modifications to the initial financial planning of the project.

3. ASSESSMENT BY THE SCIENTIST IN CHARGE ON THE FELLOW'S WORK DURING THE FELLOWSHIP

Dr. Krishnan has successfully established a new line of experiment in our group, where nanofluidic devices consisting of nanoscopic topographic indentations are used to manipulate tiny nanoparticles. The underlying mechanical potential for this mechanism is caused by electrostatic charge distribution on the walls of the nanofluidic device. The great advantage of this technique is that it allows application of substantial forces on small particles, which would not be easy to manipulate via optical tweezers and related methods. Dr. Krishnan and her collaborators in the group have demonstrated trapping of a range of nanoparticles as small as 20 nm in size. In addition, they have mapped the motion of the nanoparticles in three dimensions with nanometer accuracy and a high temporal resolution. In addition to the design and fabrication of the nanofluidic devices, Dr. Krishnan has also performed extensive calculations and simulations to investigate the performance of the geometry-based electrostatic traps and guides. The results have been very encouraging and have shown a very good agreement with the measured data on the trap potentials.

After demonstrating the basic operation of the nanofluidic three-dimensional traps, which was published in Nature, Dr. Krishnan has pursued several possible applications. One direction has been to see if proteins can be trapped. Here the challenge is not the size or charge of the protein, but the fact that the trap potential depends on salt concentration; the trap is weakend for physiologically relevant concentrations. However, smaller geometries can, in principle, recover a sufficiently deep potential even at salt concentrations of 30 mM. Another application prospect that Dr. Krishnan has followed is the development of traps for asymmetric particles. The strategy has been to carve the topographic indentations according to the shape of the particle to be trapped; the simplest example of such particles is the rod shape.

Dr. Krishnan has successfully carried through the project that was discussed in the proposal. However, her work has also opened unforeseen doors for trapping and sorting nanoparticles according to charge, shape or orientation. These features are currently not available by any other means and therefore promise to raise great attention. Overall, Madhavi Krishnan has proven to be a very determined and ambitious young scientist with great promise for an academic career.

The undersigned agree:
- that the Commission may publish information contained in this report **Yes** ☒ **No** ☐
- to allow the Commission to divulge future contact details to national representatives or to organisations carrying out services for the Commission **Yes** ☒ **No** ☐

Signatures:

Name of fellow: Madhavi Krishnan	Name of scientist in charge: Vahid Sandoghdar
Date: 1 May 2011	Date: 1 May 2011
Signature of fellow:	Signature of scientist in charge:
________________________________	________________________________

RESEARCH RESULTS (Summarise the results obtained by the contractor since the beginning of the project)**:**

Participation in conferences and other scientific events:

Please indicate the number of participation to scientific events by the beneficiary of the contract. List the participation on a separate sheet following the classification used below.

	Number			
	Active participation			Passive participation
	Oral	Poster	Of which were invited presentations (oral + poster)	
Conferences	3			
Workshops				
Other Scientific Meetings	4		3	

Patents:

Please indicate the number and status of patents, which have been the direct results of the research project. List the patents on a separate sheet giving their complete reference number and briefly stating the applicability of each patent.

	Number of Patents		
	Application filed	Pending	Granted
National Patents:	--	--	--
- Member States and/or Associated States			
- Third Countries:			
- US			
- Japan			
- Other			
European Patents (EP number):			
International Patents (WO number):			

Publications:

Please indicate the number of publications resulting directly from the project. List the publications on a separate sheet following the classification used below, indicating any invited contributions. In publications resulting from collaboration with other institutions, indicate name and country of institution.

	Number of Publications		
	As main author	Total	Of which were co-authored with researchers from other institutions
A. Peer Reviewed ***(incl. in press)***			
- Articles in Journals	1	1	0
- Chapters in Books			
- Articles in Conference Proceedings			
- Books and Monographs			
B. Non-Peer Reviewed ***(incl. in press)***			
C. Submitted			
D. Manuscripts in preparation	3	3	1

Teaching and Transfer of Knowledge:

Please indicate the number of hours of lectures, which have been delivered by the beneficiaries of the project and training courses, which have been organised by the contractor(s) on the research carried out in the project. List on a separate sheet the lectures and/or training courses delivered.

		Number of participants	
	Number of Hours	Early stage researchers	Other
Lectures	--		
Training Courses	--		

Other outcomes:

Please list other outcomes of the project than those mentioned above. Such outcomes may be further academic qualifications, spin-off companies, prizes, awards, media coverage, etc.

	Number	Type
Academic qualifications		
Prizes and Awards **Media Coverage**	1	Newspaper article in leading Swiss daily, Neue Zürcher Zeitung (NZZ) (Appended to the report.)
Spin-off companies		

Annex I

Presentations at meetings & seminars:

• "Geometry-induced trapping, levitation and assembly of nanometric objects in a fluid" , 1st *European Optical Society Optofluidics Meeting*, Munich, Germany, May 23, 2011.

• *RNAi and Single Cell Biology Meeting* 2011, Apr 4, 2011, Boston, MA.

• *Physical Sciences Seminar, University of Gothenburg,* Gothenburg, Sweden. Dec 14, 2010. [invited]

• *École Polytechnique Fédérale de Lausanne*, Lausanne, Switzerland. Oct 15, 2010. [invited]

• *Physical Chemistry Colloquium*, *University of Zurich*, Zurich, Switzerland. Sep 23, 2010. [invited]

• *Second International Conference on Nanobiotechnology*, Zurich, Switzerland. Aug 26, 2010.

• *Rowland Institute at Harvard*, Boston, MA. Jun 12, 2010.

Other Activities:

1of 4 researchers at ETH Zurich in a panel discussion on "*Scientific Research and Society*" with former UN Secretary General Kofi Annan, on the occasion of the second Richard R. Ernst Lectureship, Zurich, Switzerland, June 18, 2010.

Manuscripts in preparation:

- "Oriented trapping and assembly of nanorods in a fluid"
- "Measuring the properties of single nanometric objects using an electrostatic fluidic trap"
- "The free energy of an object in an electrostatic fluidic trap"