Summary of the Doctoral Thesis

Title: Differentiated Thyroid Carcinoma of Follicular Cell Origin: Molecular Biomarkers and Therapeutic Strategies

PhD Candidate: Tiucă Robert-Aurelian

PhD Supervisor: Prof. Univ. Dr. Pașcanu Ionela Maria

Differentiated thyroid carcinoma (DTC) of follicular cell origin is the most common malignant endocrine neoplasm, with a globally increasing prevalence over the past decades. This pathology dominates endocrine oncology, requiring a multidisciplinary approach and long-term monitoring.

Within this category, the main histological types include papillary thyroid carcinoma, follicular thyroid carcinoma, and oncocytic thyroid carcinoma. Among these, papillary thyroid carcinoma is the most frequently encountered. Given their origin in the thyroid follicular cell, these types of thyroid cancer share many functional characteristics with normal thyroid tissue, often displaying an indolent course and favorable response to specific therapies, such as radioactive iodine treatment. However, there are also forms of DTC with unfavorable clinical progression due to resistance to initial therapy or dedifferentiation processes, which can lead to the development of distant metastases.

The etiopathogenesis of DTC is complex, being influenced by both environmental and genetic factors. In recent years, international research has focused on identifying molecular biomarkers with diagnostic and prognostic value, as well as on developing personalized strategies for managing patients diagnosed with this condition. The ultimate goal of these studies is to optimize treatment efficacy, minimize adverse effects, and improve both survival rates and quality of life for patients.

The doctoral thesis presents a personal contribution structured in three studies. The main objective was, on the one hand, to investigate the effectiveness of radioactive iodine treatment in patients diagnosed with DTC, and on the other hand, to evaluate the prognostic role of the rs1799983 and rs2070744 polymorphisms of the nitric oxide synthase 3 (*NOS3*) gene in this pathology. In this context, three research directions were defined, aimed at investigating the aforementioned major objective.

The first study was a case-control study aiming to evaluate the therapeutic response in patients diagnosed with DTC by comparing the evolution of those who received radioactive iodine with those who did not. The study included 76 subjects: 58 patients diagnosed with DTC who received radioactive iodine therapy (case group) and 18 DTC patients who did not receive such treatment (control group). The results showed that DTC patients treated with radioactive iodine had lower rates of incomplete biochemical response compared to those who did not receive this treatment ($p = 0.019$). Additionally, it was found that radioactive iodine treatment could be effective in reducing the risk of incomplete biochemical response, even in cases with indolent histology ($p = 0.030$). Furthermore, the study showed that male sex ($p = 0.008$) and incomplete structural response ($p = 0.002$) were significant predictive factors for incomplete biochemical response, regardless of the histological type analyzed. This study represents an important contribution to the limited number of national and international studies that have evaluated the efficacy of radioactive iodine therapy in Eastern European populations, especially the Romanian population.

The second study aimed to evaluate the potential role of the rs1799983 and rs2070744 polymorphisms of the *NOS3* gene in the clinical evolution of DTC. This case-control study included 172 subjects: 88 diagnosed with DTC (case group) and 84 healthy individuals (control group). For the genetic analysis, a 4 mL venous blood sample was collected from each participant after obtaining informed consent. DNA was isolated using the PureLink Genomic DNA kit, and genotyping of NOS3 polymorphisms was performed using TaqMan assays on a real-time PCR system. The results showed that the analyzed polymorphisms were not significantly associated with the risk of developing DTC, incomplete biochemical control, or the presence of metastases. This is the first study of its kind, both nationally and internationally, to investigate the association of these polymorphisms with DTC. Although no statistically significant associations were found, the results contribute to shaping the view that these genetic variations have a limited role as individual predictive factors in DTC and highlight the importance of a multifactorial genetic approach in the study of tumor evolution.

The third study extended the genetic analysis from the previous study, investigating the associations between the rs1799983 and rs2070744 polymorphisms of the *NOS3* gene and the clinicopathological characteristics of patients diagnosed with DTC. This study was also a case-control study and included 134 subjects: 66 diagnosed with DTC (case group) and 68 healthy individuals (control group). For genetic analysis, a 4 mL venous blood sample was collected from each participant after signing an informed consent form. DNA was extracted using the PureLink Genomic DNA Kit, and NOS3 polymorphisms were genotyped using TaqMan assays on a real-time PCR system. The results revealed that the rs1799983 polymorphism was significantly associated with tumor size ($p = 0.004$), suggesting a possible role of this genetic variation in influencing tumor aggressiveness. Additionally, the CT genotype heterozygous variant of rs2070744 was found to be a significant protective factor for structural control ($p = 0.032$). Like the previous study, this is the first in the literature to analyze these genetic variations in relation to the clinicopathological features of patients diagnosed with DTC, providing a solid foundation for future validation and in-depth research.

In conclusion, the doctoral thesis brings relevant contributions to the approach of DTC by analyzing therapeutic and genetic factors involved in disease progression and by opening new research directions in the field of thyroid oncology.