Immune cells shape the outcome of various anticancer therapies. Pre-existing immunity against tumor cells is the factor that influences the response to chemo-and immunotherapy. It is now believed that antitumor immunity is activated strongly or is expanded from pre-existing immunity during the initial cycle of chemotherapy. During the subsequent cycles, together with incoming acquired drug resistance of the tumor cells, loss of the so-called immune participation strongly impairs efficacy of treatment. The same concept applies to immunotherapy with checkpoint inhibitors (ICIs). These drugs have in fact the capacity to unleash existing T cells in an unspecific mode, thus breaking the tolerance against self and non-self neoantigens associated with the tumor and permitting the expansion of effector T cells capable not only to recognize but also to destroy the tumor. Interference with inhibitory pathways in the effector T cells and concomitant elimination of immune-suppressive cells such as Treg cells are the dominant mechanisms of enhanced antitumor activity (Melero I, Nat Rev, 2015). Randomized trials using anti-CTLA4, anti-PD-1 and anti-PDL1 have revolutionized treatment and outcome of severe and often fatal cancer disease like metastatic melanoma and metastatic lung cancer demonstrating long-term tumor control and extended patient survival. More recently, combination regimens have furthermore increased clinical responses and survival providing cancer patients with novel treatments. Since ICIs affect T-cell functions, it is not surprising that side-effects profile are entirely distinct from chemo and molecular targeted therapies. This has challenged medical oncologists with a spectrum of novel immune-related toxicities. The main toxicities, observed in a significant proportion of patients, are autoimmune related (Weber JS, JCO 2015). Skin-related toxicity occurs first, colitis appears next and hepatitis and endocrinopathies occur last. Corticosteroids can reverse nearly all of the toxic manifestations. Little is instead known about long-term effect of combination therapy and whether a different kind and range of toxic effects will develop with chronic exposure. Therefore, despite the efficacy of ICI treatments, severe toxicity represents a major cause of reduced dosage, delayed drug administration and therapy discontinuation. The loss of the protective function of intestinal barriers that interact with the environment measured as increased intestinal permeability and the changes occurring in the microbiota composition has been proposed as a mechanism potentially explaining the pathogenesis of colitis. Understanding the role of the intestinal barrier and microbiota dynamics in ICI-treated patients responding and non-responding to immunotherapy can lead in the identification of substances such as antibiotics, probiotics and foods that can influence microbiota population causing alteration of the permeability and drug activity/toxicity (Vétizou M, Science 2015). Unfortunately, selection criteria for anti-CTLA4 and anti-PD-1 therapies are uncertain and currently no predictive markers are available to guide which patients will benefit from these treatments or are less sensible to toxicity. The type of local immunity as well as the circulating immune cells could also predict toxicity, indication on schedules and timing (Fecher LA, The Oncologist, 2013). Biomarkers that have been indicated as relevant in predicting responses are modulation of immune infiltrates in the tumor of both T cells but also immune-inhibitory molecules such as IDO and myeloid-derived suppressor cells (MSDC) (Solito S, Ann N Y Acad Sci, 2014). Genetic profiling of the immune repertoire in the tumor and applied to evaluate the mutational load related to emerging neo-epitopes appear to be important and have been shown to at various degree to correlate with response. Many strategies are under investigation to convert T cell poor tumor in T cell inflamed tumor, in order to increase response to immune therapies. The combination of chemotherapy, radiation therapy or targeted therapy with immunotherapy could influence microenvironment altering of the permeability and drug activity/toxicity. Immunotherapy is improving the survival of patients affected by MM and NSCLC, but it is associated to a new spectrum of toxicities in a high percentage of patients, especially immune related toxicities. Despite efficacy, severe toxicity represents a major cause of reduced dosage, delayed drug administration and therapy discontinuation. New predictive markers of toxicities could help us to reduce the incidence of severe toxicities and improve compliance to treatment. In the context of multiple treatment possibilities, the identification of predictive markers of response and toxicity is a challenging approach for drug selection in order to obtain the best clinical benefit while minimizing the side effects for each patient.

On the basis of literature data we propose to study the association between alteration of intestinal permeability, microbiome and overall survival (OS) - Progression free survival (PFS) and immune-related side effects in patients affected IV NSCLC treated with immunotherapy.
**PRELIMINARY RESULTS**

1. We evaluated 15 patients for serum level of testosterone and Response to treatment and we found an association between KYN/TRYP ratio and PFS ($p < 0.007$). Immunomonitoring evaluations and microbiome evaluations are on going.

**PUBBLICAZIONI 2017**


Roma, 29.09.2017

Prof.ssa M. Torrisi

Prof. P. Marchetti

Dott. Andrea Botticelli