[redacted]

Daniele Pirone

Nationality: [redacted] **Date of birth:** [redacted] **Gender:** [redacted]

Phone number: [redacted] **Email address:** [redacted]

Home: [redacted]

SHORT BIO

Something about myself

I was born in Italy on 14th April 1994. I am currently working as Postdoc student within the group of Dr. Pietro Ferraro at the Institute of Applied Sciences and Intelligent Systems (ISASI) of the Italian National Research Council (CNR). I received the bachelor's degree (cum laude) in 2017 in Biomedical Engineering at the University of Naples "Federico II" (UNINA) with a thesis about plasmonics, based on the modeling of light absorption in metallic nanoparticles for thermal ablation of tumors. Then, I received the master's degree (cum laude) in 2019 in Biomedical Engineering at UNINA. At ISASI-CNR, I developed my M.S. thesis and my Ph.D. research activity, that I started in 2020. In 2022, I spent three months of my Ph.D. program in Switzerland at the Optics Laboratory of the École Polytechnique Fédérale de Lausanne (EPFL) within the group of Prof. Demetri Psaltis, who is one of the most relevant researchers in the optics and photonics field. Among the several activities carried out there, I had also the opportunity to present my results at the annual EPFL workshop organized by ZEISS. During 2022, I was president of the OPTICA Student Chapter "PhASER". I took the final Ph.D. exam in January 2023 within the "Information and Communication Technology for Health" course at UNINA (with additional certification of Doctor Europaeus).

Since 2020, results of my research activity were published in 18 papers in peer-reviewed journals (of which, 12 papers as firsth author and 2 papers as corresponding author) and presented in 40 proceedings at 16 international conferences (OPTICA, SPIE, IEEE, EOSAM), including 6 invited talks (see my Google Scholar webpage https://scholar.google.com/citations?user=SUQxkbsAAAAJ&hl=it&oi=ao). I have achieved the most relevant results in the single-cell analysis performed by means of our unique tomographic phase imaging flow cytometry system, reporting them in prestigious international journals (e.g., *Nature Photonics - IF 39.7*, *Light: Science and Applications – IF 20.3*, *Opto-Electronic Advances – IF 14.1*, *Nano Letters - IF 12.3*, *Sensors and Actuators B: Chemical – IF 9.2*, *Lab on a Chip – IF 7.5*, etc.). Moreover, 1 international patent was filed (see: https://patents.google.com/patent/WO2023002355A1/en?oq=WO2023002355A1). Finally, I am carrying out a Reviewer activity for international peer-reviewed journals (OPTICA, SPIE, IEEE, ELSEVIER).

Honours and Awards

Some results about my research activity had a wide appeal in the national and international press (e.g., https://www.greenme.it/salute-e-alimentazione/salute/cancro-misurare-cellule-tumorali-in-3d-tecnica-innovativa-italiana/ and https://phys.org/news/2022-11-door-stain-free-cellular-components.html, https://www.eurekalert.org/news-releases/964316, https://www.dire.it/newsletter/odm/anno/2021/agosto/04/?news=N02, https://s-citizenship.com/biologicamente/esplorare-le-cellule-in-3d-con-il-metaverso/).

In particular, thanks to a publication in the Nature Photonics journal (https://doi.org/10.1038/s41566-022-01096-7), in 2023 I received the Meritorious Citizenship by the Municipality of Baronissi (Salerno, Italy), that is the highest municipal honour, for my contribution to the scientific research. In fact, single-cell analysis provided by fluorescence imaging flow cytometry is fundamental for studying the cell diversity. However, advanced studies are limited by the staining process, which is invasive, expensive, time-consuming, operator-sensitive, and allows only a 2D imaging with low informative content. Therefore, label-free imaging techniques are the key-enabling-technologies to overcome these drawbacks. My research contributed to demonstrate that the new concept of Holographic Tomography in Flow Cytometry mode (HTFC) can be, in the next future, a powerful imaging mode for advanced single-cell analysis with high-throughput. Indeed, in HTFC, the computed tomography (CT) paradigm is adapted in microscopy to reconstruct the volumetric distribution of the refractive indices (RIs) of cells flowing in microfluidics, which is used as the intrinsic endogenous marker. HTFC allows detailed statistical studies about heterogeneous cell populations, which is ideal for example for defining personalized therapies or for searching for rare cells. In this latter case, we set the perspectives for employing HTFC for detecting circulating tumor cells in body fluids, i.e., a label-free liquid biopsy aiming at the early cancer diagnosis. However, to fully exploit the great HTFC potential, it was necessary to fix its main drawback, that is the lack of intracellular specificity due to the missing fluorescence staining. In my interdisciplinary research, a breakthrough approach based on a fully computational method exploiting the RI statistical-volumetric distribution has been demonstrated for the identification of the stain-free nucleus inside the HTFC tomograms of cancer cells. Thanks to its statistical working principle, this method could be extended to segment any intra-cellular organelle, thus filling the specificity gap with fluorescence microscopy by allowing the potential identification of new cell biomarkers and open the route towards the clinical practice.

EDUCATION AND TRAINING

Bachelor's Degree
University of Naples "Federico II" [08/2013 – 26/01/2017]

City: Naples

Country: Italy

Website: http://ingegneria-biomedica.dieti.unina.it/

Field(s) of study: Biomedical Engineering

Final grade: Cum laude – **Level in EQF:** EQF level 6

Thesis: Modeling of Light Absorption in Metallic Nanoparticles for Thermal Ablation of Tumors

I learnt the basics of biomedical engineering by taking exams such as Mathematical Analysis, General Physics, Fundamentals of Information Technology, Electronics Calculators, Thermodynamics and Transport Phenomena, Mathematical Methods for Engineering, Principles of Electrical Engineering, Fundamentals of Chemistry and Biomaterials, Biomedical Applications of Chemical Engineering, Signal Theory, Mechanics of Materials and Structures, Principles of Bioengineering and Biomedical Instrumentation, Biomedical Signal and Data Processing, Electromagnetic Fields.
In my final thesis, I studied the Plasmonics as a phenomenon capable of inducing thermal ablation of tumour cells through the light absorption in gold nanoparticles. This technique consists in delivering a lethal dose of heat to the volume of diseased tissue by laser while preserving the surrounding healthy tissues. This method requires the use of photothermal agents such as gold nanoparticles, because, excited in resonance conditions by electromagnetic radiation, they show a high absorption followed by a strong heat dissipation, therefore resulting in damages to the surrounding cancer cells. These nanoparticles, smaller than 100nm, make the local heating action extremely selective thanks to the passive and active targeting processes that allow for an accumulation limited to the tumour site only. Furthermore, the optical absorption in healthy tissue is minimal since for gold nanoparticles the resonance falls in the near infrared optical window. In this region, in fact, the absorption coefficients of water and hemoglobin are at their lowest level, therefore the light can penetrate deep into the tumour tissues. Hence, this technique is able to provide a minimally invasive alternative in the treatment of tumours if compared to traditional therapies (surgery, radiotherapy and chemotherapy). In particular, I studied the dependence of the plasmon resonance phenomenon on the shape and size of the nanoparticles. To this aim, gold nanoparticles were chemically created in the laboratories of the Institute for Microelectronics and Microsystems (IMM) of the CNR of Naples and were recorded in TEM images. From the TEM images, I modelled the geometry of gold nanoparticles through a CAD program (Gmsh). Then, through numerical codes developed at the University of Naples "Federico II", I modelled the electromagnetic absorption process that takes place when visible light interacts with the simulated nanoparticles. Finally, I compared the theoretical results obtained through the simulations with the experimental measurements. Through this comparison, I demonstrated that electromagnetic models are able to satisfactorily predict the interaction of light with metallic nanomaterials and thus that these models can be used as tools in the nanoparticle engineering process, which consists in controlling their shape and size to optimize the absorption properties and increase the efficiency of thermal ablation therapy.

Master's Degree
University of Naples "Federico II" [02/2017 – 23/10/2019]

City: Naples (NA)

Country: Italy

Website: http://ingegneria-biomedica.dieti.unina.it/

Field(s) of study: Biomedical Engineering

Final grade: Cum laude – **Level in EQF:** EQF level 7

Thesis: Tracking Algorithms for Phase Contrast Tomography

I learnt the advanced notions of biomedical engineering by taking exams such as Electromagnetic Fields in Diagnosis and Therapy, Biomedical Instrumentation, Advanced Instrumentation for Diagnosis and Therapy, Advanced Analysis of Biomedical Signals for Clinical Applications, Analysis of Multimedia Signals, Tomography and Imaging, Biomedical Signal and Image Processing, Health Physics, General Pathophysiology, Applied Biochemistry, Health Systems Management, Clinical Engineering, Health Information Systems, Hospital Systems, Diagnostics for Images and Radiotherapy.
In my final thesis, that I developed at Institute of Applied Sciences and Intelligent Systems (CNR-ISASI) in Pozzuoli (NA), I studied the Phase Contrast Tomography (PCT) as a technique to reconstruct 3D tomograms of single tumour cells. PCT exploits the quantitative phase contrast images of an object acquired from different orientations to provide a complete 3D map of its refractive index (RI). All PCT setups observe the sample along different directions. The methods mostly used to record images to obtain the tomography of microscopic objects were two, i.e. variation of the illumination beam direction with fixed sample and mechanical/optical sample's rotation with fixed illumination beam direction. In both cases, there were a complete knowledge of the lighting angle. However, these techniques had several limitations that reduced the accuracy of the tomographic reconstruction. In the first case, there were a loss of information since the object was not observed from all possible directions (maximum illumination angle tipically less than 150°). In the second case, external manipulation introduced the risk of altering the sample and considerably reduced the throughput of the technique. Therefore, an alternative tomographic approach had recently been proposed based on the use of fixed illumination and the rotation of the sample induced by the hydrodynamic forces of a laminar flow in a microfluidic environment. Each object is recorded by a digital holographic setup while it moves and rotates in a continuous microfluidic flow, so there is no information about its 3D position and orientation. To retrieve this information, required by the tomographic algorithms, it is necessary to use suitable tracking algorithms. Therefore, I validated this new PCT technique, defining a precise and robust strategy of 3D localization of the objects flowing in the microfluidic channel, that exploited the total information within the holographic sequence. Then, I developed a new tracking-based method to recover the unknown rolling angles of the flowing object. I tested these algorithms on a flowing 3D numerical cell phantom. Finally, I combined the opto-microfluidic holographic recording, the 3D holographic

processing and the rolling angles recovery method to reconstruct the 3D RI tomograms of single Circulating Tumour Cells of a breast cancer cell line (MCF-7) and two neuroblastoma lines (SK-N-SH and CHP134).

PhD
University of Naples "Federico II" [11/2019 – 01/2023]

City: Naples (NA)

Country: Italy

Website: https://icth.dieti.unina.it/

Field(s) of study: Information and Communication Technology for Health (ICTH)

Level in EQF: EQF level 8

The goal of my PhD research activity was the development of new diagnostic systems at lab-on-a-chip scale and computational tools for single Circulating Tumor Cells (CTCs) analysis, through the high-throughput tomographic imaging of cells in a microfluidic platform. Phase Contrast Tomography (PCT) is one of the most powerful microscopic imaging tools for single-cell analysis. It exploits the acquisition of quantitative phase contrast maps around the object to provide a 3D tomogram of its refractive index (RI). Traditional methods of PCT are two, i.e variation of the illumination beam with fixed sample and mechanical sample rotation with fixed illumination beam. They show different limits that reduce the accuracy of the tomographic reconstruction. The first one has a low resolution caused by the maximum illumination angle that is typically less than 150°; the second one has a high resolution but the throughput is low and the sample is altered by the external forces. To overcome these drawbacks, recently at ISASI-CNR a new alternative tomographic approach has been proposed. It is based on the use of a digital holographic microscope that records the digital holograms of cells while they are flowing within a microfluidic channel and rotating due to hydrodynamic forces produced by a laminar flow. Therefore, I dealt with the development of algorithms for (i) detection and 3D holographic tracking techniques of CTCs to identify the trajectory of each cell within the microfluidic channel; (ii) the retrieval of the rolling angles of cells exploiting their phase maps signature and the microfluidic channel properties; (iii) appropriate strategies for the fast and accurate tomography to achieve the 3D RI mapping at the single cell level, possibly using high performance computing algorithms. Finally, I identified appropriate label-free biomarkers able to discriminate among CTCs and from White Blood Cells of comparable sizes, thus providing a first proof of concept about the early detection of tumors through the blood collection (i.e., liquid biopsy).
Prof. Angelo Liseno, Prof. Amedeo Capozzoli, Prof. Claudio Curcio (University of Naples “Federico II”), Dr. Pasquale Memmolo and Dr. Pietro Ferraro (Institute of Applied Sciences and Intelligent Systems ISASI-CNR) were my PhD supervisors. My research activity was hosted in the laboratories of ISASI-CNR located in Pozzuoli (NA), where Pietro Ferraro’s group has developed a digital holographic microscope to perform experiments on CTCs along with processing tools needed to recover the tomographic reconstructions.

WORK EXPERIENCE

Research Fellow
National Research Council (CNR) - Institute of Applied Sciences and Intelligent Systems (ISASI) [16/02/2020 – Current]

City: Pozzuoli (NA)

Country: Italy

The research activity I am carrying out at ISASI-CNR in the Dr. Pietro Ferraro's group is framed in the field of label-free computational optical microscopy for biomedical applications (e.g., diagnosis, therapy, biological studies of cell populations) and for the environmental monitoring (e.g., microplastics detection and plant cell studies). It is mainly focused on the implementation of computational methods in label-free optical microscopy for applications in biomedicine (mammalian cell and tissue analysis) and environmental monitoring (plant cell analysis and microplastic detection). The employed imaging techniques belong to the field of 2D and 3D Quantitative Phase Imaging, such as Digital Holography (DH) and Holographic Tomography (HT), both static and in flow cytometry, and Fourier Ptychographic Microscopy (FPM). In particular, I achieved the main optics-related scientific results in the field of Holographic Tomography in Flow Cytometry mode (HTFC). Thanks to my background as biomedical engineer, to my research work at the ISASI-CNR, and my abroad experience at the EPFL, I have developed an interdisciplinary expertise in label-free optical microscopy.
One of the hot topics I am currently working on is label-free liquid biopsy, which is framed within a project PRIN 2017 - MORphological biomarkers For Early diagnosis in Oncology (MORFEO) Prot. 2017N7R2CJ - funded by the Italian Ministry of University & Research. The MORFEO project aims at the development of HTFC as a biomedical tool for recognizing the presence of circulating tumor cells in body fluids, also by exploiting the artificial intelligence. The final goal is the implementation of label-free liquid biopsy for the early cancer diagnosis and the definition of personalized therapies in non-invasive and cost-effective way, with the further possibility to exploit the holographic properties for a lab-on-chip miniaturization into a point-of-care medical device.
Furthermore, I am also working on other projects, like the identification of drug resistance in cancer cells, the study of cell-nanoparticles interactions for drug delivery applications, the detection of microplastics in marine samples, and the tissue analysis through the Fourier Ptychography Microscopy.

Reviewer
OPTICA;SPIE;ELSEVIER;IEEE [12/2019 – Current]

President of OPTICA Student Chapter
[10/2021 – 10/2022]

City: Pozzuoli

Country: Italy

RESEARCH ACTIVITY

Main Topic

Single-cell analysis provided by fluorescence imaging flow cytometry is fundamental for studying the cell diversity. However, advanced studies are limited by the staining process, which is invasive, expensive, time-consuming, operator-sensitive, and allows only a 2D imaging with low informative content. Therefore, label-free imaging techniques are the key-enabling-technologies to overcome these drawbacks. My research contributed to demonstrate that the new concept of HTFC can be, in the next future, a powerful imaging mode for advanced single-cell analysis with high-throughput. Indeed, in HTFC, the computed tomography paradigm is adapted in microscopy to reconstruct the volumetric distribution of the refractive indices (RIs) of cells flowing in microfluidics, which is used as the intrinsic endogenous marker. HTFC allows detailed statistical studies about heterogeneous cell populations, which is ideal for example for defining personalized therapies or for searching for rare cells. Starting from the first proof of concept of HTFC, I tackled many aspects of this tool to fully exploit its theoretical potential and fix its main drawbacks for a practical implementation. From the technological point-of-view, I implemented several computational strategies for reaching the high-throughput property in 3D. Moreover, I have started to fill the main drawback of label-free imaging in HTFC, i.e. the lack of intracellular specificity due to the missing fluorescence staining. Among the others, I developed a breakthrough approach based on a fully computational method exploiting the RI statistical-volumetric distribution for the identification of the stain-free nucleus inside the HTFC tomograms of mammalian single cells. Then, I generalized this algorithm for the segmentation of vacuoles inside budding yeast cells. In this latter activity, the output of the HTFC technique combined to the computational segmentation algorithm, i.e. the segmented 3D tomogram, was given in input for the first time to a virtual reality tool for a fascinating and innovative visualization of the cell from the inside. Finally, I exploited the big HTFC datasets full of informative content about 3D morphological and biophysical biomarkers at the single-cell level for feeding the data hunger typical of Artificial Intelligence, above all for cell classification scopes (for both diagnosis and therapy). But, at the same time, I employed deep learning for greatly speeding up the heavy HTFC numerical processing through a fast, accurate, and lightweight neural network, towards lab-on-chip implementations.

Other Topics

In addition to the flow cytometry mode, I also worked on the HT imaging implemented in static conditions by a non-conventional modality. Indeed, I demonstrated a strategy to reconstruct the 3D RI tomogram directly at the nuclear level within a layer of plant cells, after inducing their rotation by exploiting the natural and reversible phenomenon of dehydration. Such a method could be exploited for the environmental monitoring of the dehydration process within plant cells along with the study of its effects.

Remaining in the field of environmental monitoring, I have trained a machine learning model to detect microplastics among diatoms within the marine environment. At this aim, I used fractal geometry to create a set of complex features able to describe the most distinctive fingerprint hidden in the holographic diffraction patterns recorded through a DH microscope.

Finally, I worked on the tissue analysis through FPM based on its unique feature of imaging samples with simultaneously high resolution and large field-of-view. Furthermore, thanks to FPM, I implemented a strategy for the multi-scale monitoring of the behavior of a cell layer onto a micropatterned substrate according to its fabrication features.

PUBLICATIONS

Conference Proceedings

C1 - Holographic phase imaging to observe intracellular dynamics of plant cells during dehydration

https://doi.org/10.1364/FIO.2020.FTh1A.1

Wang, Z., Bianco, V., **Pirone, D.**, Memmolo, P., Paturzo, M., & Ferraro, P. (2020, September). Holographic phase imaging to observe intracellular dynamics of plant cells during dehydration. In *Frontiers in Optics* (pp. FTh1A-1). Optical Society of America.

C2 - Tracking-based rolling angles recovery method for holographic tomography of flowing cells

https://doi.org/10.1117/12.2592591

Pirone, D.,[P] Memmolo, P., Merola, F., Miccio, L., Mugnano, M., Capozzoli, A., Curcio, C., Liseno, A. & Ferraro, P. (2021, June). Tracking-based rolling angles recovery method for holographic tomography of flowing cells. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials V* (Vol. 11786, p. 117860H). International Society for Optics and Photonics.

C3 - Investigation of plant cells intracellular dynamics by digital holography

https://doi.org/10.1117/12.2592678

Wang, Z., Bianco, V., **Pirone, D.**, Memmolo, P., Villone, M. M., Maffettone, P. L., & Ferraro, P. (2021, June). Investigation of plant cells intracellular dynamics by digital holography. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials V* (Vol. 11786, p. 117860M). International Society for Optics and Photonics.

P - Presenter

BARONISSI, 16/12/2023 [redacted]

Conference Proceedings

C4 - Cytocompatibility and 3D biodistribution with oxidized nanographene assessed by digital holographic microscopy

https://doi.org/10.1117/12.2592731

Mugnano, M., Lama, G. C., Castaldo, R., Merola, F., del Giudice, D., Grilli, S., Gentile, G., Ambrogi, V., Cerruti, P., Memmolo, P., Pagliarulo, V., **Pirone, D.** & Ferraro, P. (2021, June). Cytocompatibility and 3D biodistribution with oxidized nanographene assessed by digital holographic microscopy. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials V* (Vol. 11786, p. 117860N). International Society for Optics and Photonics.

C5 - Holographic fingerprint as a morphological marker to identify micro-plastics

https://doi.org/10.1117/12.2593832

Bianco, V., Memmolo, P., **Pirone, D.**, Carcagnì, P., Merola, F., Miccio, L., Paturzo, M., Distante, C. & Ferraro, P. (2021, June). Holographic fingerprint as a morphological marker to identify micro-plastics. In *Optical Measurement Systems for Industrial Inspection XII* (Vol. 11782, p. 117820J). International Society for Optics and Photonics.

C6 - Tomographic flow cytometry as the key-enabling technology for label-free liquid biopsy

https://doi.org/10.1109/MED51440.2021.9480257

Memmolo, P., **Pirone, D.**, Miccio, L., Cimmino, F., Kurelac, I., Villone, M. M., Bianco, V., Merola, F., Mugnano, M., Capasso, M., Iolascon, A., Maffettone, P.L., & Ferraro, P. (2021, June). Tomographic flow cytometry as the key-enabling technology for label-free liquid biopsy. In *2021 29th Mediterranean Conference on Control and Automation (MED)* (pp. 267-272). IEEE.

C7 - Label-free microfluidic platform for blood analysis based on phase-contrast imaging

https://doi.org/10.1109/MetroAeroSpace51421.2021.9511700

Miccio, L., Cimmino, F., Kurelac, I., Villone, M. M., Bianco, V., Mugnano, M., Merola, F., Memmolo, P., **Pirone, D.**, Capasso, M., Iolascon, A., Maffettone, P.L., & Ferraro, P. (2021, June). Label-free microfluidic platform for blood analysis based on phase-contrast imaging. In *2021 IEEE 8th International Workshop on Metrology for AeroSpace (MetroAeroSpace)* (pp. 464-468). IEEE.

C8 - Holographic tomography for single-cell analysis

https://www.europeanoptics.org/events/eos/eosam2021.html

Pirone, D.,[P] Memmolo, P., Miccio, L., Bianco, V., Mugnano, M., and Ferraro, P., "Holographic tomography for single-cell analysis", *European Optical Society Annual Meeting (EOSAM) 2021*.

C9 - Induced dehydration as a method to enhance phase-contrast observation of plant cells intracellular dynamics

https://doi.org/10.1364/DH.2021.DW4C.1

Wang, Z., **Pirone, D.**, Bianco, V., Memmolo, P., Villone, M. M., Maffettone, P. L., & Ferraro, P. (2021, July). Induced dehydration as a method to enhance phase-contrast observation of plant cells intracellular dynamics. In *Digital Holography and Three-Dimensional Imaging* (pp. DW4C-1). Optical Society of America.

C10 - Raw holograms based machine learning for cancer cells classification in microfluidics

https://doi.org/10.1364/DH.2021.DTh1D.3

Priscoli, M. D., Memmolo, P., Ciaparrone, G., Bianco, V., Merola, F., Miccio, L., Bardozzo, F., **Pirone, D.**, Mugnano, M., Cimmino, F., Capasso, M., Iolascon, A., Ferraro, P. & Tagliaferri, R. (2021, July). Raw holograms based machine learning for cancer cells classification in microfluidics. In *Digital Holography and Three-Dimensional Imaging* (pp. DTh1D-3). Optical Society of America.

C11 - Microalgae as potential bioindicators for heavy metal pollution

https://doi.org/10.1109/MetroSea52177.2021.9611610

Cavalletti, E., Sardo, A., Bianco, V., Miccio, L., **Pirone, D.**, Sirico, D., Valentino, M., Spinelli, M. & Ferraro, P. (2021, October). Microalgae as potential bioindicators for heavy metal pollution. In *2021 International Workshop on Metrology for the Sea; Learning to Measure Sea Health Parameters (MetroSea)* (pp. 449-453). IEEE.

C12 - A fractal analysis of the holographic diffraction patterns for detecting microplastics among diatoms

https://doi.org/10.1109/MetroSea52177.2021.9611578

Pirone, D.,[P] Bianco, V., Memmolo, P., Merola, F., & Ferraro, P. (2021, October). A fractal analysis of the holographic diffraction patterns for detecting microplastics among diatoms. In *2021 International Workshop on Metrology for the Sea; Learning to Measure Sea Health Parameters (MetroSea)* (pp. 173-177). IEEE.

C13 - Holographic tracking and imaging of free-swimming Tetraselmis by off-axis holographic microscopy

https://doi.org/10.1109/MetroSea52177.2021.9611576

P - Presenter

BARONISSI, 16/12/2023 [redacted]

Conference Proceedings

Sirico, D., Cavalletti, E., Miccio, L., Bianco, V., **Pirone, D.**, Memmolo, P., Sardo, A. & Ferraro, P. (2021, October). Holographic tracking and imaging of free-swimming Tetraselmis by off-axis holographic microscopy. In *2021 International Workshop on Metrology for the Sea; Learning to Measure Sea Health Parameters (MetroSea)* (pp. 234-238). IEEE.

C14 - Tomographic phase microscopy at single cell scale without a-priori knowledge of cell orientations: smart strategies for rotation angles recovery

https://doi.org/10.1117/12.2626452

Miccio, L., **Pirone, D.**, Sirico, D., Merola, F., Memmolo, P., Bianco, V., Wang, Z., Behal, J., Del Giudice, D., Mugnano, M., & Ferraro, P. (2022, May). Tomographic phase microscopy at single cell scale without a-priori knowledge of cell orientations: smart strategies for rotation angles recovery. In *Unconventional Optical Imaging III* (Vol. 12136, pp. 211-217). SPIE.

C15 - On the use of multilook Fourier ptychographic microscopy for observing cells and tissues

https://doi.org/10.1117/12.2625364

Bianco, V., Memmolo, P., Běhal, J., **Pirone, D.**, Valentino, M., Mugnano, M., Pagliarulo, V., Miccio, L., & Ferraro, P. (2022, May). On the use of multilook Fourier ptychographic microscopy for observing cells and tissues. In *Unconventional Optical Imaging III* (Vol. 12136, pp. 218-224). SPIE.

C16 - Digital holography in microplastic identification

https://doi.org/10.1117/12.2625368

Bianco, V., Valentino, M., Běhal, J., **Pirone, D.**, Itri, S., Mossotti, R., Dalla Fontana, G., Stella, E., Miccio, L., Memmolo, P., & Ferraro, P. (2022, May). Digital holography in microplastic identification. In *Unconventional Optical Imaging III* (Vol. 12136, pp. 232-240). SPIE.

C17 - Real-time FPM reconstruction and misalignment correction by numerical Multi-Look and GAN

https://doi.org/10.1364/COSI.2022.CTh3C.4

Bianco, V., Priscoli, M. D., Valentino, M., **Pirone, D.**, Behal, J., Zanfardino, G., Memmolo, P., Bardozzo, F., Miccio, L., Ciaparrone, G., Tagliaferri, R., & Ferraro, P. (2022, July). Real-time FPM reconstruction and misalignment correction by numerical Multi-Look and GAN. In *Computational Optical Sensing and Imaging* (pp. CTh3C-4). Optica Publishing Group.

C18 - Deep learning for faster holographic reconstruction processing in microfluidics

https://doi.org/10.1364/DH.2022.Tu4A.7

Pirone, D., Sirico, D. G., Miccio, L., Bianco, V., Mugnano, M., Ferraro, P., & Memmolo, P. (2022, August). Deep learning for faster holographic reconstruction processing in microfluidics. In *Digital Holography and Three-Dimensional Imaging* (pp. Tu4A-7). Optica Publishing Group.

C19 - On the 3D Pose Dynamics of Flowing Cells in Holo-Tomographic Flow Cytometry

https://doi.org/10.1364/DH.2022.M4A.4

Pirone, D.,[P] Sirico, D. G., Miccio, L., Bianco, V., Capozzoli, A., Curcio, C., Liseno, A., Memmolo, P., & Ferraro, P. (2022, August). On the 3D Pose Dynamics of Flowing Cells in Holo-Tomographic Flow Cytometry. In *Digital Holography and Three-Dimensional Imaging* (pp. M4A-4). Optica Publishing Group.

C20 - Intelligent Digital Holographic systems to counteract microplastic pollution in marine waters

https://doi.org/10.1109/MetroSea55331.2022.9950910

Valentino, M., **Pirone, D.**, Béhal, J., Itri, S., Miccio, L., Memmolo, P., Bianco, V., & Ferraro, P. (2022, October). Intelligent Digital Holographic systems to counteract microplastic pollution in marine waters. In *2022 IEEE International Workshop on Metrology for the Sea; Learning to Measure Sea Health Parameters (MetroSea)* (pp. 344-348). IEEE.

C21 - Photonics through biological lenses

https://doi.org/10.1117/12.2651260

Valentino, M., Behal, J., Giugliano, G., **Pirone, D.**, Memmolo, P., Bianco, V., Miccio, L., & Ferraro, P. (2023, March). Photonics through biological lenses. In *Quantum Sensing and Nano Electronics and Photonics XIX* (Vol. 12430, pp. 114-119). SPIE.

C22 - Toward the specificity in QPI 3D tomographic cell flow cytometry holography: recent achievements and perspectives in biomedical sciences

https://doi.org/10.1117/12.2665523

Pirone, D., Lim, J., Merola, F., Miccio, L., Mugnano, M., Bianco, V., Valentino, M., Giugliano, G., Cimmino, F., Visconte, F., Montella, A., Capasso, M., Iolascon, A., Memmolo, P., Psaltis, D., & Ferraro, P. (2023). Toward the specificity in QPI 3D

P - Presenter

tomographic cell flow cytometry holography: recent achievements and perspectives in biomedical sciences. *Quantitative Phase Imaging IX*, *12389*, 1238902.

C23 - Label-free 3D visualization and quantification of endogenous and exogenous intracellular particles in single cells by phase-contrast holographic flow tomography

https://doi.org/10.1117/12.2663945

Pirone, D., Mugnano, M., Sirico, D., Miccio, L., Bianco, V., Merola, F., Del Giudice, D., Lama, G. C., Castaldo, R., Grilli, S., Pasquinelli, G., Valente, S., Lemma, S., Iommarini, L., Kurelac, I., Memmolo, P., & Ferraro, P. (2023, March). Label-free 3D visualization and quantification of endogenous and exogenous intracellular particles in single cells by phase-contrast holographic flow tomography. In *Label-free Biomedical Imaging and Sensing (LBIS) 2023* (Vol. 12391, pp. 63-68). SPIE.

C24 - Tomographic phase microscopy by 3D Zernike polynomials

https://doi.org/10.1364/DH.2023.HTh4B.1

Memmolo, P., **Pirone, D.**, Giugliano, G., Bianco, V., Miccio, L., & Ferraro, P. (2023, August). Tomographic phase microscopy by 3D Zernike polynomials. In *Digital Holography and Three-Dimensional Imaging* (pp. HTh4B-1). Optica Publishing Group.

C25 - Soft-matter based micro-lenses: from liquids to living cells

https://doi.org/10.1364/3D.2023.DW3A.1

Conference Proceedings

Miccio, L., Memmolo, P., Behal, J., **Pirone, D.**, Bianco, V., & Ferraro, P. (2023, August). Soft-matter based micro-lenses: from liquids to living cells. In *3D Image Acquisition and Display: Technology, Perception and Applications* (pp. DW3A-1). Optica Publishing Group.

C26 - Diagnosis of space-induced effects on blood components by label-free optical technique and microfluidics

https://doi.org/10.1109/MetroAeroSpace57412.2023.10189985

Behal, J., Wang, Z., Miccio, L., Mugnano, M., Memmolo, P., **Pirone, D.**, ... & Maffettone, P. (2023, June). Diagnosis of space-induced effects on blood components by label-free optical technique and microfluidics. In *2023 IEEE 10th International Workshop on Metrology for AeroSpace (MetroAeroSpace)* (pp. 157-161). IEEE.

C27 - End-to-end neural network for speeding up the tomographic reconstruction in holographic imaging flow cytometry

https://doi.org/10.1117/12.2674981

Pirone, D.,[P] Sirico, D. G., Miccio, L., Bianco, V., Mugnano, M., Ferraro, P., & Memmolo, P. (2023, August). End-to-end neural network for speeding up the tomographic reconstruction in holographic imaging flow cytometry. In *Multimodal Sensing and Artificial Intelligence: Technologies and Applications III* (Vol. 12621, pp. 201-205). SPIE.

C28 - Holo-tomographic flow cytometry: a new paradigm in diagnostics by high-throughput and stain-free single-cell imaging

https://doi.org/10.1117/12.2675702

Miccio, L., **Pirone, D.**, Behal, J., Giugliano, G., Schiavo, M., Bianco, V., ... & Ferraro, P. (2023, August). Holo-tomographic flow cytometry: a new paradigm in diagnostics by high-throughput and stain-free single-cell imaging. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials VI* (Vol. 12622, pp. 41-43). SPIE.

C29 - Differences between wild type and LSD populations of HeLa and MEF cells through investigation of lysosomal aggregation and morphological variations by digital holography

https://doi.org/10.1117/12.2675432

Giugliano, G., Behal, J., **Pirone, D.**, Schiavo, M., Capuozzo, A., Montefusco, S., ... & Ferraro, P. (2023, August). Differences between wild type and LSD populations of HeLa and MEF cells through investigation of lysosomal aggregation and morphological variations by digital holography. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials VI* (Vol. 12622, pp. 117-120). SPIE.

C30 - Computational nucleus specificity for holographic tomography in flow cytometry

https://doi.org/10.1117/12.2674991

Pirone, D.,[P] Lim, J., Merola, F., Miccio, L., Mugnano, M., Bianco, V., ... & Ferraro, P. (2023, August). Computational nucleus specificity for holographic tomography in flow cytometry. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials VI* (Vol. 12622, pp. 4-9). SPIE.

C31 - Detection of cells with lipid droplets exploiting their biolens' signature in holographic imaging flow cytometry

https://doi.org/10.1117/12.2674987

P - Presenter

BARONISSI, 16/12/2023 [redacted]

Conference Proceedings

Pirone, D.,[P] Sirico, D. G., Mugnano, M., Del Giudice, D., Kurelac, I., Cavina, B., ... & Ferraro, P. (2023, August). Detection of cells with lipid droplets exploiting their biolens' signature in holographic imaging flow cytometry. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials VI* (Vol. 12622, pp. 255-259). SPIE.

C32 - Three-dimensional holographic particle tracking in space-time digital holography

https://doi.org/10.1117/12.2675460

Huang, Z., Wang, Z., **Pirone, D.**, Bianco, V., Memmolo, P., Cao, L., & Ferraro, P. (2023, August). Three-dimensional holographic particle tracking in space-time digital holography. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials VI* (Vol. 12622, pp. 161-166). SPIE.

C33 - Encoding single-cell phase-contrast tomograms by 3D Zernike descriptors

https://doi.org/10.1117/12.2674829

Memmolo, P., **Pirone, D.**, Sirico, D. G., Miccio, L., Bianco, V., Ayoub, A. B., ... & Ferraro, P. (2023, August). Encoding single-cell phase-contrast tomograms by 3D Zernike descriptors. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials VI* (Vol. 12622, pp. 62-64). SPIE.

C34 - Label-free imaging and microfluidics to diagnose space-induced effects on blood components

https://doi.org/10.1117/12.2675596

Miccio, L., Behal, J., **Pirone, D.**, Memmolo, P., Bianco, V., Mugnano, M., ... & Maffettone, P. L. (2023, August). Label-free imaging and microfluidics to diagnose space-induced effects on blood components. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials VI* (Vol. 12622, pp. 105-107). SPIE.

C35 - Quantitative evaluation of nanoparticles internalization in living cells by holographic microscopy

https://doi.org/10.1117/12.2674984

Pirone, D.,[P] Mugnano, M., Memmolo, P., Merola, F., Lama, G. C., Castaldo, R., ... & Ferraro, P. (2023, August). Quantitative evaluation of nanoparticles internalization in living cells by holographic microscopy. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials VI* (Vol. 12622, pp. 252-254). SPIE.

C36 - Deep learning assisted Fourier ptychography for cells and tissue analysis

https://doi.org/10.1117/12.2674881

Bianco, V., Valentino, M., Behal, J., **Pirone, D.**, Bardozzo, F., Memmolo, P., ... & Ferraro, P. (2023, August). Deep learning assisted Fourier ptychography for cells and tissue analysis. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials VI* (Vol. 12622, pp. 65-70). SPIE.

C37 - Phase-contrast tomography of plant cells' nuclei enabled by reversible induced dehydration

https://doi.org/10.1117/12.2675076

Wang, Z., **Pirone, D.**, Bianco, V., Memmolo, P., Villone, M. M., Maffettone, P. L., & Ferraro, P. (2023, August). Phase-contrast tomography of plant cells' nuclei enabled by reversible induced dehydration. In *Optical Methods for Inspection, Characterization, and Imaging of Biomaterials VI* (Vol. 12622, pp. 260-263). SPIE.

C38 - Computational methodologies for reliable tomographic phase imaging flow cytometry

https://doi.org/10.1117/12.2674832

Memmolo, P., **Pirone, D.**, Valentino, M., Giugliano, G., Bianco, V., Miccio, L., & Ferraro, P. (2023, August). Computational methodologies for reliable tomographic phase imaging flow cytometry. In *Modeling Aspects in Optical Metrology IX* (Vol. 12619, pp. 173-176). SPIE.

C39 - On the use of machine learning for microplastic identification from holographic phase-contrast signatures

https://doi.org/10.1117/12.2674886

Valentino, M., Behàl, J., **Pirone, D.**, Memmolo, P., Miccio, L., Bianco, V., & Ferraro, P. (2023, August). On the use of machine learning for microplastic identification from holographic phase-contrast signatures. In *Multimodal Sensing and Artificial Intelligence: Technologies and Applications III* (Vol. 12621, pp. 216-223). SPIE.

C40 - Diatoms as bio-sentinels to probe the dose-dependent impact of copper on aquatic environment: a multi-scale fractal analysis in Fourier Ptychographic Microscopy

https://doi.org/10.1109/MetroSea58055.2023.10317522

Bianco, V., **Pirone, D.**, Miccio, L., Cavalletti, E., Behal, J., Memmolo, P., ... & Ferraro, P. (2023, October). Diatoms as bio-sentinels to probe the dose-dependent impact of copper on aquatic environment: a multi-scale fractal analysis in Fourier Ptychographic

Microscopy. In *2023 IEEE International Workshop on Metrology for the Sea; Learning to Measure Sea Health Parameters (MetroSea)* (pp. 243-248). IEEE.

Journal Papers

J1 - Rolling angles recovery of flowing cells in holographic tomography exploiting the phase similarity

https://doi.org/10.1364/AO.404376

Pirone, D.,[1,*] Memmolo, P., Merola, F., Miccio, L., Mugnano, M., Capozzoli, A., Curcio, C., Liseno, A. & Ferraro, P. (2021). Rolling angle recovery of flowing cells in holographic tomography exploiting the phase similarity. *Applied Optics*, *60*(4), A277-A284.

J2 - Neuroblastoma cells classification through learning approaches by direct analysis of digital holograms

https://doi.org/10.1109/JSTQE.2021.3059532

Dellipriscoli, M., Memmolo, P., Ciaparrone, G., Bianco, V., Merola, F., Miccio, L., Bardozzo, F., **Pirone, D.**, Mugnano, M., Cimmino, F., Capasso, M., Iolascon, A., Ferraro, P. & Tagliaferri, R. (2021). Neuroblastoma cells classification through learning approaches by direct analysis of digital holograms. *IEEE Journal of Selected Topics in Quantum Electronics, 27*(5), 1-9.

J3 - Identification of Microplastics Based on the Fractal Properties of Their Holographic Fingerprint

https://doi.org/10.1021/acsphotonics.1c00591

Bianco, V., **Pirone, D.**,[1] Memmolo, P., Merola, F., & Ferraro, P. (2021). Identification of Microplastics Based on the Fractal Properties of Their Holographic Fingerprint. *ACS Photonics, 8*(7), 2148-2157.

J4 - Three-dimensional quantitative intracellular visualization of graphene oxide nanoparticles by tomographic flow cytometry

https://doi.org/10.1021/acs.nanolett.1c00868

Pirone, D.,[1] Mugnano, M., Memmolo, P., Merola, F., Lama, G. C., Castaldo, R., Miccio, L., Bianco, V., Grilli, S. & Ferraro, P. (2021). Three-dimensional quantitative intracellular visualization of graphene oxide nanoparticles by tomographic flow cytometry. *Na no Letters*, *21*(14), 5958-5966.

J5 - Dehydration of plant cells shoves nuclei rotation allowing for 3D phase-contrast tomography

https://doi.org/10.1038/s41377-021-00626-2

Wang, Z., Bianco, V., **Pirone, D.**, Memmolo, P., Villone, M. M., Maffettone, P. L., & Ferraro, P. (2021). Dehydration of plant cells shoves nuclei rotation allowing for 3D phase-contrast tomography. *Light: Science & Applications*, *10*(1), 1-16.

J6 - Speeding up reconstruction of 3D tomograms in holographic flow cytometry via deep learning

https://doi.org/10.1039/D1LC01087E

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J7 - Deep learning-based, misalignment resilient, real-time Fourier Ptychographic Microscopy reconstruction of biological tissue slides

https://doi.org/10.1109/JSTQE.2022.3154236

Bianco, V., Priscoli, M. D., **Pirone, D.**, Zanfardino, G., Memmolo, P., Bardozzo, F., Miccio, L., Ciaparrone, G., Ferraro, P., & Tagliaferri, R. (2022). Deep learning-based, misalignment resilient, real-time Fourier Ptychographic Microscopy reconstruction of biological tissue slides. *IEEE Journal of Selected Topics in Quantum Electronics*, *28*(4), 1-10.

J8 - Fourier ptychographic microscope allows multi-scale monitoring of cells layout onto micropatterned substrates

https://doi.org/10.1016/j.optlaseng.2022.107103

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J9 - On the hydrodynamic mutual interactions among cells for high-throughput microfluidic holographic cyto-tomography

https://doi.org/10.1016/j.optlaseng.2022.107190

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J10 - Finding intracellular lipid droplets from the single-cell biolens' signature in a holographic flow-cytometry assay

https://doi.org/10.1364/BOE.460204

Pirone, D.,[1,*] Sirico, D. G., Mugnano, M., Del Giudice, D., Kurelac, I., Cavina, B., Memmolo, P., Miccio, L., & Ferraro, P. (2022). Finding intracellular lipid droplets from the single-cell biolens' signature in a holographic flow-cytometry assay. *Biomedical Optics Express*, *13*(11), 5585-5598.

J11 - Stain-free identification of cell nuclei using tomographic phase microscopy in flow cytometry

https://doi.org/10.1038/s41566-022-01096-7

Pirone, D.,[1] Lim, J., Merola, F., Miccio, L., Mugnano, M., Bianco, V., Cimmino, F., Visconte, F., Montella, A., Capasso, M., Iolascon, A., Memmolo, P., Psaltis, D. & Ferraro, P. (2022). Stain-free identification of cell nuclei using tomographic phase microscopy in flow cytometry. *Nature Photonics, 16*, 851–859.

J12 - Loss minimized data reduction in single-cell tomographic phase microscopy using 3D zernike descriptors

https://doi.org/10.34133/icomputing.0010

Memmolo, P., **Pirone, D.**, Sirico, D. G., Miccio, L., Bianco, V., Ayoub, A. B., Psaltis, D., & Ferraro, P. (2023). Loss minimized data reduction in single-cell tomographic phase microscopy using 3D zernike descriptors. *Intelligent Computing*, *2*, 0010.

Journal Papers

J13 - 3D imaging lipidometry in single cell by in-flow holographic tomography

https://doi.org/10.29026/oea.2023.220048

Pirone, D.,[1] Sirico, D., Miccio, L., Bianco, V., Mugnano, M., Del Giudice, D., Pasquinelli, G., Valente, S., Lemma, S., Iommarini. L., Kurelac, I., Memmolo, P. & Ferraro, P. (2023). 3D imaging lipidometry in single cell by in-flow holographic tomography. *Opto Electronic Adv*, *6*, 220048-220041.

J14 - Identification of drug-resistant cancer cells in flow cytometry combining 3D holographic tomography with machine learning

https://doi.org/10.1016/j.snb.2022.132963

Pirone, D.,[1] Xin, L., Bianco, V., Miccio, L., Xiao, W., Che, L., Li, X., Memmolo, P., Pan, F., & Ferraro, P. (2023). Identification of drug-resistant cancer cells in flow cytometry combining 3D holographic tomography with machine learning. *Sensors and Actuators B: Chemical*, *375*, 132963.

J15 - On monocytes and lymphocytes biolens clustering by in flow holographic microscopy

https://doi.org/10.1002/cyto.a.24685

Běhal, J., **Pirone, D.**, Sirico, D., Bianco, V., Mugnano, M., Del Giudice, D., Cavina, B., Kurelac, I., Memmolo, P., Miccio, L., & Ferraro, P. (2023). On monocytes and lymphocytes biolens clustering by in flow holographic microscopy. *Cytometry Part A*, *103*(3), 251-259.

J16 - Label-free liquid biopsy through the identification of tumor cells by machine learning-powered tomographic phase imaging flow cytometry

https://doi.org/10.1038/s41598-023-32110-9

Pirone, D.,[1] Montella, A., Sirico, D. G., Mugnano, M., Villone, M. M., Bianco, V., Miccio, L., Porcelli, A. M., Kurelac, I., Capasso, M., Iolascon, A., Maffettone, P. L., Memmolo, P., & Ferraro, P. (2023). Label-free liquid biopsy through the identification of tumor cells by machine learning-powered tomographic phase imaging flow cytometry. *Scientific Reports*, *13*(1), 6042.

J17 - Phenotyping neuroblastoma cells through intelligent scrutiny of stain-free biomarkers in holographic flow cytometry

https://doi.org/10.1063/5.0159399

Pirone, D.,[1] Montella, A., Sirico, D., Mugnano, M., Del Giudice, D., Kurelac, I., ... & Ferraro, P. (2023). Phenotyping neuroblastoma cells through intelligent scrutiny of stain-free biomarkers in holographic flow cytometry. *APL bioengineering*, *7*(3).

J18 - Label-Free Intracellular Multi-Specificity in Yeast Cells by Phase-Contrast Tomographic Flow Cytometry

https://doi.org/10.1002/smtd.202300447

Bianco, V., D'Agostino, M., **Pirone, D.**, Giugliano, G., Mosca, N., Di Summa, M., ... & Ferraro, P. (2023). Label-Free Intracellular Multi-Specificity in Yeast Cells by Phase-Contrast Tomographic Flow Cytometry. *Small Methods*, 2300447.

Patents

P1 - Computer-implemented method and corresponding apparatus for identifying subcellular structures in the non-chemical staining mode from phase contrast tomography reconstructions in flow cytometry

[2022]

1 - First Author
* - Corresponding Author

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https://patents.google.com/patent/WO2023002355A1/en?oq=WO2023002355A1

Pirone, D., Ferraro, P., Memmolo, P., Miccio, L., Bianco, V., Psaltis, D., Lim, J. *International Patent*

DIGITAL SKILLS

MATLAB&Simulink / Machine Learning / Convolutional Neural Networks / Deep Learning / Basics Of Python / Basics of C++ / Basics of Assembly / Cisco Packet Tracer / Simul8 / Basics of Mathematica / Graphical User Interface / Microsoft Office

LANGUAGE SKILLS

Mother tongue(s): **Italian**

Other language(s):

English

LISTENING A2 **READING** B1 **WRITING** B1

SPOKEN PRODUCTION A2 **SPOKEN INTERACTION** A2

Levels: A1 and A2: Basic user; B1 and B2: Independent user; C1 and C2: Proficient user

DRIVING LICENCE

Driving Licence: B

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