Can intercalating chemotherapy with Epidermal Growth Factor Receptor inhibitors delay development of treatment resistance in advanced Non-Small Cell Lung Cancer?

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1. Introduction

Lung cancer is the leading cause of cancer-related mortality in the world. Non-small cell lung cancer (NSCLC) accounts for more than 80% of all lung cancers. By the time of diagnosis, the majority of patients are already at advanced stage of disease, and they can only benefit from a systemic palliative treatment. At present, the choice of pharmacological treatment for patients with metastatic disease is based on subject's characteristics, on tumor histology and on genetic alterations (namely, Epidermal Growth Factor Receptor (*EGFR*) activating mutations, Anaplastic Lymphoma Kinase (*ALK*) and Proto-Oncogene Tyrosine-Protein Kinase reactive oxygen species (*ROS-1*) translocations), that are present in only about 20% of Western patients. While platinum-based chemotherapy is the standard of care for patients who do not exhibit *EGFR* activating mutations or other oncogene addictions, the first-line option for NSCLC patients who have tumors that harbor activating *EGFR* mutations is treatment with EGFR-tyrosine kinase inhibitors (EGFR-TKIs), such as gefitinib, erlotinib or afatinib. **[1]** In these patients, EGFR-TKIs are better than chemotherapy as first-line treatment, in terms of progression-free survival (PFS), objective responses rates (ORR), safety profile and health-related quality of life.**[2]** Unfortunately, despite the initial efficacy of the treatments, drug resistance and disease progression almost always emerge. Although the combination of anti-angiogenic drugs and EGFR-TKIs (namely, the addition of the anti-VEGF bevacizumab to erlotinib) as first-line therapy has produced a significant improvement in PFS, the occurrence of resistance remains the clinical rule even in patients treated with this combination.**[3]**

Therefore, together with the identification of better treatments to use as first-line therapy of *EGFR* mutated NSCLC patients, another challenging open question regards the optimal management of those patients who develop resistance to first-line EGFR-TKIs. Different mechanisms can lead to acquired resistance to EGFR-TKIs: among the mechanisms described, the threonine to methionine point mutation in codon 790 of exon

20 *(T790M),* the human *HER2* amplification, and the activation of secondary signaling such as *MET* amplification or *phosphatidylinositol 3-kinase* mutation. In addition, the transformation from NSCLC to SCLC or the epithelial-to-mesenchymal transition have been identified as further mechanisms of acquired resistance to EGFR-TKIs. Among these mechanisms, the *T790M* missense mutation is responsible for the acquired resistance in nearly 60% of patients.**[4]** Osimertinib, a third-generation EGFR-TKI, has shown high activity in *EGFR T790M* positive cases.**[5]** However, although exciting data of PFS and ORR have been reported in patients treated with osimertinib and other third-generation EGFR-TKIs, unfortunately acquired resistance occurs in all patients. Furthermore, the mechanisms of acquired resistance to third-generation EGFR-TKIs have not been completely understood and seems to be extremely various and heterogeneous, probably more complex than resistance to first- and second-generation EGFR-TKIs. **[6]** Thus nowadays, for *T790M* positive disease progressing after treatment with osimertinib, there are no other targeted agents approved. On the other hand, in *T790M* negative cases progressing after first-line EGFR-TKIs, no active biological drugs are available in clinical practice. **[3]** Therefore, chemotherapy remains the standard of care for patients with *T790M* positive tumor progressing after first-line EGFR-TKIs and second-line osimertinib, and for *T790M* negative patients progressing after first-line EGFR-TKIs. Immune checkpoint inhibitors (such as nivolumab, pembrolizumab, atezolizumab) are potential alternative salvage treatments in NSCLC, but subgroup analyses of randomized trials did not show a significant improvement in OS over chemotherapy in EGFR-mutated advanced NSCLC. **[7]**

2. Body of evidence

To date, combination regimens of EGFR-TKIs and chemotherapy have no role in clinical practice. Several randomized phase III trials testing continuous daily administration of erlotinib or gefitinib, in combination with platinum-based chemotherapy doublets, as first-line treatment of patients with advanced NSCLC, have failed to improve survival.**[8,9]** The phase III TRIBUTE trial randomly assigned patients with advanced NSCLC to erlotinib or placebo combined with up to six cycles of carboplatin and paclitaxel, followed by maintenance monotherapy with erlotinib. Median survival was 10.6 months for patients treated with erlotinib and 10.5 months for placebo (hazard ratio, HR 0.99; 95% CI, 0.86 to 1.16; P= .95).**[8]** The phase III INTACT 2 trial, randomized untreated patients with advanced NSCLC to receive chemotherapy with paclitaxel and carboplatin plus the EGFR-TKI gefitinib at the dose of 500 mg/day, or gefitinib 250 mg/day, or placebo. There was no difference in overall survival (OS) (median, 8.7, 9.8, and 9.9 months for gefitinib 500 mg/day, 250 mg/day, and placebo, respectively; P =0.64), TTP, or RR between arms.**[9]** Those negative clinical results were obtained in a population of patients unselected for *EGFR* mutational status, whereas exist evidences of PFS and OS improvement with first-line concurrent combination therapies of the EGFR-TKI gefitinib plus platinum-based doublet chemotherapy (carboplatin/pemetrexed) compared to the sequential alternating administration of gefitinib and chemotherapy. **[10]** A phase III study comparing gefitinib with gefitinib plus concomitant carboplatin/pemetrexed in EGFR mutated NSCLC has recently concluded enrollment, and results are still not available. **[11]**

Recently, Cheng *et al* reported the results of a randomized phase II trial comparing gefitinib alone vs. pemetrexed + gefitinib (continuous administration) as first-line therapy in 195 East Asian patients with advanced nonsquamous NSCLC with activating *EGFR mutations*. **[12]** PFS, that was the primary endpoint, was significantly longer with pemetrexed plus gefitinib (median, 15.8 months) than with gefitinib (median, 10.9 months).

Although the latter promising results underline that combination of chemotherapy and EGFR TKIs could perform better when tested in properly selected patients, it has been speculated that the previous negative clinical results - that were obtained in a population of patients unselected for *EGFR* mutational status - were possibly due not only to inadequate selection of patients, but also to the cell-cycle timing interference. There is growing laboratory evidence of a possible sequence-dependent antagonism as a result of the well-known G_1-phase arrest of tumor cells by EGFR-TKIs, which protect tumor cells from the cell cycle–specific cytotoxic agents.**[13]** Similarly, the attempt of combining continuous administration of EGFR-TKIs with platinum-based doublets chemotherapy after progression to EGFR-TKIs in patients affected by *EGFR* mutation positive NSCLC, did not produce significant improvement in patients' outcome.**[14]** With this background, based on several preclinical data, some interest has grown for a strategy of sequencing and intercalating administration of chemotherapy and EGFR-TKIs. In particular, chemotherapy should be given after EGFR-TKI (pharmacodynamic separation), in order to avoid cell cycle-specific antagonism. Li *et al.* showed that interrupting erlotinib before the subsequent administration of pemetrexed in NSCLC cells should be the most effective sequence for combined treatment with these agents.**[15]** Interestingly, the synergism observed was independent from the mutational status of the *EGFR* or the intrinsic sensitivity to erlotinib.

In another preclinical experience, La Monica *et al* demonstrated that simultaneous treatment with pemetrexed and gefitinib in NSCLC cell lines harboring a deletion in exon 19 of EGFR gene enhanced cell growth inhibition and prevented the appearance of gefitinib resistance of mediated by T790M mutation, only when pemetrexed was the initial treatment. **[16]**

Cheng *et al.* proved that the sequence of paclitaxel followed by gefitinib may be superior to other sequences in treating NSCLC cell lines; in fact, they demonstrated that exposure to paclitaxel in these cell lines resulted in an increased level of phosphorylated EGFR, and

this increase in phosphorylation was inhibited by subsequent exposure to gefitinib.**[17]** In addition, Bello *et al.,* with the aim of identifying the optimal schedule for translation into the clinical setting, demonstrated the efficacy of different treatment schedules involving vinorelbine and gefitinib in NSCLC cell lines (either with the L858R/T790M *EGFR* mutation or wild-type).**[18]** Tsai et al., using a panel of 12 NSCLC cell lines that had no sensitizing *EGFR* mutations *(*SEM*),* demonstrated synergism in combining gefitinib with antimicrotubule agents.**[19]** The same authors tested six different gefitinib–drug combinations in 15 NSCLC cell lines with and without SEMs*.* They showed that in the *EGFR* wild-type NSCLC cells, gefitinib created synergism when treated in combination with pemetrexed or other citotoxic agents (antagonism with cisplatin). Instead, NSCLC cells with SEMs seemed be more chemo-refractory and showed a tendency toward consistent antagonism when gefitinib was used concurrently with chemotherapy. They concluded that concomitant administration of EGFR-TKI and chemotherapeutic agents is not a good treatment strategy for NSCLC patients harboring SEMs, and suggested a strategy of intercalated administration of same agents.**[20]**

Based on those encouraging preclinical evidence, the intercalated administration of EGFR-TKIs (in particular gefitinib or erlotinib), and chemotherapy in patients with advanced NSCLC has been investigated in phase II and phase III, randomized and non-randomized, clinical trials.**[21, 22, 23, 24]** For instance, a phase II study evaluated the administration of cisplatin and docetaxel based chemotherapy plus intercalated gefitinib for advanced NSCLC EGFR mutated patients. **[24]** The primary endpoint of the study was the two–year PFS rate. The 1–, 2–, and 3–year estimated PFS rates were 59.4%, 37.5%, and 33.8%, respectively, and the median PFS time was 19.2 months. The 1–, 2–, and 3–year estimated survival rates were 90.0%, 82.9%, and 62.4%, respectively, and the median survival time had not been reached at the time of analysis. A recent meta-analysis of randomized trials (RCTs) **[21]** showed that this therapeutic strategy might be effective in

the treatment of advanced NSCLC patients, not selected by *EGFR* mutational status. According to the results of this meta-analysis, the intercalated schedule is associated with a significant improvement in OS (hazard ratio [HR], 0.82; 95% confidence interval [CI], 0.71-0.95; P = .01), PFS (HR, 0.60; 95% CI, 0.53-0.68; P < .00001), and objective response rate (ORR; odds ratio [OR], 2.70; 95% CI, 2.08-3.49; P < .00001), compared to chemotherapy alone. Notably, the RCTs considered in this meta-analysis were heterogeneous for the EGFR-TKI used, for the type of chemotherapy (platinum-based or single-agent), for the line of treatment (first-line versus pretreated patients), for the *EGFR* mutational status (which was known only in 43% of the patients included) and for the clinical characteristics of eligible patients (in terms of ethnicity, age, PS, smoking history and tumor histology). Subgroup analyses suggested that in patients with tumors harboring *EGFR*-activating *mutations*, there was a significant benefit in PFS (HR, 0.24; 95% CI, 0.16-0.37; P < .00001) and ORR (OR, 11.59; 95% CI, 5.54-24.25; P < .00001) with the addition of intercalated EGFR-TKI to chemotherapy.

3. Expert opinion

Unfortunately, the results of the meta-analysis**[21]** document the efficacy of the combination compared to chemotherapy, while there is no robust evidence to define the relative efficacy of the intercalated combination compared to single-agent EGFR-TKIs. However, the addition of an EGFR-TKI to chemotherapy could be of interest in the treatment of patients with unknown *EGFR* mutational status: this strategy should be tested in dedicated prospective trials to better define its role, although the lack of information about *EGFR* mutational status should be no longer acceptable in modern clinical practice. As for patients with *EGFR* mutation, who are candidates to receive chemotherapy as second-line (if *T790M* negative) or further line of therapy (if *T790M* positive), the results of the meta-analysis are not directly applicable, because most patients included in the trials considered were treatment-naïve and were not pre-treated with an EGFR-TKI. However, despite the occurrence of molecular changes associated with resistance, the original *EGFR* mutation remains detectable at the onset of resistance, and this finding suggests that *EGFR* should be considered a principal driver for progressing neoplastic clones even after disease progression. **[19]** As a consequence, on the basis of the preclinical data reported and considering the positive results obtained in response rate and survival endpoints with the intercalated combination of EGFR-TKI and chemotherapy in NSCLC, we suggest that intercalated schedule could represent not only a promising way to delay the development of treatment resistance, but also a potentially active strategy after EGFR-TKI resistance development.**[23]** Of course, only well designed randomized trials may help in verifying this hypothesis.

In conclusion, dedicated prospective studies are needed to assess the impact of the intercalated regimen as a weapon to delay or revert the onset of EGFR-TKI resistance in NSCLC patients. Currently, several different clinical trials regarding intercalated EGFR-TKI and chemotherapy in advanced NSCLC are ongoing: both single-arm and randomized

studies, regarding first- or subsequent lines setting, with heterogeneous treatment administered **[Table 1]**. Some of these trials will help to better define the role of intercalated administration of EGFR-TKIs and chemotherapy.

Table 1. Ongoing trials with intercalated EGFR-TKI and chemotherapy in patients with advanced NSCLC

Clinicaltrials.gov Identifier	Phase	Patients	Control arm	Experimental arm	Primary endpoint	Accrual*
NCT02031601	III	EGFR mutated, first-line treatment	Single-agent TKI (erlotinib or gefitinib or icotinib)	Chemotherapy (docetaxel or pemetrexed plus platinum), day 1, plus TKI (erlotinib or gefitinib or icotinib), days 2-15, every 3 weeks	PFS	Recruiting
NCT02064491	II	EGFR mutated, progressed after first-line EGFR-TKI	Chemotherapy (cisplatin or carboplatin plus docetaxel or paclitaxel or pemetrexed), day 1 every 3 weeks	Chemotherapy (same as in control arm), day 1, plus intercalated erlotinib, every 3 weeks	PFS	Completed
NCT02775006	III	EGFR wild type, progressedafter one platinum- containing regimen	Chemotherapy (docetaxel), day 1 every 3 weeks	Chemotherapy (docetaxel), day 1, plus intercalated erlotinib, days 2-16 every 3 weeks	PFS	Recruiting
NCT03151161	II	EGFR mutated, first-line treatment	Chemotherapy (carboplatin plus pemetrexed), day 1 every 3 weeks	Chemotherapy (carboplatin plus pemetrexed), day 1, plus intercalated icotinib, days 2-15, every 3 weeks	Response rate	Recruiting
UMIN000020242	III	EGFR mutated, first-line treatment	Single-agent TKI Gefitinib	Gefitinib on days 1-56 ,cisplatin and pemetrexed on days 71, 92, 113 and again Gefitinib on day 134	OS	Recruiting

EGFR: Epidermal Growth Factor Receptor; TKI: tyrosine kinase inhibitor; NSCLC: non-small cell lung cancer; PFS: progression-free survival;

**as reported in ClinicalTrials.gov and UMIN-CTR Clinical Trial on July 2, 2017,*

Declaration of interest

In accordance with Taylor & Francis policy and our ethical obligation as researchers, we are reporting that Massimo Di Maio has received honoraria for lectures from Novartis, Amgen, Bristol Myers Squibb, and acted as consultant for AstraZeneca, Boehringer Ingelheim, Merck Sharp & Dohme. Antonio Rossi has received honoraria for lectures from Eli-Lilly, Roche, AstraZeneca, Boehringer Ingelheim, and acted as consultant for Eli-Lilly, AstraZeneca, Roche. Anna La Salvia and Emmanuele De Luca have no conflicts of interest to declare. We have disclosed those interests fully to Taylor & Francis.

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