PHYSICIAN SCIENTIST
LA MEDICINA INCONTRA LA RICERCA
promosso da: AIRC Fondazione CARIPLO | con il supporto di: SIEMM Società Italiana di Medicina Molecolare | UNIVERSITÀ DEGLI STUDI DI MILANO

Principal Investigator	Lugli Enrico
Hosting institution	Humanitas Mirasole S.p.A.
Proposal title	Biomarkers of anti-tumor response in solid tumors treated with combination immunotherapy targeting immunosuppression
Keywords	Immunotherapy; Treg cells; Biomarkers; Immunosuppression and/or suppressor cells; Tumor-Infiltrating Lymphocytes (TIL)
PhD project description	Immune checkpoint blockade revolutionized cancer therapy but a number of patients fail to respond because of primary or secondary mechanisms of resistance, in many cases involving a suppressive architecture of the tumor microenvironment. Our lab has previously identified intratumoral and systemic T-cell characteristics associated with improved progression and enhanced response to cancer immunotherapy. Immunosuppressive T cell populations, particularly those with effector characteristics, play a critical role in blocking anti-tumor immune responses and are associated with poor prognosis across multiple solid tumor types. Preclinical data suggest that combining immunosuppressive T-cell depletion with anti-PD-1/PD-L1 immunotherapy enhances anti-tumor activity, yet its immunodynamic effects in humans remain unexplored. At the Humanitas Research Hospital in Milan, we designed a clinical trial targeting immunosuppressive T cell populations in combination with immune checkpoint blockade immunotherapy in metastatic solid tumor patients, with a major focus on translational endpoints and biomarker discovery. Serial multi-tissue biopsies and blood samples will be collected pre- and post-treatment to capture immune changes. Multiomic cellular, molecular and spatial technologies will profile circulating immune subsets, the spatial architecture of tumors and systemic changes to identify biomarkers undergoing modulation with, and predicting response to combination immunotherapy. Following results from this trial, we anticipate to conduct follow-up clinical and laboratory studies where the cohort of most sensitive tumors will be expanded and the combined immunotherapy be optimized for improved efficacy. The successful MD candidate will have clinical expertise in solid tumor oncology and basic laboratory expertise in cellular immunology, molecular biology, biochemistry or related fields. Access to Humanitas facilities (flow cytometry, genomics, histology, microscopy, metabolomics and advanced bioinformatics) will be granted.
Main topics of the lab	Identifying molecular mechanisms of immune dysfunction and immunosuppression in solid tumors
Short description of the lab activity	Over the past decade, my research has focused on characterizing stem-like memory T cells and their pivotal role in cancer immunotherapy. During my postdoctoral training, I co-led the discovery of stem cell

	memory T cells (Tscm), a subset with self-renewing and long-lived properties, which significantly enhance the efficacy of adoptive T cell transfer (ACT) therapies in cancer (Gattinoni*, Lugli* et al., Nat Med, 2011). My lab has since shown that Tscm cells support immune reconstitution post-transplantation (Roberto et al., Blood 2015) and uncovered the molecular cues guiding their differentiation (Kared et al., Nat Commun, 2020). We demonstrated that antioxidant treatment can sustain Tscm features and improve the anti-tumor performance of CAR T cells (Pilipow et al., JCI Insight, 2018). In the tumor context, we were the first to describe a stem-like subset among exhausted PD-1+ T cells (now termed Tpex), which are crucial for the success of immune checkpoint blockade (Brummelman et al., J Exp Med, 2018). Using single-cell and multi-omics approaches, we further redefined human T cell differentiation by identifying two distinct stem-like progenitor populations—Tstem and Tpex—with different functional outcomes (Galletti et al., Nat Immunol, 2020), and proposed a new model of "hybrid" T cell differentiation (Lugli et al., Trends Immunol, 2020). More recently, we found that NaCl supplementation can prevent T cell exhaustion and enhance anti-tumor immunity (Scirgolea et al., Nat Immunol, 2024). Complementing this, our work has explored immunosuppression within the tumor microenvironment. By applying advanced single-cell technologies, we uncovered novel regulatory T cell (Treg) subsets that contribute to immune evasion. Specifically, we showed that IRF4 and MEOX1 regulate highly suppressive effector Tregs in tumors (Alvisi et al., J Clin Invest, 2020; J Hepatol, 2022), and further identified FOXP3– immunosuppressive T cell populations (Bonnal*, Rossetti*, Lugli* et al., Nat Immunol, 2021; Whiteside et al., Sci Immunol, 2023). We continue to investigate the transcriptional regulators shaping these immunosuppressive networks. A third major focus has been the development of cutting-edge single-cell tools for immune profiling. In collaboration with the Humanitas Flow Cytometry Core, we pioneered the implementation of 30-parameter flow cytometry and associated computational workflows (Mazza et al., Cytometry, 2018; Brummelman et al., J Exp Med, 2018). To support the field, we published a step-by-step protocol for panel design (Brummelman et al., Nat Protocols, 2019) and introduced user-friendly computational tools for data analysis (Puccio et al., Nat Commun, 2023).
Main research area	Immunology
Group composition	The laboratory comprises 18 people, including 3 postdocs, 5 PhD students, 1 postgraduate fellow, 4 computational biologists (2 senior, 2 junior), 3 technicians and 2 technologists serving the flow cytometry facility
Institutional page link	https://www.humanitas-research.com/
Lab website link	https://www.humanitas-research.com/groups/enrico-lugli-group/
Social media link	nan
Lab bibliography	IRF4 instructs effector Treg differentiation and immune suppression in human cancer. Alvisi G, Brummelman J, Puccio S, Mazza EMC, Paoluzzi Tomada E, Losurdo A, Zanon V, Peano C, Colombo FS, Scarpa A, Alloisio M, Vasanthakumar A, Roychoudhuri R, Kallikourdis M, Pagani M, Lopci E, Novellis P, Blume J, Kallies A, Veronesi G, Lugli E J CLIN INVEST 2020 Jun; 130: 3137