**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**

**Doctorate of National Interest**

**RNA THERAPEUTICS AND GENE THERAPY**

**SELECT ONE OF THE FOLLOWING RESEARCH AREA:**

☐ **Mechanisms of Diseases and Drug Target Identification**

☐ **Design and Delivery of New Gene Therapy and RNA-Based Medicines**

✠ **Validation and Safety In Preclinical and Clinical Studies**

**LOCATION OF THE RESEARCH ACTIVITY (INSTITUTION/DEPARTMENT):**

DEPARTMENT OF DRUG SCIENCES, UNIVERSITY OF PAVIA

**TUTOR:** CRISTINA TRAVELLI

**PROPOSED RESEARCH ACTIVITIES (max 300 words)**

The research aims to screen the potential immunotoxicological properties of nucleic acids-loaded nanoparticles *in vitro* and *in vivo*. Nanoparticles present peculiar properties impacting on their immunological behaviour. These can be due not only to the features of the RNA cargo, but also to the composition of the carrier materials and their physico-chemical properties. These factors could contribute to the induction of organ/tissue and cell-specific toxicological and immune reactions. This project will be focused on the characterization of the organ/tissue-specific immunotoxicological profile of nanoparticles *in vitro* and *in vivo*, in rodent models (assessed in the CNS and in the GI track as model systems). The research activities will include:
- gastrointestinal toxicity, evaluated by IHC and flow cytometry analysis. The following morphologic criteria will be considered: score 0, no damage; score 1, focal epithelial edema and necrosis; score 2, diffuse swelling and necrosis of the villi; score 3, presence of neutrophil infiltrate in the submucosa; score 4, necrosis with neutrophil infiltrate; score 5, massive neutrophil infiltrate and hemorrhage.
- neurotoxicity, for CNS-specific immuno-toxicological analyses. Brain tissue will be dissected and longitudinally split in two halves: one will be homogenized and stained for flow cytometric analysis to assess the

immunophenotypic profile of CNS-resident as well as CNS-infiltrating immune system cells; the other half will be fixed with formalin or PFA to allow histological and immunohistochemical investigation of markers of neurodegeneration, glymphatic system functionality, as well as neuroinflammation. In parallel, CSF and blood (serum/plasma) will be collected to allow the investigation of CNS-specific biomarkers (indicative of ongoing neurodegenerative/neuroinflammatory processes) as a non-invasive readout of the toxicity induced by exposure of the CNS to the NPs (in a direct as well as indirect manner).

- ex vivo evaluations: haemotoxicological evaluation using acute phase markers haematological phenotypes, blood cytokine and platelet aggregation capacity