# Abstract

The scientific activity, professional and academic work presented in this thesis was performed in field of **Immunology** starting with the PhD thesis (1996) until 2013. The habilitation thesis entitled ***Harnessing the immune system - new insights in diagnosis and therapeutical approaches of major diseases*** is based on several original studies carried out over the last 17 years, published in ISI journals, other data base indexed journals and books, and were financed by international and national research projects led by me these years.

The main body of the habilitation thesis is represented by Part I *Scientific and Professional Achievements* that describes the main scientific contributions; this part being organized according to the defined fields.

The section dedicated to *in vitro* and *in vivo* modulation, performed by immune peptides, presents results demonstrating that, administration of peptides acting as MHC class II antagonists can inhibit the induction of T cell-dependent primary and secondary antibody responses. Inhibition of *in vivo* antibody responses is associated to prevention of complex formation between antigenic peptides and class II molecules indicating MHC blockade as the mechanism hindering T helper cell activation. Therefore, MHC class II antagonists may induce selective immunosuppression in autoimmune diseases, including diseases like myasthenia gravis, where autoantibodies have a direct pathogenic role.

Regarding our results focusing on another peptide, Prothymosin-alpha, results have been published regarding its immunologically active fragment, located at the C-terminus of the polypeptide [proTα(100–109)], sequence that can increase *in vitro* T cell proliferation and enhance NK- and LAK-cell cytotoxicity in cancer patients. Moreover besides the reported immunoregulatory activity of proTα(100–109) and entire proTα on cells of the adaptive arm of immunity, data are presented supporting an additional effect of proTα and its immunoreactive fragment proTα(100–109) in augmenting some of the functions of neutrophils in cancer patients.

On-going studies will develop the potential of the proTα(100–109) decapeptide as a promising candidate adjuvant molecule that could be incorporated in cancer immunotherapeutic protocols.

The section focusing on the study of respiratory burst presents the published results regarding mechanisms of respiratory burst generation upon chemotactic peptides stimulation *via* N-formylated peptides and CR3 receptors, characteristics of the inflammatory site. We have shown that the chemotactic peptide sustains the respiratory burst, mainly its early phase, the chemotactic factors exerting a modulatory action on the microbicidal oxygen-dependent functions of PMNs. Short chains of polysaccharide, like low molecular weight heparins (LMWH), used as drugs in cardiovascular diseases, can influence the overall activity of PMNs from patients. Besides the known pharmacological action of LMWH the results shown in this section show that there is a reduction of superoxide anion release by activated PMNs upon LMWHs, decreasing thus the destructive oxidative stress triggered by immune complexes, reducing tissue damage, ischemia and reperfusion injury. Our results show that LMWHs can have additional effects independent of their anticoagulant activity, these effects influencing the “inflammatory component" of the atherosclerotic process.

Part I *Scientific and Professional Achievements* continues with a section that shows the results obtained regarding immune parameters investigated in healthy ageing and in major diseases like cardiovascular diseases and skin cancer.

We have shown that investigating the cellular immune parameters of unstable angina patients, the alteration pattern can announce the development of an acute coronary syndrome. The unstable angina presents alteration of some cellular immune parameters that indicate an inflammatory syndrome associated with an increased risk of coronary heart disease, having also a prediction value for the plaque instability. Although the clinical symptoms and ECG characteristics of the angina don't point out an unstable stage, the immune investigated parameters announce the development of an unstable event. In these patients plasma level of platelet-activating factor acetylhydrolase is increased in correlation with PMN’s activity reflecting the atherothrombotic process. We can ascertain that the immunological parameters are useful to further stratify cardiovascular risk and that therapy should be manipulated accordingly with the immunological status of the patient.

Evaluating normal ageing process through the immunological point of view, results are presented showing that during the physiological ageing, individuals are subjected to multiple inflammatory processes that can direct the percentages of neutrophils toward increased values needed to compensate an individual cell decreased oxidative activity. It is possible that the registered susceptibility to infections of aged individuals, commonly agreed upon, to be the consequence of the absolute number of neutrophils that can respond to the stimuli and to the actual individual capacity of oxidative activity. Regarding adaptive

immunity in ageing, in the over 70 years group, total CD3+ and its subpopulations, CD8+ and CD4+ are altered, thus the CD4+/CD8+ ratio statistically raises with ageing.

The physiological process of ageing is characterized by a series of discrete alteration in both innate and adaptive immunity that can participate to the increased susceptibility of aged individuals to diseases like infections, cancer and other major diseases.

The original section of results continues with an extended part showing the most recent studies regarding immune parameters in skin cancer. We are presenting results showing tumor infiltrating immune cell populations, emphasizing local immune-suppression, generation of systemic anti-tumoral responses to tumor antigens high lightening immune-biomarkers in circulation (molecules and cells), and systemic immune-suppression. All these immune features are represented in high-risk group for developing cancer. Being the largest immune organ, skin ***covers*** the largest aggression surface and must cope with the most complex array of immune mechanisms keeping a distinct equilibrium between external aggressors and any internal alteration of self.

Scrutinizing the immune system and the immune responses developed in skin cancer, we have developed an experimental therapy in cutaneous melanoma animal model using whole body irradiation with microwave in an original equipment, therapy that increases melanoma sensitivity to dacarbazine. Moreover we have studied the dynamics of immune parameters during melanoma development and the possible immune markers that can indicate a good therapeutical behaviour. Combined therapy (low doses dacarbazine + microwave irradiation) had the best clinical evolution of the experimental cutaneous melanoma and these findings encourage us to state that the used microwave therapy can increase the therapeutical effect of dacarbazine.

In melanoma patients investigated by us we have found particular immune parameters, circulatory cells and immune molecules that can predict the metastatic process. Moreover using mass spectrometry technology we have pin-pointed immune-related molecules that can have predictive power in these patients.

Due to the complexity of interactions between skin tumors and the immune system, mechanisms that suppress the antitumor responses and the discovery of predictive biomarkers have been particularly challenging. The specific role of immune cell populations in melanoma and in non-melanoma skin cancers continues to be explored *in search of new biomarkers* and *new immune networks*. We are confident that correlations between clinical, immunological and immunohistochemical data can be useful in the disease management and personalized immune-therapy.

The original results presented herein are based on papers, reviews and chapters in international books and were financed through national and international research projects as stated in detail for each section. The thesis continues with Part II *Plans for Advancement and Career Development* that is structured in future research and teaching development plans and ends with Part III comprising 480 references.