

Principal Investigator	Mavilio Domenico
Hosting institution	Università degli Studi di Milano
Proposal title	Targeting tumor immune-evasion mechanisms to limit the metastatic progression of human colorectal cancer to liver.
Keywords	Innate immunity; Liver development and/or regeneration; Microenvironment; Colorectal and/or Intestinal ca.; Metastasis
PhD project description	<p>Colorectal cancer (CRC) poses a significant health challenge, with approximately 50% of patients developing colorectal liver metastases (CRLM), a primary cause of CRC-related mortality. Within six months of an initial CRC diagnosis, 25% of patients are found to have CRLM, underscoring the urgency of effective intervention strategies. Moreover, even after initial treatment, a significant percentage of patients experience liver recurrence, further complicating disease management. Current treatments, including curative resection and chemotherapy, offer limited 5-year survival rates of 20-50%. Hence, there is an imperative need to discover new: i) therapeutic approaches; ii) diagnostic markers for detecting liver recurrence; iii) prognostic indicators of treatment outcomes. Our group already identified several immune-mechanisms conferring protective effects against both primary CRC and metastatic liver tumors. Using scRNA-seq technology and multi-parametric spectral flow cytometry, we characterized distinct subsets of unconventional gamma-delta T cells and tumor-reactive cytotoxic Natural Killer cells associated with favorable clinical outcomes. However, the molecular and cellular modalities that either promote or suppress immune recruitment and infiltration in CRLM are unclear. In particular, significant gaps persist in our understanding of how the spatial organization of the CRLM TME influence disease progression and therapeutic responses. This study aims to implement novel therapeutic and prognostic strategies by integrating analyses of immune and tumor cell-state regulators, tumor genetic variations and spatial biology within the CRLM tumor microenvironment (TME). The specific objectives are: a) Spatiomolecular profiling of the CRLM TME; b) Identification of malignant cell states and genetic factors predicting immune susceptibility and evasion; c) Development of predictive models for CRLM outcome and recurrence risk. By combining our expertise in clinical CRLM and immuno-oncology, access to an extensive patient cohort with detailed clinical data, cutting-edge single-cell spatial transcriptomic, computational analysis, and machine learning approach, this project will identify key immunoregulatory mechanisms to develop novel therapies for CRLM.</p>
Main topics of the lab	Impact of Innate immune responses in Cancer and viral infections
Short description of the lab activity	The Unit Of Clinical and Experimental Immunology (UCEI) at University of Milan investigates on different model of human immune-mediated

diseases by undertaking project of Translational Medicine. Translational Medicine and Immunology. In particular, physicians and scientists at UCEI aim to disclose the pathophysiology of innate immune responses in cancer and viral infections within "bed-to-bench side" projects addressing several unmet clinical needs. The feasibility of these projects is sustained by clinical, technical, experimental and administrative facilities available at UCEI. Our projects relies on the collections of human specimens and clinical data through large programs of patient recruitment and longitudinal follow-up. These clinical activities of UCEI are supported by the IRCCS Humanitas Research Hospital (HRH), accredited by the Joint Commission International (JCI) for adherence to strict international best clinical practices. Thanks to an official agreement between the University of Milan and HRH, UCEI has full access to large clinical programs enrolling patients donating their surgical specimens and blood for research purposes. UCEI also run experimental and clinical trial either monitoring patients' follow-ups or testing new drugs. The laboratory of UCEI operates within a cutting-edge research ecosystem designed to optimize experimental workflows and ensure the efficient management of samples, reagents, and instruments. Core facilities and specialized platforms are supervised by highly qualified personnel, with dedicated teams responsible for daily instrument maintenance, certified calibrations, and regular technical audits. Continuous staff training on emerging technologies and regulatory standards ensures the highest levels of performance, safety, and scientific excellence. Our platforms and core facilities are equipped with state-of-the-art technologies for immunological research, including: a) BSL-2/3 laboratories for the safe handling of biological materials, equipped with biosafety cabinets, CO2 incubators, centrifuges, chemical hoods, freezers, liquid nitrogen cell storage systems, and platforms for cell molecular analyses and genetic editing; b) Advanced conventional and spectral flow cytometry platforms, including advanced multiparametric flow cytometry, Hyperion Imaging Mass Cytometry analyzers and cell sorters; c) Molecular biology platforms featuring the 10x Genomics Chromium system for scRNA/TCR-seq and Illumina NGS technologies; d) Spatial transcriptomics platforms, including 10x Genomics Xenium in Situ and Visium Spatial Gene Expression, for high resolution spatially resolved analysis e) Imaging platforms, such as confocal microscopy, live cell imaging, IVIS in vivo imaging, laser micro-dissection; f) A Bioinformatics Unit offering secure big data storage, advanced analysis pipelines, and consultancy services; g) Immunohistochemistry and multiplex protein analysis systems. UCEI is also highly engaged in education programs and courses at University of Milan within the International Medical School of University of Milan. In this regard, UCEI founded UniMiFlow, which is the Flow Cytometry Academy of the University of Milan (<https://flowcytometryacademy.com/en/>). UniMiFlow is a new project that provides training for modern high-dimensional flow cytometry, for its computational analytic approaches. Our long-lasting experience in this potent and versatile technology is applied both in clinical protocols and research projects. UCEI research

	programs are funded by competitive grants from Italian Ministry of Health, Italian Ministry of Research, Fondazione Cariplo, Fondazione Umberto Veronesi, Associazione Italiana per la Ricerca sul Cancro (AIRC) and the European Union.
Main research area	Immunology
Group composition	As Tenured/Principal Investigator, Head of UCEI at HRH and Full Professor of Translational immunology, Prof. Mavilio coordinates a research team composed by 2 physician-scientist, 4 staff scientists (including 1 physician scientist), 1 Laboratory Manager, 2 Technicians, 1 Technologist, 3 Post-Doctoral Fellows, 7 Ph.D. students (including 3 bio-informaticians with expertise in computational biology), 5 master students.
Institutional page link	https://www.unimi.it/it
Lab website link	https://www.labmavilio.it
Social media link	linkedin.com/in/lab-mavilio-74b78a1a3
Lab bibliography	Tumor microenvironment in primary liver tumors: A challenging role of natural killer cells. Polidoro MA, Mikulak J, Cazzetta V, Lleo A, Mavilio D, Torzilli G, Donadon M WORLD J GASTROENTERO 2020 Sep; 26: 4900