SUMMARY

This dissertation presents an integrative omics and machine learning (ML) approach to the mechanistic understanding of carbon nanotube (CNT)-induced pulmonary toxicity. Considering the increasing production and diverse applications of single-walled and multi-walled CNTs (S- and MWCNTs), evaluating their safety, especially following inhalation exposure, has become a regulatory and scientific priority. To address this gap, the research bridges cheminformatics and transcriptomics and Adverse Outcome Pathway (AOP)-informed modeling to develop a predictive Nano-Quantitative-Structure-Activity-Relationship (Nano-QSAR) framework aligned with the principles of New Approach Methodologies (NAMs) and Next-Generation Risk Assessment (NGRA).

The central research question underpinning this work is**:** ***How can transcriptomics and machine learning be integrated to develop predictive models for CNT-induced acute phase-driven inflammation following inhalation exposure?*** The core hypothesis posits that CNTs with similar physicochemical characteristics and transcriptomic signatures are likely to induce comparable adverse effects in lung tissue. To investigate this, the study focused on the *acute phase response (AR) signaling pathway*, a well-established early marker of pulmonary inflammation and fibrosis.

To validate this hypothesis, the research pursued three primary objectives. First, an AOP-informed Nano-QSAR model was developed to quantitatively link the physicochemical properties of multi-walled CNTs (MWCNTs) with transcriptional perturbations in the AR pathway. Second, a comparative analysis of single-walled and multi-walled CNTs (SWCNTs and MWCNTs) was conducted to identify shared and distinct structural features that impact on acute phase response pathway. Third, the global Nano-QSAR model was developed to assess how residual metal impurities (such as Fe_2O_3 and CoO) contribute to early transcriptomic changes associated with lung inflammation and fibrosis.

These models successfully captured key molecular drivers and early key events (KEs), demonstrating strong predictive performance consistent with the OECD principles for QSAR validation. Key physicochemical determinants, including aspect

ratio, surface area/functionalization, and specific metal impurities, were identified as critical contributors of inflammatory and fibrotic responses.

A key innovation of this dissertation lies in the systematic integration of transcriptomic data into the Nano-QSAR modeling framework. Unlike traditional QSAR models that often rely on apical endpoints (e.g., histopathology or organ-level toxicity), this approach incorporates genome-wide gene expression profiles to capture early molecular and cellular responses within the acute phase pathway. This molecular-level integration enhances the biological interpretability and improves predictivity, enabling a more precise identification of pathways perturbed by specific physicochemical properties of CNTs. By anchoring transcriptomic responses to structural attributes, the strategy supports more nuanced and mechanistically grounded toxicity predictions. Moreover, it aligns with current regulatory trends emphazing mechanism-based hazard assessment and facilitates the development of safer-by-design (SbD) nanomaterials within the AOP and NAMs frameworks.

In summary, this dissertation establishes a comprehensive and innovative framework for the mechanistically informed prediction of CNT-induced toxicity. It demonstrates the value of combining cheminformatics, machine learning, and systems biology to advance regulatory toxicology and to support the ethical, efficient, and sustainable development of nanotechnology.