ALMA MATER STUDIORUM
UNIVERSITÀ DI BOLOGNA
DIPARTIMENTO
DI FARMACIA
E BIOTECNOLOGIE

All. 1

Titolo: Dissecting GD2's Role in Neuroblastoma: From Epigenetic Regulation to Next-Generation Therapeutic Nanovectors

Neuroblastoma (NB), the most common extracranial solid tumor in children, remains challenging due to high relapse rates and poor prognosis. GD2, a disialoganglioside selectively expressed on NB cells, has emerged as a critical target for immunotherapy. However, a subset of NBs exhibits low or absent GD2 levels, limiting the effectiveness of immunological treatments. Overcoming this antigen heterogeneity is critical to improve therapeutic responses and reduce disease recurrence.
We propose that epigenetic/transcriptional silencing, particularly of the ST8SIA1 gene responsible for GD2 biosynthesis, is the main cause of reduced expression of GD2 in GD2-negative tumors. By genetically and pharmacologically reversing these genetic/epigenetic mechanisms, it may be possible to restore GD2expression and sensitivity to immunotherapies. This reactivation is expected to enhance antibody dependent cytotoxicity and reduce relapse rate. We also propose a novel therapeutic approach, alternative to immunotherapy, that is based on M13GD2 phage nanovectors targeting GD2 positive cells with high specificity and efficacy.
The proposal is based on three main aims:
1: Identification and characterization of the genetic and epigenetic factors governing GD2 silencing in NB. 2: Comprehension of the role of GD2 expression in NB arising and progression and impact on cancer microenvironment.
3: Development of a novel therapeutic approach to treat GD2-positive NB tumors based on M13GD2 phage nanovectors.
It is anticipated to identify key epigenetic regulators responsible for GD2 silencing, that can be pharmacologically inhibited to allow reactivation of ST8SIA1 and subsequent restoration of GD2 in NB models. Experiments with zebrafish should unveil how GD2 influence cell microenvironment and cancer severity. Moreover, drug-loaded M13 phage nanovectors should demonstrate targeted delivery, high cytotoxicity in GD2positive cells, and reduced off target effects, establishing a novel platform for precision cancer treatment. The impact on cancer of this research proposal is substantial. By uncovering the genetic and epigenetic mechanisms underlying GD2 silencing, this study attempts to develop novel therapeutic interventions that restore or enhance GD2 expression in tumors, currently evading antì-GD2 immunotherapy. Demonstrating that GD2 silencing is a reversible process rather than a fixed resistance mechanism redefines the paradigm of antigen-targeted cancer therapy. Furthermore, the introduction of M13 phage-based nanovectors capable of delivering antineoplastic agents directly to GD2-positive cells represents a highly innovative approach to improve treatment efficacy while reducing systemic toxicity. Finally, the broad expression of GD2 in multiple adult malignancies, suggests that findings from this study could extend well beyond Neuroblastoma.

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PROF. GIOVANNI PERINI
Via Francesco Selmi 3 40126 Bologna, Italia Tel. +39051209416 giovanni.perini@unibo.it

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Executive and Training Plan of fellowship:

Fellows will use advanced genetic and epigenetic analyses to identify critical regulators that silence genes required for GD2 biosynthesis. Those with potentially druggable activities will be tested for responsiveness to specific inhibitors in NB cell lines and patient-derived organoids. Zebrafish KO and xenograft models will help investigate how changes in GD2 expression affect overall tumor-host interactions within the microenvironment and impact on disease severity. Next, M13GD2 phage nanovectors will be developed that can selectively bind GD2-positive Neuroblastoma cells and deliver cytotoxic agents. By optimizing drug payloads, release kinetics, and tumor targeting, we aim to establish a powerful new platform for treating GD2-expressing Neuroblastomas with minimal off-target effects. The activity will be monitored through regular meetings with the research team to track progress, address challenges, and support scientific and professional growth.

PROF. GIOVANNI PERINI
Via Francesco Selmi 3 40126 Bologna, Italia Tel. +39051209416 giovanni.perini@unibo.it