Molecular target therapy and immunotherapy of rare lung tumours

**Tutor:** Prof. Ardizzoni Andrea

**Background:**

In the last years, the discovery of new therapeutic approaches in the treatment of non-small cell lung cancer (NSCLC) has been an event of necessity in contingency. Classic therapy approaches such as chemotherapy treatments, however, encounter side-effect limitations due to their mechanism of action. Despite their ability to kill both cancer and normal cells, secondary acquired-resistance to anti-cancer therapies have dramatically increased in the past few years thus the chance of cancer relapse in patients. Because the fragmentary discoveries of NSCLC-derived molecular landscape, rare but clinically novel driver oncogenes continue to emerge. Notably, new molecular profiling technologies in the evaluation of patient with NSCLC has recently led to the discovery of novel actionable driver oncogenes involving RET gene fusion rearrangements. (1-2) Despite the most reported KIF5B-RET gene fusion type in NSCLC clinical subset, many other RET fusion genes such as CCDC6-RET, NCOA4-RET and TRIM33-RET have been reported as oncogene drivers in 1-2% of NSCLC patients so far. Upon molecular analysis of clinically reported RET rearrangements in NSCLC patients, it has been observed that the aberrant RET genes preserve their tyrosine kinase domain yet upregulating their activity in cancer cells. As consequence of the constitutive activation, increased RET pathway activity downstream the chimeric oncoprotein leads to the proliferation and survival of tumour cells. (3) Interestingly, it is important to note that RET rearrangements typically do not co-occur with other well-established oncogenic mutations in NSCLC such as EGFR, KRAS, ALK, HER2 and BRAF, they are believed to harbor independent oncogenic driver potential.

The benefit of the most developed RET inhibitors such as Cabozantinib, Vandetanib and Lenvatinib, in terms of tumour-response and median progression-free survival has been already demonstrated. (4) Cabozantinib is an oral small molecule tyrosine kinase inhibitor, which interfere with the activity of several receptors such as the vascular endothelial growth factor receptor (VEGFR), MET, AXL and RET. Based on the results of previous studies, Cabozantinib showed activity in patients with RET-rearranged advanced NSCLC (5). Another therapeutic approach that has been developed considerably in the last few years is immunotherapy.

Moreover, emerging data on immune-oncology (I-O) agents paved the way to a new era for the treatment of different solid tumours. In chemo-naïve metastatic NSCLC patients expressing programmed death ligand 1 (PD-L1) over 50%, Pembrolizumab displayed an improvement of progression free survival (PFS) and overall survival (OS) (7-8). Immunotherapy with ICIs has consolidate its role in the treatment of squamous and non-squamous NSCLC over the last few years across different settings and treatment options such as Pembrolizumab in PD-L1+ tumours, Nivolumab for the treatment of all advanced NSCLC patients and Atezolizumab for the treatment of locally advanced or metastatic NSCLC progressing on or after a prior chemotherapy (and an EGFR-or ALK-directed targeted therapy in presence of sensitizing EGFR or ALK genetic alterations) (9,10,11,12).

Nevertheless data about Atezolizumab treatment, as well as other ICIs, in NSCLC of rare histology are lacking and whether these treatments can be an option for these patients is unknown.
This project involves two clinical trials CRETA and CHANCE which will investigate the activity of target therapy and immunotherapy treatments in patients with rare lung tumours.

The results of these studies could provide missing information for the treatment of these otherwise neglected populations and possibly lead to the expansion of the Atezolizumab and Cabozantinib indication in rare NSCLC.

Finally, the exploratory analysis on the role of PD-L1 gene expression and RET rearrangement in both tumour biopsies and circulating tumour cells could provide further information in regards to the interaction between these diseases and the immune system as well as the possible mechanisms which cause the onset of acquired secondary resistance.

**Objectives of the study:**

The main objective of the current project is to follow the activity of data management for the CRETA and CHANCE experimental clinical trials, thus verifying the clinical efficacy, safety, mechanism of action and potential acquired resistance process of target therapy and immunotherapy in patients with rare lung tumor. Furthermore, for CRETA trial, search for RET alteration on circulating tumor cells in the bloodstream is an exploratory goal which would help to deeply describe the possible mechanisms of resistance of the tumor upon Cabozantinib treatment. To do so, analysis of the DNA extracted from the circulating CTC tumor cells and/or tumor cells within the biopsy sample will be optional performed. Consequently, these data could shed light on the potential mechanisms of acquired resistance to the RET inhibition.

**Study design**

- CHANCE trials: Phase II, open-label study of Atezolizumab in 43 patients, advanced non-small cell lung cancer at 12 national centers with rare histological subtypes.
- CRETA: Phase II, open-label study of Cabozantinib in 25 patients at 11 national centers with RET-rearranged non-small cell lung cancer.

**Stages of research**

1) In the first year the data management activity is responsible for the development of both CRETA and CHANCE clinical studies, sponsored by the University of Bologna and by GOIRC, respectively, in patients with rare lung cancer, as regards the design of the clinical trial, the protocol development and related study documents.

Moreover, the activity of data management also includes the following objectives:

1. Assist with the preparation of regulatory applications, amendments, supplements, reports, and advisory panel presentations needed to conduct studies and approve products. (Ethical Committee application and AIFA application)
2. Assure that clinical studies are adequately managed to meet the protocol objectives and schedules (Study documentation development)
3. Establish and maintain first-line contact with investigators, site coordinators, etc.
4. Oversees study compliance/safety including: protocol deviations, adverse events, etc.
5. Ensure sponsor and site compliance to domestic and international government regulations.
6. Prepare, review, and distribute study agreements, informed consent forms, and pre-study visit materials.
2) In second year the activity of data management consists in:

1. Create two custom electronic CRFs based on the characteristics of the two protocols.
2. Assist medical personnel in identifying 2-3 patients pretreated with advanced rearranged NSCLC in coordinator’s center. Data quality and monitoring data entry for all centers.
3. Coordinate the delivery of the cytological/histological material. Moreover, for patients who have joined the translational phase delivery of the hematological sample for CTC research analysis of blood samples is required.
4. Cleaning of the raw data and statistical analyses to evaluate primary and secondary endpoints of the studies and their closure.

Methods

Once the routine diagnostic analysis has been completed, the residual cytological and histological material present in the archive will be viewed by a pathologist (MF) to assess the suitability of their enrollment in the studies.

CHANCE (Immunohistochemical evaluation of PD-L1 expression)

The IHC analysis will be performed with the Benchmark ULTRA automated system (Roche-Ventana) available at the molecular pathology laboratory of the Policlinico (name policlinico) (PAD. 26), using detection/amplification systems and monoclonal antibodies:

1) Anti-PD-L1 (clone SP263, Ventana)
2) Anti- PD-L1 (clone 28-8, Abcam)
3) Anti- PD-L1 (clone 22C3 PharmDX, Dako)

The evaluation of PD-L1 expression in neoplastic cells will be performed in a semi-quantitative way (percentage of positive cells) counting at least 200 neoplastic cells. In the retrospective phase of the study, the results of the three in both cytological and histological samples will be compared each other. In order to assess sensitivity and specificity of each antibody, each type of sample will be assessed using the clone antibody SP263 as standard control. Later it will also be the agreement between different samples for the same case. The best cutting value for each antibody, based on sensitivity and specificity, will be calculated on a ROC curve.

CRETA (Evaluation RET- Status by FISH)

1. The evaluation of RET-translocation will be performed by break-apart FISH probe for the 10p11 locus in tumour biopsies.

References


7. Hui Yu, MD, PhD, Theresa A. Boyle PD-L1 Expression in Lung Cancer. Journal of Thoracic Oncology Vol. 11 No. 7: 964-975


