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Title of the proposed project:	Single-cell predictors biomarkers of sacituzumab govitecan (SG) or SG + pembrolizumab in muscle-invasive bladder cancer
Short description of the project	<p>Sacituzumab govitecan (SG) is an antibody-drug conjugate (ADC) targeting TROP2 and delivering the topoisomerase-I inhibitor SN-38. SG has demonstrated significant clinical activity in advanced and metastatic urothelial carcinoma and is now being explored in earlier disease settings, including muscle-invasive bladder cancer (MIBC). The SURE-01 and SURE-02 phase II trials, led by Andrea Necchi and colleagues at IRCCS San Raffaele Hospital, investigated perioperative SG-based strategies in cisplatin-ineligible patients with MIBC. SURE-01 evaluates neoadjuvant SG monotherapy prior to radical cystectomy, while SURE-02 investigates the combination of SG with pembrolizumab in a bladder-preserving strategy. Interim analyses from these trials have reported encouraging pathological and clinical complete response rates, supporting the therapeutic potential of anti-TROP2 approaches alone or combined with immune checkpoint inhibition. Importantly, exploratory translational analyses based on bulk transcriptomic profiling have suggested that molecular subtype may influence treatment sensitivity, with luminal tumors appearing particularly responsive to SG ± pembrolizumab. Additional transcriptomic correlates of response and resistance have also begun to emerge, including TOP1 expression (SURE-01)¹ or stromal signature (SURE-02). Despite these advances, the biological determinants of response to SG-based therapy remain incompletely understood. Bulk transcriptomic analyses average signals across all cells within a tumor specimen and therefore cannot distinguish tumor-intrinsic programs from microenvironmental features that may critically influence ADC sensitivity and response to immunotherapy. Moreover, bulk approaches cannot resolve intratumor heterogeneity, which may play a major role in treatment resistance through the coexistence of distinct tumor cell states with variable TROP2 and TOP1 expression, proliferative capacity, DNA damage response programs, or immune interactions. Understanding how tumor cells and the tumor microenvironment jointly shape response to SG and SG + pembrolizumab is therefore essential to optimize patient selection, identify biomarkers of resistance, and rationally design future combination strategies.</p>
Main research area for the project	Cancer biology
5 keywords for the project	Immunotherapy - Antibody/mAb therapy - Bladder tumor - Chemoimmunotherapy - Neoadjuvant therapy

LAB INFO	
Main topic/s of the lab	Single cell transcriptomic analyses
Short description of the lab activity	<p>While molecular profiling has become increasingly integrated into cancer care, it remains largely confined to bulk tumor or liquid biopsy analyses. These approaches, though informative, do not differentiate between tumor-intrinsic features and those originating from the surrounding microenvironment that could strongly influence ADC response and sensitivity to immune checkpoint blockade. In contrast, single-cell technologies offer the capability to dissect tissues at cellular resolution, enabling detailed molecular characterization of both tumor cells and the surrounding microenvironment. We will use a comprehensive tumor profiling solution combining single-nucleus transcriptomics with AI-powered analytical algorithms to uncover cellular and molecular determinants of treatment response from clinical samples, including frozen and FFPE-preserved tissues. In bladder cancer, this technology was previously used to unravel tumor and macrophage cell states associated with response to immune checkpoint inhibitors in a metastatic setting. Ongoing projects involve this technology to identify single-cell biomarkers of response to the anti-NECTIN4 ADC enfortumab vedotin. Importantly, single-cell analysis may substantially extend the findings generated from bulk RNA-seq in the SURE trials by:</p> <ul style="list-style-type: none"> • refining the characterization of luminal cells associated with sensitivity to SG ± pembrolizumab, • assessing heterogeneity of TROP2 and TOP1 expression, and other ADC-related pathways across malignant subclones, • characterizing immune microenvironment states associated with durable response, <p>In this study, leveraging a single-cell platform, we aim to evaluate the clinical utility of single-cell transcriptomics to predict response to SG and SG + pembrolizumab in muscle-invasive bladder cancer patients enrolled in the SURE-01 and SURE-02 clinical trials.</p> <p>Hypothesis & Aims We hypothesize that single-cell profiling can capture the tumor-cell and immune-cell features that determine response and resistance to SG-based therapies in MIBC. Our objectives are: 1. to evaluate the feasibility of generating high-quality single-cell transcriptomic data from FFPE-preserved MIBC samples from the SURE-01 and SURE-02 trials, 2. to identify tumor-intrinsic and microenvironmental determinants of response to SG and SG + pembrolizumab, 3. to characterize intratumor heterogeneity associated with resistance, 4. and to discover predictive biomarkers that could guide future patient stratification and combination therapies.</p> <p>Analysis Plan We will analyze FFPE material from approximately 100 MIBC patients enrolled in the SURE-01 and SURE-02 trials, including patients with favorable and unfavorable clinical outcomes following treatment with SG alone or SG + pembrolizumab. We will generate and analyze</p>

	single-nucleus RNA-seq data from these samples to uncover cellular determinants of therapeutic response and resistance. Integrated analyses with existing bulk transcriptomic and clinical data generated within the SURE trials will further strengthen biological interpretation.
Recent bibliography	Neoadjuvant sacituzumab govitecan plus pembrolizumab, followed by adjuvant pembrolizumab, in patients with muscle-invasive bladder cancer (SURE-02): a single-arm, phase 2 study. LANCET ONCOL 2026 Apr; 27: 442 Neoadjuvant Sacituzumab Govitecan in Patients With Muscle-Invasive Bladder Cancer: Primary Results of the SURE-01 Trial. J CLIN ONCOL 2026 May; : JCO2600142
Group composition	The Group will be composed by the following members: 4 staff oncologists, 3 PhD students (2 PhD students already in staff + one additional PhD student with the present grant) 1 postdoc 2 research nurses
Institutional page link	www.hsr.it