PNRR Missione 4, Componente 2, Investimento 1.4 "Potenziamento strutture di ricerca e creazione di "campioni nazionali di R&S" su alcune Key Enabling Technologies"
Iniziativa finanziata dall'Unione europea –- NextGenerationEU.
National Center for Gene Therapy and Drugs based on RNA Technology
Sviluppo di terapia genica e farmaci con tecnologia a RNA
Codice progetto MUR: **CN00000041** – CUP UNINA: **E63C22000940007**

SPOKE 8: Platform for DNA-RNA delivery

Development of chicken ChorioAllantoicMembrane (CAM)-based cancer models to screen trafficking, delivery efficiency, and toxicity of RNA based nanoplatforms.

The purpose of this research project is to evaluate the safety and efficacy of therapeutic RNAs, appropriately delivered into nanocarriers, in cancer cell lines that are sensitive or resistant to common chemotherapeutic drugs. The main goal is to develop a human CAM-based tumor model as a preclinical screening platform for nano-formulated RNA. In particular, we plan to: i) generate cancer cell lines stably expressing reporter genes such as GFP, luciferase, or GAPDH (2D and 3D/spheroids); ii) set up the optimized procedure for the CAM model. We plan to use as a model a colon cancer cells lacking p53 already present in our laboratory, HCT $116^{p53-/-}$ cells, and uL3ΔHCT $116^{p53-/-}$ cells, derived from the HCT $116^{p53-/-}$ cell line and stably silenced for the ribosomal protein uL3; the absence of uL3 resulted in chemoresistance to the common chemotherapeutic drugs. GFP based measurement serves as a quantitative read-out for target cell viability. Next, to develop three-dimensional (3D) cultures that more closely reflect the pathophysiology of tumors *in vivo*, we plan to generate spheroids from HCT $116^{p53-/-}$ cells, and uL3ΔHCT $116^{p53-/-}$ cells.
These cancer cell lines (2D and 3D/spheroids) will be utilized to evaluate the safety and efficacy of therapeutic RNAs when appropriately delivered into nanocarriers in a CAM-based model. The immunodeficient CAM model can efficiently (in 5-10 days) test multiple targeted therapies on different cancer cell lines and also on fragments derived from the same tumor; enabling imaging to monitor human tumor growth rate and observation of tumor/host vascular counterparts. The CAM assay will also be used to study angiogenesis and assess invasion and metastasis.

References

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