BLIND DECONVOLUTION AND 3D PSF MODELING IN BIOLOGICAL MICROSCOPY

1 Introduction: research topic and state of the art

Multidimensional microscopy is an essential tool for research and industry in the areas of cellular biology and molecular medicine, cell-based drug discovery and cellular therapies. Modern microscopic methodologies have brought the possibility to follow live cells in action, responding to various perturbations. These capabilities include not only detailed dynamic information about cell morphology, but highly sensitive spatio-temporal data about the behavior of specific proteins in cells. Experimental systems are being developed to model mechanisms in healthy and sick cell lines, and probe the various components mediating these mechanisms, thus resolving the molecular networks underlying complex cellular processes.

While microscopy has been vastly employed in the analysis of abundant proteins, the detection of rare ones is facing difficulties. One of the most limiting factor is the fact that the full resolution of the microscope cannot be realized for three-dimensional thick samples. The reason is that imaging without aberrations (in practice, with aberrations smaller then the diffraction-limit resolution) can only be achieved under well defined conditions. For biological microscopy these are set for samples just under a cover-slide of well defined thickness. As soon as the focus of the objective is moved into the sample depth, the resolution of the optical system degrades.

There are several ways to correct these depth aberrations: the most flexible method is to use objectives with correction collar. However, this imposes severe limitations on the speed of three-dimensional image acquisition. It is the purpose of this project to develop computational alternative methods that will be compatible with modern three-dimensional microscope imaging procedures.

Three-dimensional image deconvolution is a post-acquisition method that uses the known properties of the microscope optics to reconstruct better images. Deconvolution has been used for over two decades with great success for a wide range of applications in astronomy and in microscopy.

Many deconvolution methods have already been proposed for 3D microscopy, such as Agard and Sedat [1, 3], Tikhonov-Miller inverse filter [26], Carrington [24] and Richardson-Lucy (RL) algorithms [18, 22]. The latter has been used extensively in astrophysical or microscopic imaging [26], and is of particular interest for confocal microscopy because it is adapted to Poisson noise. An important drawback of RL deconvolution, however, is that it amplifies noise after a few iterations. This sensitivity to noise can be avoided with the help of regularization constraints, leading to much improved results. Conchello et al. [6] and van Kempen et al. [24, 25] have presented a RL algorithm using energy-based regularization applied to biological images. Conchello's regularization term introduces oscillations enhanced with the number of RL iterations in homogeneous areas. Tikhonov-Miller based term, on the contrary, regularizes too much, resulting in smoothed edges. The previous work of the team financed first by ARC Demitri funded by INRIA from 2002 to 2004 and then by the P2R program for the period 2005-2006 has resulted in a deconvolution method based on the Richarson-Lucy algorithm regularized by the Total Variation [9, 10] in order to reduce the influence of noise on the performances of the RL method used alone and provides smoothing in homogeneous areas while preserving edges.

A second class of restoration methods contains multiresolution models. In particular, wavelet denoising offers an alternative method of regularization for deconvolution. Boutet de Monvel et al. [8] propose a denoising method for confocal image stacks, using Daubechies' wavelets for each

direction, before applying a MAP or a non-regularized Richardson-Lucy method for deconvolution. In [28], Willett et al. use a multiscale approach based on platelets to denoise 2D images in the presence of a Poisson noise. Platelets are localized functions at various scales, locations, and orientations, which produce piecewise linear image approximations. This platelet-based method is very well suited to Poisson noise and preserves edges in the images. In [17], Kervann and Trubuil propose a method to denoise confocal images as a pre-processing before using a deconvolution algorithm based on Richardson-Lucy. The denoising method is based on a locally piecewise constant modelling of the image with an adaptive choice of window around each pixel [21]. They apply the Anscombe transform to transform the Poisson noise into a noise distribution with stabilized variance.

We have recently developped a denoising method based on a 3D Complex Wavelet Transform (CWT) [4]. The CWT was proposed by N. Kingsbury in order to obtain a shift and rotation invariant transform with reduced redundancy (2^d if d is the dimension of the space, $d = 3$ in confocal microscopy application). The denoising results show that this transform is well adapted to biological 3D image restoration compared to real wavelet transforms. The goal now is to propose a restoration method using this Complex wavelet transform and including the deconvolution process.

The team will also investigate two other alternatives that stem from activities in which the Technion group has experience. One is based on an alternative denoising technique commonly referred to as Basis-Pursuit Denoising [5] while the second uses a new iterative-shrinkage method called Parallel Coordinate Descent (PCD) [19].

However, in 3D microscopy most of the deconvolution algorithms fail to function well when the optical characteristics in the working conditions deviate from the assumed model. These characteristics are usually represented by the Point Spread Function (PSF), the image of a sub-resolution point source. For deconvolution methods in microscopic imagery, it is important to know precisely the degradation function (ie. PSF) which caracterizes the microscope in several experimental contexts and which defines the degradations induced by all the elements of the set optical system/sample. Several approaches can be used to obtain this function. One is experimental. The PSF is given by the image acquisition of small (a hundred of nanometers) fluorescent beads. Each microsphere defines a point source and its image gives the PSF of the optical system. However the image of one bead is very noisy and it is recommended to average over several beads. Another approach consists in using a parametric physical model. The PSF we have used in previous work modelizes the diffraction of the light with the default of focus and the effect of the finite size of the pinhole [2, 11].

For high accuracy, simple models of the non aberrated PSF are however not precise enough. Our goal is to devise accurate modelling of PSFs with aberrations and to find accurate approximations with analytical expressions depending only of a small number of parameters. This will be sought for both confocal and wide field PSF models. During the course of the 2005-2006 P2R project, the team has established accurate Gaussian approximation models of non- aberrated PSFs in order to reduce the computational requirements of deconvolution algorithms [29].

The proposed project will develop deconvolution software that will use the modeling of aberrated PSFs to reconstruct the three-dimensional image and the PSF under aberrated conditions. To define the best model according to the degraded image, we will use the approach of estimation theory. Based on the experience acquired in such estimation methods for satellite images [13], we will propose a blind deconvolution algorithm for microscopy images.

The goals of this research are expected to be relatively reachable for confocal microscopy, where the PSF stay rather compact in real space and is only distorted by the aberrations [20]. However, the existence of aberrations causes un-retrievable loss of intensity during the acquisition stage, and therefore the ability to reconstruct the image at the full resolution is limited by signal-to-noise ratio. The situation is different for wide-field microscopy, where aberrations do not cause loss in intensity, but spread the same total intensity over larger volume. In this case reconstruction algorithms based on blind deconvolutions have a chance to reconstruct the full intensity of the image. The difficulty here emerges from the complex behavior of the 3D PSF. We nevertheless have PSF models for wide field microscopy that can cover a wide range of aberrations by a small number of parameters, provided the un-aberrated PSF is known. Given this, the blind deconvolution algorithms will be

extended to wide-field microscopy.

2 Scientific and technological background

The team comprises two groups that are world-wide recognized leaders in specific image processing topics (wavelet, variational and stochastic processes, denoising, deconvolution, super-resolution), and two groups that are working in image processing applications for cell biology of infectious diseases and drug screening in two world-wide recognized biological institutes. Collaborative work performed in the framework of first the ARC Demitri project (2003-2004) funded by INRIA and then of a P2R project during the period 2005-2006 has yielded a 3D modified Richardson-Lucy deconvolution method which takes into account the specificity of the Poisson noise of microscopic images and uses a non-linear regularization technique based on Total Variation (TV) regularisation [10]. The global structure of the project reflects the multi-disciplinarity of the team. This guaranties to foster collaborations and to make the best use of the complementarity of the research teams. Moreover, as some of the project teams are either affiliated to or have active collaborations with major biological institutions, the evaluation of the methodology will be done on real-life applications.

The Quantitative Image Analysis Unit develops image processing methods and programs for the automatic analysis and quantitation of microscopic images. The main research topics include dynamic object segmentation, spot and particle tracking in dynamic microscopy, fluorescence quantification as well as detection methods for temporal sequences.

Ariana team has a long experience in ill-posed inverse problems, using variational or stochastic methods and also wavelet analysis. For satellite image deconvolution, Ariana team has developed in the past restoration methods by using non-linear regularization with maximum likelihood parameter estimation [14], or by using complex wavelet packet decomposition [15]. Ariana team has used the Richardson-Lucy approach with various regularization constraints. This team also recently developed a denoising and deconvolution method using 3D complex wavelet decomposition for biological imagery [4]. The Ariana team has also proposed a blind image deconvolution method for satellite images [13]. This method has been successfully tested by the Space French Agency. A patent deposit was done in 2001 in France and in 2002 in several other countries.

The Weizmann team have recently developed deconvolutions to create synthetic projections from "sweeping focus" acquisition mode, in order to obtain microscope imaging of thick samples in very fast high-throughput cell screening experiments. The method is useful for subcellular localization of fluorescently-labeled molecules in high-magnification microscopy applications for drug development.

The Technion team is specialized in image processing and restoration from uncomplete data, super-resolution and adaptive filtering of multidimensional data.

3 Detailed research program

The aims of this proposal are two-fold:

- First, we want to derive analytical PSFs expressions for modelling the degradation of the images by an aberrated microscope. We also want to find accurate Gaussian approximations that will be used by the deconvolution algorithm developed in the second part of the project.

- Second, we want to derive a blind deconvolution algorithm that will make use of the PSFs models above by estimating the parameters of the approximative PSFs thanks to an information-theoretic approach.

3.1 FR1 Contribution

The PSFs of fluorescence microscopes play a central role in understanding their imaging performances, such as the theoretical resolution limit and the optical sectioning capacity. A great amount of research has focused on deriving more and more accurate PSF models based on wave optics ([12] and cited references). Despite the availability of the rigorous physical models of PSFs, approximative PSFs, particularly separable Gaussian approximations are widely used in practical microscopic data processing. Indeed, compared with a physical PSF model, which usually involves non-trivial terms such as integrals and infinite series, a Gaussian approximative PSF is much simpler and can be computed much faster. Furthermore, due to its special analytical form and good properties, a Gaussian PSF is often preferred to facilitate theoretical analysis and modeling, such as the analysis of convergence properties of EM deconvolution [7].

Despite the popularity of Gaussian approximations, most of the above mentioned works either assume the validity of the approximations or justify them only empirically on observed data. The approximation accuracy and the selection of Gaussian parameters have rarely been rigorously investigated with any physical PSF model, leaving these approximations essentially arbitrary. To the best of our knowledge, only a few works [27, 23] have considered the PSF approximations based on physical models, but they solely covered the paraxial WFFM PSF case with an L^∞ constraint.

In this project, which is a generalization of our previous work [29], we will study comprehensively the least squares Gaussian approximations of the diffraction-limited 2D/3D paraxial/non-paraxial PSFs of confocal and wide-field microscopes in the aberrated case. The PSFs will be described using the scalar or vectorial Debye integral. Numerical simulations will be used to characterize the approximations, before test with real data are used for blind deconvolution in collaboration with the teams FR2, IL1 and IL2. The derivation of Gaussian PSF approximations is part of the work of a Ph.D. student (Bo Zhang) financed by the CNRS and the Institut Pasteur.

3.2 FR2 Contribution

Biological specimens imaged by microscopy are usually rich in information, containing small details and thin structures. Blurred and noisy biological image restoration methods include a regularisation of the solution in order to stabilize the inversion. The regularisation methods must preserve small structures of the images. For that purpose, we propose to develop a regularisation technique using a 3D wavelet transform.

Wavelet denoising offers an alternative method of regularization for deconvolution.cWe have developed a denoising method based on a 3D Complex Wavelet Transform (CWT) [4]. The CWT was proposed by N. Kingsbury in order to obtain a shift and rotation invariant transform with reduced redundancy (2xD if D is the dimension of the space, D=3 in confocal microscopy application). The denoising results show that this transform is well adapted to biological 3D image restoration compared to real wavelet transforms.

This method of denoising could be combined with deblurring according to three schemes:

- The first one is to denoise the image and then apply a deconvolution algorithm. This deconvolution method should take into account the residual noise. Therefore a regularisation must be introduced in the deconvolution process.

- The second one is the opposite. It consists in first deblurring the image and then denoising the deconvolved signal. This technique has been used successfully for satellite image deconvolution [15] but should be used carefully in the context of confocal microscopy because the PSF could exhibit zeros in its spectrum.

- The third one consists in using the proposed denoising technique embedded in an iterative deblurring algorithm. The threshold should be chosen with special attention. Here again, because of zeros in the spectrum of the PSF of confocal microscopy, a regularisation technique could be necessary.

Tests will be conducted on simulated and real data provided by Pasteur and Weizmann and the different schemes will be compared. Another part of Ariana's research work will be done in collaboration with Technion for blind deconvolution. As for the non aberrated case, we will try to recover the PSF (using the model proposed by Pasteur during this proposal) from the observed image itself by using MLE (Maximum Likelihood Estimation) and EM (Expectation Maximisation) methods. Tests will be done on synthetic and real data provided by both Pasteur and Weizmann. The approach of using 3D complex wavelet will be the post-doc work of Caroline Chaux (one year post-doc in 2007 funded by INRIA) whereas the estimation work for blind deconvolution will be conducted by one PhD student (Praveen Pankajakshan) who will start in september 2006 and will be funded during 3 years by INRIA.

3.3 IL1 Contribution

As described by the FR2 group, using wavelet denoising offers an interesting alternative method for regularization for deconvolution. While we plan to take part in investigating this possibility we also plan to investigate two other alternatives as well. These alternatives stem from activities in which the Technion group has experience. One is based on an alternative denoising technique commonly referred to as Basis-Pursuit Denoising [5]. The Basis-Pursuit algorithm has a wide use in the context of sparse representations. As the images commonly observed through biological microscopes are quite sparse, we feel that this approach can be very applicable here. Recently, a new iterative-shrinkage method called Parallel Coordinate Descent (PCD) has been developed at the Technion [19]. An accelerated version of this algorithm was presented in [19]. This algorithm has the potential of being an efficient approach for solving our blind deconvolution problem.

Yet another idea we plan to pursue has to do with a particular modeling of the 3D PSF of the biological microscope. A very common model in communication channels is the multi-path model. A received signal is the result of the transmitted signal passing through a number individual, simple, channels. As a result, many efficient tools have been developed to equalize the effects of this channel on the signal, an action which amounts to deconvolving the received signal. We intend to investigate the possibility of modeling the microscope PSF in its depth dimension (z) as a multi-path of a finite number of depth levels. Once proven successful, a whole arsenal of efficient deconvolution methods can be applied.

Both described approaches will be the graduate work of a new M.Sc. student, Michal Lahav, which will be working on it full time for the proposed two years.

3.4 IL2 Contribution

At Weizmann deconvolutions are routinely applied to widefield microscopy. Within this research program, the Ariana algorithms will be used to deblur cytoskeleton images of both fixed and immunostained data and to live cell expressing fluorescent protein labeled components of the cytoskeleton and other cell organelles. Quantitative analyses of such images based on segmentation of labeled structures are expected to yield cleaner and more precise data needed for studying cell mechanisms and pathways. Deconvolution algorithms for the reconstruction of true 3D projections of thick samples using high-magnification microscopy will be developed and applied for high-throughput applications based on imaging of cells in multiwell microplates. Continuing previous work [16] the Weizmann team will also develop various models for theoretical description of the microscope point-spread function in ideal and in aberrated conditions, and will test these models using fluorescent microspheres and by incorporating them in the Ariana deconvolution algorithms, and comparing blind deconvolution results for the PSF with expected aberration in the real acquisition process (e.g. depth-dependent spherical aberration).

4 Expected scientific, social and economic contribution

Over the past ten years, a considerable part of research efforts in biology have been directed towards integrating large molecular data sets into in vivo systems and relating exhaustive

genetic information to functional analysis. The recent development of new advanced in vivo microscopy techniques together with the possibility of manipulating and tagging proteins with, e.g., GFP-derived probes, have been instrumental in allowing for a shift in basic and applied biological research. As an example, large scale studies allow nowadays for a global and exhaustive documentation of biological processes within the context of living cellular or tissue systems, even though cell functions involve complex networks of many interacting molecules, many not yet identified. The use of the programs developed in the context of this project will enable to dig further into these directions and will be used in collaborating biological institutes to develop and strengthen the following topics:

- The data produced by cell-based assays will be of great interest for basic understanding of complex cell behavior, is expected to have direct implications in diagnostics and treatment of diseases highly relevant for pharmaceutical companies. Not only may the traditional drug discovery process adopt cell-based screening if it is shown fast and effective for sorting potential drug leads before going into slow and expensive animal studies, but avenues for new methods not possible before will also be opened. For example, synergistic drug cocktails are very seldomly designed systematically, although the multiple malfunctions associated with many diseases are well recognized. Sensitive and quantitative cell-based imaging assays bear the potential of becoming a paradigmatic platform for such drug cocktails development with lower concentrations of each compound therefore lower toxicity and side effects, yet with better specificity due to affects of multiple pathways that mediate the targeted malfunction.

- Multidimensional microscopy assays in cell biology. The most general source of visual information decoded in biological system concerns the dynamic topology of a target. Targets can be on the levels of single proteins, protein complexes, organelles or entire integrated systems, starting from viruses, prokaryotic cells, eukaryotic cells, parasites, etc. 3-D image analysis provides the required spatial organization that is complementary to the functional relations established by array technologies. Acquiring 3-D data of cells in vivo over time yields a 4D image sequence, which allows dynamic studies of biological processes.The novel tools developed to analyze 3-D multi-probe signals acquired by high-resolution multidimensional microscopy will permit us to quantitatively describe the organization of labelled probes and proteins in the cell during the cell cycle.

5 Conformity of the project to the priority research fields

The proposed project is submitted within area 3 of the CFP (ie. Novel Fields of Medical and Biological applications). It proposes to use the synergy of two leader groups in mathematical image processing (one located at INRIA and the other at Technion) and two outstanding research groups working in image processing applications for cell biology of infectious disease and drugs screening (one located at Institut Pasteur and the other at Weizmann Institute). The proposed research program will promote new fields of application such as cell based assays for therapeutic research, and multidimensional microscopy assays in cell biology.

6 Available research resources

6.1 FR1: Institut Pasteur research group

6.1.1 Research team members

- **Jean-Christophe Olivo-Marin, Director of research**

  Biography
  Jean-Christophe Olivo-Marin received his Ph.D. in 1989 and the "Habilitation à Diriger des

Recherches" in 1998 both from the Institut d'Optique Théorique et Appliquée, University of Paris-Orsay, France. He is the head of the Quantitative Image Analysis Unit at the Institut Pasteur. He was a co-founder and Chief Technology Officer of the Institut Pasteur Korea, Seoul, from 2004 to May 2005. Previous to that he was a staff scientist from 1990 to 1998 at the European Molecular Biology Laboratory, Heidelberg. He has a long experience of multi-disciplinary approaches in biological imaging, and his research interests are in image analysis of multidimensional microscopy images, pattern recognition and motion analysis for cellular dynamics studies. He has organized several special sessions dedicated to biological image analysis in international conferences (ELMI'02, ELSO'03, ISBI'04, ICASSP'06). He is a member of the Bio Imaging and Signal Processing Technical Committee (BISP-TC) of the IEEE Signal Processing Society, SPIE, the Pattern Recognition Society and member of the Editorial Board of the journal Medical Image Analysis.

Publications

Genovesio, A., Liedl, T., Emiliani, V., Parak, W., Coppey-Moisan, M. and Olivo-Marin, J.-C. (2006) Multiple particle tracking in 3D+t microscopy : method and application to the tracking of endocytozed Quantum Dots, IEEE Trans. Image Processing, 15, 5, pp. 1062-1070

Zimmer, C., Zhang, B., Dufour, A., Thébaud, A., Berlemont, S., Meas-Yedid, V., and Olivo-Marin, J.-C (2006) On the digital trail of mobile cells, IEEE Signal Processing Mag., 23, 3, pp. 54-62

Zimmer, C. and Olivo-Marin, J.-C (2005) Coupled parametric active contours, IEEE Trans. Pattern Analysis and Machine Intelligence, 27, 11, pp. 1838-1842

Olivo-Marin, J.-C. (2002) Extraction of spots in biological images using multiscale products, Pattern Recognition, 35, 9, pp. 1989-1996.

Zimmer, C., Labruyère, E., Meas-Yedid, V., Guillén, N. and Olivo-Marin, J.-C. (2002) Segmentation and tracking of migrating cells in videomicroscopy with parametric active contours : a tool for cell-based drug testing, IEEE Trans. on Medical Imaging, 21, 10, pp. 1212-1221.

Galy, V., Olivo-Marin, J.-C., Scherthan, H., Doyle, V., Rascalou, N.and Nerhbass, U. (2000) Nuclear pore complexes in the organization of silent telomeric chromatin, Nature, 403, pp. 108-112.

- **Christophe Zimmer, Senior Research scientist**

Biography
Christophe Zimmer graduated from Ecole Polytechnique in 1993 and obtained a doctorate in astrophysics and space techniques from Université Paris 7 in 1997. From 1998 to 2000, he worked as Assistant Research Geophysicist under a NASA contract at University of California Los Angeles, on the analysis and modeling of Jupiter's magnetic field data. In 2000, he joined the Quantitative Image Analysis group of Institut Pasteur, where he develops automated methods for extracting biological information from dynamic microscopy data. He is a permanent researcher of Institut Pasteur since 2003.

Publications
Zimmer, C., Zhang, B., Dufour, A., Thébaud, A., Berlemont, S., Meas-Yedid, V., and

Olivo-Marin, J.-C (2006) On the digital trail of mobile cells, IEEE Signal Processing Mag., 23, 3, pp. 54-62

Zimmer, C. and Olivo-Marin, J.-C (2005) Coupled parametric active contours, IEEE Trans. Pattern Analysis and Machine Intelligence, 27, 11, pp. 1838-1842

Dufour, A., Shinin, V., Tajbakhsh, S., Guillén, N., Olivo-Marin, J.-C. and Zimmer, C. (2005) Segmenting and tracking fluorescent cells in 4D microscopy with coupled active surfaces, IEEE Trans.Image Processing, 14, 9, pp. 1396-1410

C. Zimmer, E. Labruyère, V. Meas-Yedid, N. Guillén and J-C. Olivo-Marin. Segmentation and tracking of migrating cells in videomicroscopy with parametric active contours: a tool for cell-based drug testing, IEEE Transactions on Medical Imaging, vol. 21, no. 10, 2002.

M.G. Kivelson, K.K. Khurana, C.T. Russell, M. Volwerk, R.J. Walker, and C. Zimmer, Galileo magnetometer measurements strengthen the case for a subsurface ocean at Europa, Science, 289, 1340-1343, 2000.

- **Bo Zhang, PhD student**

Biography
Bo Zhang received his engineer degree from Ecole Nationale Sup?rieure des T?l?communications (ENST) in 2003 and his Bsc degree from Nanjing University, China, in 2001. After completing a DEA "Mathématiques, Vision, Apprentissage" at the ENS Cachan, he is currently pursuing a PhD in the Quantitative Image Analysis Group at the Institut Pasteur, Paris.

Publications
Zhang, B., Fadili, M.J., and Starck, J.-L. (2006) Multi-scale variance stabilizing transform for multi-dimensional Poisson count image denoising, IEEE Intern. Conf. on Acoustics, Speech, Signal Processing, ICASSP 2006, II, pp. 81-84

Zimmer, C., Zhang, B., Dufour, A., Thébaud, A., Berlemont, S., Meas-Yedid, V., and Olivo-Marin, J.-C (2006) On the digital trail of mobile cells, IEEE Signal Processing Mag., 23, 3, pp. 54-62

Zhang, B., Zerubia, J., and Olivo-Marin, J.-C. (2006) A study of Gaussian approximations of microscopic PSF, SPIE, 3D and Multidimensional Microscopy, San Jose, January 2006, 6090, pp. 6090K:1?6090K:11

Zhang, B., Zimmer, C., and Olivo-Marin, J.-C. (2004) Tracking fluorescent cells with coupled geometric active contours, IEEE Intern. Symp. on Biomedical Imaging, ISBI 2004 , Arlington, April 2004, pp.476-479.

Genovesio, A., Zhang, B. and Olivo-Marin, J.-C. (2004) Interacting multiple model based method to track moving fluorescent biological spots, IEEE Intern. Symp. on Biomedical Imaging, ISBI 2004, Arlington, April 2004, pp. 1239-1242.

6.1.2 Inventory of relevant equipment

- 10 PC workstations
- 1 SGI server
- 1 automated Nikon microscope for transmission imaging
- several software packages for image analysis

6.2 FR2: Ariana project team

6.2.1 Research team members

- **Josiane Zerubia, Director of research, INRIA**

Biography

Josiane Zerubia has been a permanent research scientist at INRIA since 1989, and director of research since July 1995. She was head of the PASTIS remote sensing laboratory (INRIA Sophia-Antipolis) from mid-1995 to 1997. Since January 1998, she has been head of the Ariana research group (INRIA/CNRS/University of Nice), which also works on remote sensing. She has been adjunct professor at Sup'Aero (ENSAE) in Toulouse since 1999. Before that, she was with the Signal and Image Processing Institute of the University of Southern California (USC) in Los-Angeles as a postdoc. She also worked as a researcher for the LASSY (University of Nice/CNRS) from 1984 to 1988 and in the Research Laboratory of Hewlett Packard in France and in Palo-Alto (CA) from 1982 to 1984. She received the MSc degree from the Department of Electrical Engineering at ENSIEG, Grenoble, France in 1981, and the Doctor of Engineering degree, her Ph.D., and her 'Habilitation', in 1986, 1988, and 1994 respectively, all from the University of Nice Sophia-Antipolis, France.

She is a Fellow of the IEEE. She was a member of the IEEE IMDSP Technical Committee (SP Society) from 1997 to 2003; associate editor of IEEE Trans. on IP from 1998 to 2002; guest co-editor of a special issue of IEEE Trans. on PAMI in 2003; and member-at-large of the Board of Governors of the IEEE SP Society from 2002 to 2004. She has been area editor of IEEE Trans. on IP since 2003. She has also been a member of the editorial board of the French Society for Photogrammetry and Remote Sensing (SFPT) since 1998, and of the International Journal of Computer Vision since 2004. She has been a member of the IEEE BISP Technical Committee (SP Society) since 2005.

She was co-chair of two workshops on Energy Minimization Methods in Computer Vision and Pattern Recognition (EMMCVPR'01, Sophia Antipolis, France, and EMMCVPR'03, Lisbon, Portugal); co-chair of a workshop on Image Processing and Related Mathematical Fields (IPRM'02, Moscow, Russia); chair of a workshop on Photogrammetry and Remote Sensing for Urban Areas, Marne La Vallee, France, 2003; and co-chair of the special sessions at IEEE ICASSP 2006 (Toulouse, France).

Her current research interests are in image processing using probabilistic models and variational methods. She also works on parameter estimation and optimization techniques.

Publications

G. Moser, J. Zerubia and S.B. Serpico, "Dictionary-Based Stochastic Expectation-Maximization for SAR Amplitude Probability Density Function Estimation" IEEE Trans. Geoscience and Remote Sensing, 44(1): pages 188-200, January 2006.

N. Dey, L. Blanc-Fraud, C. Zimmer, Z. Kam, P. Roux, J.C. Olivo-Marin and J. Zerubia "Richardson-Lucy Algorithm with Total Variation Regularization for 3D Confocal Microscope Deconvolution" Microscopy Research and Technique, 69: pp. 260-266, 2006.

G. Poggi, G. Scarpa and J. Zerubia "Supervised Segmentation of Remote Sensing Images Based on a Tree-Structure MRF Model" IEEE Trans. Geoscience and Remote Sensing, 43(8): pages 1901-1911, August 2005.

A. Jalobeanu, L. Blanc-Féraud, J. Zerubia. "Satellite image debbluring using complex wavelet packets", Int'l J. Comp. Vis., 3, 51, 2003, p. 205-217.

X. Descombes, J. Zerubia. "Marked Point Processes in Image Analysis", IEEE Signal Processing Magazine, 5, 19, 2002.

- **Laure Blanc-Féraud, Director of research, CNRS**

Biography
Laure Blanc-Féraud received the phD degree in image restoration in 1989 and the "Habilitation à Diriger des Recherches" on inverse problems in image processing in 2000, both from the University of Nice-Sophia Antipolis, France. She is currently director of research at CNRS in Sophia Antipolis. Her research interests are inverse problems in image processing by deterministic approach using calculus of variation and PDEs. She is also interested in stochastic models for parameter estimation and their relationship with the deterministic approach. She is currently working in the Ariana research group (I3S/INRIA) which is focussed on Earth observation. She is vice-director of the I3S laboratory (CNRS and University of Nice-Sophia Antipolis) since 2003 and in the head committee of the french research network on Signal and Image Processing (GDR ISIS).

Publications
G. Aubert, L. Blanc-Féraud, R. March "An approximation of the Mumford-Shah energy by a family of dicrete edge-preserving functionals" Journal of Nonlinear Analysis, Vol. 64, pp. 1908-1930, 2006.

N. Dey, L. Blanc-Fraud, C. Zimmer, Z. Kam, P. Roux, J.C. Olivo-Marin and J. Zerubia "Richardson-Lucy Algorithm with Total Variation Regularization for 3D Confocal Microscope Deconvolution" Microscopy Research and Technique, 69: pp. 260-266, 2006.

J-F Aujol, G. Aubert, L. Blanc-Féraud, A. Chambolle "Image decomposition into a bounded variation component and an oscillating component" Journal of Mathematical Imaging and Vision, Vol. 22(1), janvier 2005.

J. Bect, L. Blanc-Féraud, G. Aubert, A. Chambolle"A l^1-unified variational framework for image restoration", Computer Vision-ECCV 2004, T. Pajdla and J. Matas Ed., Springer LNCS 3024, Vol IV, pp.1-13, 2004.

A. Jalobeanu, L. Blanc-Féraud, J. Zerubia " Satellite image debbluring using complex wavelet packets" , International Journal of Computer Vision, vol.51(3), pp.205-218, 2003.

- **PhD student**: Praveen Pankajakshan hired by INRIA for 3 years (sept. 2006 - August 2009)

- **Post-doc student**: Caroline Chaux hired by INRIA for 1 year (oct. 2006 - sept. 2007)

6.2.2 Inventory of relevant equipment

22 PCs are available as well as 10 laptops.
ENVI software for image processing and IMARIS software for vizualization of 3D biological images are available in Ariana research group.

6.3 IL1: Technion research group

6.3.1 Research team members

- **Arie Feuer, Professor**

Biography
Professor Feuer received his B.Sc. and M.Sc. in Mechanical engineering at the Technion, Haifa, Israel ('67 and '73 resp.) and his Ph.D. from Yale University, CT, in 1978. From 1967 to 1970 he was with Technomatics Inc. working on the design of automatic machines. From 1978 through 1983 he worked for Bell Labs in network performance evaluation. In 1983 he joined the faculty of Electrical Engineering at the Technion where he is currently a professor and head of the Control and Robotics lab. His research interests included adaptive systems (in control and in signal processing) and sampled data systems. Since 1992 he has been intensively involved in research in digital image enhancement, multidimensional sampling and signal representations. Specifically, some of the projects he worked on had to do with improving digital resolution capabilities of various image acquisition devices. These included scanners, digital cameras - still and video, and orthoscopic devices.

Professor Feuer is a Fellow of the IEEE and a council member of IFAC.

Publications
M. Elad and A. Feuer, " Super - Resolution Restoration of Image Sequence: Adaptive Filtering Approach" *IEEE Trans. on Image Processing*, Vol.8, No. 3 (1999), pp 387-395

N. Goldberg, A. Feuer and G.C. Goodwin, " Super - Resolution Reconstruction Using Spatio - Temporal Filtering" *J. of Visual Com. and Image Rep.* Vol. 14 (2003), pp. 508–525

A. Feuer, " On the necessity of Papoulis result formulti-dimensional GSE" *IEEE Signal Processing Letters.* Vol. 11, No. 4 (2004), pp. 420–422

Y. Dorfan, A. Feuer and B. Porat, " Modeling and Identification of LPTV Systems by Wavelets" *Signal Processing* Vol. 84, No. 8 (2004), pp. 1285–1297

A. Feuer, A. Allouche and G.C. Graham, " Motion Aided Sampling and Reconstruction" , *Proceedings of IEE*, (2004)

- **M.Sc. student:** Michal Lahav

6.3.2 Inventory of relevant equipment

Fully equipped for computerized image analysis work.

6.4 IL2: Weizmann Institute research group

6.4.1 Research team members

- **Zvi Kam, Professor**

Biography

Zvi Kam got his D.Sc. in Physics at the Technion, Haifa, and did his postdoctoral research in biophysics at the University of California, San Diego. He is a Professor at the Weizmann Institute of Science since 1981. He is developing methodologies for high-resolution microscopy, and applying light microscopy to cell biological research. His present research involve development and application of cell-based methods for drug development and genomic library screens. He has built probably the only high-magnification automated screening microscope operating today, which acquires 40-80Gbytes of image data each day, and is working on computerized analysis methods to score such experiments according to designed essays.

Publications

Zamir, E., Katz, M, Posan, Y., Erez, N., Yamada, K.M., Katz, B.-Z, Lin, S., Lin, D.C., Bershadsky, A., Kam, Z. and Geiger, B. (2000) Dynamics and Segregation of cell-matrix adhesions in cultured fibroblasts. Nature Cell Biol. 2:191-196.

Kam, Z., Gustafsson, M.G.M., Hanser, B.H. Agard. D.A. and Sedat, J.W. (2001) Computational adaptive optics for live three-dimensional biological imaging. Proceedings of the National Academy of Sciences 98:3790-3795.

Riveline, D., Zamir, E., Balaban, N.Q., Schwarz, U.S., Ishizaki, T., Narumiya, S., Kam, Z., Geiger, B., and Bershadsky, A.D. (2001) Focal contacts as mechanosensors: Externally applied local mechanical force induces growth of focal contacts by an mDial-dependent and ROCK-independent mechanism. J Cell Biol. 153:1175-1185.

Kam, Z. Zamir, E. and Geiger, B. (2001) Probing molecular processes in live cells by quantitative multidimensional microscopy. Trends in Cell Biol. 11:328-333.

Lichtenstein N, Geiger B, Kam Z. (2003). Quantitative analysis of cytoskeletal organization by digital fluorescent microscopy.Cytometry. 54A:8-18.

- **student to be named in future.**

6.4.2 Inventory of relevant equipment

The Weizmann laboratory is fully equipped for cell biological work, has 6 computerized microscope system for fixed samples, live samples and automated screens of cells in multiwell microplates. 20 various workstations (Silicon Graphics, Apple and PC) are available for image visualization, processing and large volume computation. Computer farms and Terabyte RAID disk storage serve these workstations. Multiple software packages are installed, and the major image visualization and interpretation platform used is Priism (www.msg.ucsf.edu/ive), within which a large volume of image processing and quantitative analysis software has been written.

7 Bibliography of relevant litterature

References

[1] D.A. Agard. Optical sectioning microscopy: cellular architecture in three dimensions. *Annu Rev Biophys Bioeng*, 13:191–219, 1984.

[2] D.A. Agard, Y. Hiraoka, P. Shaw, and J.W. Sedat. Fluorescence microscopy in three dimensions. *Methods Cell Biol.*, 30:353–377, 1989.

[3] D.A. Agard and J.W. Sedat. Three-dimensional architecture of a polythene nucleus. *Nature*, 302:676–681, April 1983.

[4] G. Pons Bernad. Deconvolution d'images biologiques 3D en microscopie confocale par transformee en ondelettes complexes 3d. Master report, INRIA, 2004.

[5] S. S. Chen, D. L. Donoho, and M. A. Saunders. Atomic decomposition by basis pursuit. *SIAM Journal of Scientific Computing*, 20:33–61, 1999.

[6] J.-A. Conchello and J.G. McNally. Fast regularization technique for expectation maximization algorithm for optical sectioning microscopy. In *Three-Dimensional and Multidimensional Microscopy: Image Acquisition and Processing III*, pages 199–208, 1996.

[7] J.A. Conchello. Supperresolution and convergence properties of the expectation-maximization algorithm for maximum-likelihood deconvolution of incoherent images. *Journal of Optical Society of America A*, 15(10):2609–2619, March 1998.

[8] J. Boutet de Monvel, S. Le Calvez, and M. Ulfendahl. Image restoration for confocal microscopy: Improving the limits of deconvolution, with application to the visualization of the mammalian hearing organ. *Biophysical Journal*, 80:2455–2470, May 2001.

[9] N. Dey, L. Blanc-Féraud, C. Zimmer, Z. Kam, J-C. Olivo-Marin, and J. Zerubia. A deconvolution method for confocal microscopy with total variation regularization. In *Proceedings of the IEEE International Symposium on Biomedical Imaging: From Nano to Macro (ISBI)*, 2004.

[10] N. Dey, L. Blanc-Féraud, C. Zimmer, P. Roux, Z. Kam, J-C. Olivo-Marin, and J. Zerubia. Richardson-lucy algorithm with total variation regularization for 3d confocal microscope deconvolution. *Microscopy Research and Techniques*, 69:260–266, 2006.

[11] N. Dey, A. Boucher, and M. Thonnat. Image formation model of 3-D translucent object observed in light microscopy. In *Proceedings of ICIP'02*, volume 2, pages 469–472, September 2002.

[12] M. Gu. *Advanced Optical Imaging Theory*. Springer, 2000.

[13] A. Jalobeanu, L. Blanc-Féraud, and J. Zerubia. Estimation of blur and noise parameters in remote sensing. In *Proceedings of ICASSP'2002*, May 2002.

[14] A. Jalobeanu, L. Blanc-Féraud, and J. Zerubia. Hyperparameter estimation for satellite image restoration using a MCMC Maximum Likelihood method. *Pattern Recognition*, 35(2):341–352, 2002.

[15] A. Jalobeanu, L. Blanc-Féraud, and J. Zerubia. Satellite image debbluring using complex wavelet packets. *Int. J. Comput. Vision*, 51(3):205–217, 2003.

[16] Z. Kam, B. Hanser, M.G. Gustafsson, D.A. Agard, and J.W. Sedat. Computational adaptive optics for live three-dimensional biological imaging. *Proceedings of the National Academy of Sciences*, 98:3790–3795, March 2001.

[17] C. Kervrann and A. Trubuil. An adaptive window approach for Poisson noise reduction and structure preserving in confocal microscopy. In *Proceedings of ISBI'2004*, April 2004.

[18] L.B. Lucy. An iterative technique for rectification of observed distributions. *The Astronomical Journal*, 79(6):745–765, 1974.

[19] B. Matalon. M.Sc. thesis, Technion, 2006.

[20] J.B. Pawley. *Handbook of Biological Confocal Microscopy*. Plenum Press, New York, 2nd edition, 1996.

[21] J. Polzehl and V. Spokoiny. Adaptive weigths smoothing with application to image restoration. *Journal of Royal Statistical Society B*, 62(2):335–354, 2000.

[22] W. H. Richardson. Bayesian-based iterative method of image restoration. *Journal of Optical Society of America A*, 62:55–59, 1972.

[23] D. Thomann, D. R. Rines, P. K. Sorger, and G. Danuser. Automatic fluorescent tag detection in 3D with super-resolution: application to the analysis of chromosome movement. *J. Microscopy*, 208(1):49–64, 2002.

[24] G.M.P. van Kempen and L.J. van Vliet. Background estimation in non linear image restoration. *Journal of Optical Society of America A*, 17(3):425–433, March 2000.

[25] G.M.P. van Kempen and L.J. van Vliet. The influence of the regularization parameter and the first estimate on the performance of Tikhonov regularized non-linear image restoration algorithms. *Journal of Microscopy*, 198:63–75, April 2000.

[26] G.M.P. van Kempen, L.J. van Vliet, P.J. Verveer, and H.T.M van der Voort. A quantitative comparison of image restoration methods for confocal microscopy. *Journal of Microscopy*, 185:354–365, March 1997.

[27] L. J. van Vliet. *Grey-Scale Measurements in Multi-Dimensional Digitized Images*. PhD thesis, Delft University, The Netherlands, 1993.

[28] R. Willett and R.D. Nowak. Platelets: a multiscale approach for recovering edges and surfaces in photon-limited medical imaging. *IEEE Trans. on Medical Imaging*, 22(3):332–350, March 2003.

[29] B. Zhang, J. Zerubia, and J-C. Olivo-Marin. A study of gaussian approximations of microscopic psf. In *SPIE, Three-Dimensional and Multidimensional Microscopy: Image Acquisition and Processing III*, pages 6090–1:6090–11, 2006.