Comprehensive characterization of tumor immune contexture changes induced by cytotoxic agents in lung and rectal cancer

PhD student: Zuzanna Nowicka, MD Promotor: Prof. Wojciech Fendler
Department of Biostatistics and Translational Medicine, Medical University of Lodz, Poland

BACKGROUND

Despite the significant progress in cancer, chemotherapy (CHT) and radiotherapy (RT) remain the most commonly used modalities. Conventionally, the reduction in tumor size in response to CHT/RT has been attributed to the direct killing of rapidly dividing cells. It is now evident that the immune response plays a crucial role in their efficacy (**Figure 1**). Cytotoxic agents and radiotherapy can either enhance or suppress the immune response against cancer. Induced immunogenicity mechanisms include the release of damage-associated molecular patterns, which stimulate antigen processing and exposure, as well as the production of inflammatory mediators by dying cancer cells. On the other hand, these treatment modalities have undeniable immunosuppressive effects by killing immune cells. Understanding these aspects is vital for the informed, knowledge-based design of treatment regimens combining immunotherapy with CHT and RT.

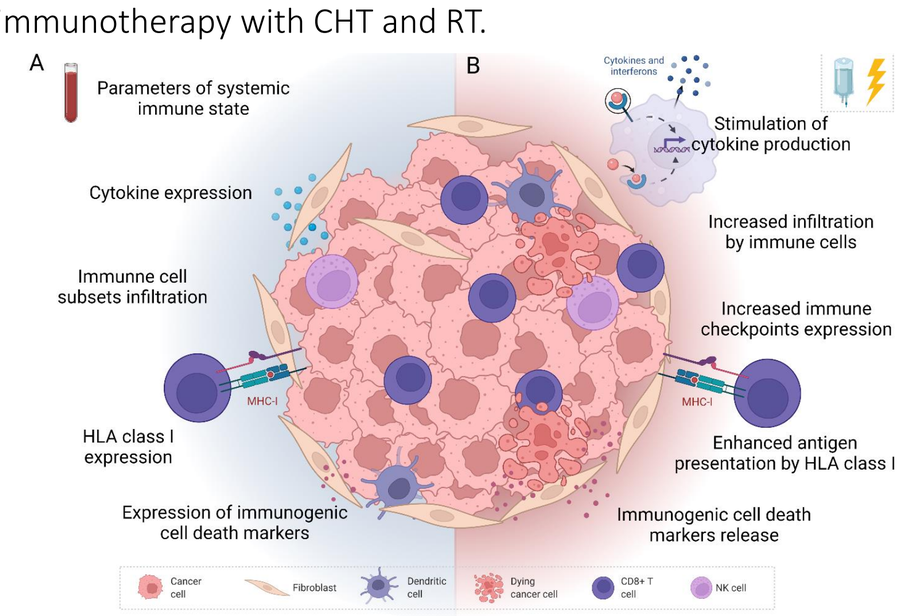

Figure 1. Interactions of the immune system and conventional anti-cancer agents. Known immune parameters that affect CHT and RT efficacy (A) and the effect of CHT and RT on different aspects of the cancer immunity cycle. HLA – human leukocyte antigen.

RESEARCH AIMS

To characterize key aspects of the cancer-immune interaction in non-small-cel lung cancer (NSCLC) tumors resected shortly after therapy with cytotoxic agents. To evaluate lymphocyte infiltration in rectal tumors resected after neoadjuvant chemoradiotherapy (CHRT) with respect to their intrinsic radiosensitivity established *via* gene expression-based indices. We hypothesize that radiotherapy is more immunogenic in tumors characterized by high intrinsic radiosensitivity.

METHODOLOGY AND RESULTS

For the study involving patients NSCLC, retrospective analysis of data from 3 oncology centers is being performed to ensure sufficient number of baseline samples from and samples resected after chemoradiotherapy; approval was obtained from the bioethics committee (RNN/102/21/KE, KE/187/22). For the study involving patients with rectal cancer, bioethics committee approval was obtained (RNN/23/23/KE) and the recruitment is planned to begin in June 2023.

METHODOLOGY AND RESULTS (CONTINUED)

The recrutiment and retrospective analysis of hospital records allowed us to secure 12 non-small-cell lung cancer samples resected between 28-63 days after radiotherapy. Immunohistochemistry (IHC) will be performed with staining for CD4, CD8, FOXP3, PD-1 and PD-L1; furthermore, in the post-RT samples we will perform spatial transcriptomic evaluation with NanoString GeoMx assay to assess the impact of treatment on the immune lung cancer microenvironment in the spatial context. We will use these data to identify infiltrating cell types and their activation states. To gain deeper understanding into the determinants of the immunogenic or immunosuppressive effect of chemo- and radiotherapy, we initiated a project aimed to evaluate whether rectal cancers with higher intrinsic radiosensitivity are characterized by a more „immune-hot" microenvironment following neoadjuvant CHRT (**Figure 2**).

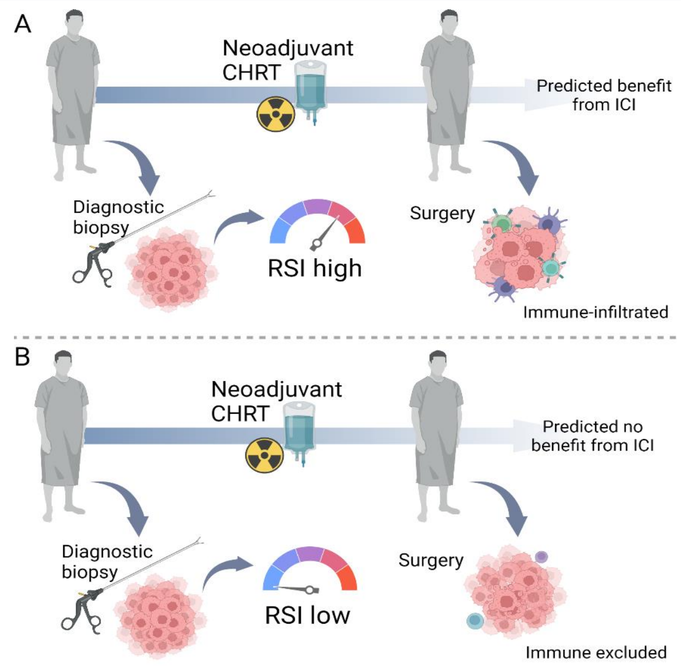

Figure 2. Radiosensitivity index (RSI) as a biomarker in radiosensitive (A) and radioresistant (B) tumours. CHRT – chemoradiotherapy, ICI – immune checkpoint inhibitors, RSI – radiosensitivity index.

The project „Intrinsic tumour radiosensitivity as a marker of chemoradiotherapy-induced immunogenic effects in rectal cancer" is funded from PRELUDIUM grant from the National Science Center awarded in 2023 call to Zuzanna Nowicka MD. To validate radiosensitivity index (RSI) as predictive marker for clinical benefit from CHRT and prospectively evaluate its' association with post-CHRT infiltration by immune cells, two cohorts of patients with rectal cancer will be prospectively recruited: patients treated with neoadjuvant CHRT before undergoing surgery and patients with Stage I-II colorectal cancer treated with surgery alone. Diagnostic tumour biopsies and post-operative material will be collected according to the standard procedures for the management of patients with rectal cancer. The IHC staining will enable the evaluation of the tumour immune environment, by characterizing immune cell subtypes (regulatory FOXP3+ T cells, cytotoxic CD8+ T cells, helper CD4+, CD57+ NK cells), the expression of proteins of the antigen processing and presentation system HLA class I, and of immune checkpoints PD-1 and PD-L1. RNA will also be extracted from pre-treatment samples and RSI will be calculated based on the expression of selected genes.

ACHIEVEMENTS

During this academic year, I participated also in computational oncology workshops in Marseille, France ("Dynamics and Statistics of Cancer Evolution: Applying Mathematics to Experimental and Clinical Data") and in Tampa, Florida (X Integrated Mathematical Oncology workshop "Cancer Communities"; **Figure 3**). As part of this second workshop my team was awarded 1st place for a presentation of mathematical model describing glioblastoma growth and response to treatment and pilot grant funding to facilitate model validation.

Figure 3. Artwork for the X Integrated Mathematical Oncology workshop „Cancer Communities" that took part between 29 October – 4 November in Moffitt Cancer Center, Tampa, Florida.

CONCLUSIONS

We believe that the results achieved during both projects led during this doctoral work will have a significant impact on improving the understanding of the interaction between cytotoxic therapy and the patient's immune system. As a result, we will provide tools to optimize drug combinations and achieve better treatment outcomes for patients with non-small cell lung cancer and rectal cancer.

FUNDING

This research is supported by the National Science Centre and Polish National Agency for Academic Exchange PRELUDIUM BIS Grant no. 2020/39/O/NZ5/01696 awarded to WF and from National Science Centre PRELUDIUM grant no. 2022/45/N/NZ5/01240 awarded to ZN.

Contact information: Zuzanna Nowicka, e-mail: zuzanna.nowicka@umed.lodz.pl