Mutation Analysis and Individualized Therapy in Locally Advanced and Metastatic Pulmonary Adenocarcinoma

PhD Thesis

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Introduction

Lung cancer is the second most common malignant disease (following non-melanoma skin cancer) and it has the highest cancer-related mortality. There was an increase of 51% in the number of newly discovered cases of lung cancer between 1985 and 2011 (the number of cases in males increased by 44% while the increase in women was 76%). Each year 1350000 new cases of the disease are detected worldwide. In Hungary, 5189 new cases were registered (52.5/%000) in 2014 according to data from the pulmonary treatment centers, however, according to the national cancer registry the incidence of lung cancer was 11646, while the prevalence was 21226 (215 %000). Non-small cell lung cancer (NSCLC) accounts for 85-90% of lung cancer cases. Within the cases of non-small cell lung cancer, the number of cases of adenocarcinoma has increased on a global scale in the past 10 years and its proportion reaches 50% in Hungary.

Change in the therapeutic regimeen in NSCLC

Only a quarter of cases of NSCLC are detected at a resectable stage. At the time of diagnosis, 60% of patients have locally advanced (stage IIIB) or metastatic (stage IV) cancer. A systemic treatment must be offered to each patient with an ECOG performance status of 0-2 and stage IIIB or IV cancer (evidence IA). In the treatment of advanced non-small cell lung cancer histology, molecular pathology results, age, performance status, co-morbidities and the patient's preferences have to be taken into consideration. The offered treatment plan must be discussed by a tumor board.

Platinum-based doublet chemotherapy prolongs patients' life and improves the quality of life in the case of performance statuses 0-2. Third-generation cytotoxic medications combined with platinum can only provide an average survival of less than 1 year. Recently there has been a major paradigm shift in the treatment of NSCLC with the discovery of activating ('driver') mutations. The term 'theranostic', which combines therapy and diagnostics to provide individualized treatment, is spreading.

Oncogenic 'driver' mutations

Oncogenic 'driver' mutations are known to be present in approximately 60% of cases of adenocarcinoma. Mutations of receptors or protein kinases can stimulate signal transduction cascades, resulting in an uncontrolled growth, proliferation and survival of tumor cells.

EGFR

The most promising predictive factor of NSCLC is epidermal growth factor receptor (EGFR) located at position 12 on the short (p) arm of chromosome 7. When EGF binds to EGFR, dimerization and autophosphorylation occur and the receptor activates several pathways, leading to cell proliferation, development of metastases and migration, in addition to the inhibition of apoptosis. First-generation EGFR TKIs including erlotinib and gefitinib target the catalytic domain of EGFR through competition of adenosine triphosphatase.

The first studies on EGFR TKIs tried to relate the efficacy of the medications to the overexpression of EGFR. The BR 21 clinical study demonstrated the efficacy of erlotinib in the entire, unselected patient population. Erlotinib was registered as a second- and third-line treatment of NSCLC based on the results of the BR 21 trial in 2005. A study by Lynch TJ. et al. made a breakthrough in the assessment of the efficacy of EGFR TKIs in 2009. Certain mutations of the tyrosine kinase domain (exon 19 and 21 deletions) were proven to predict the efficacy of erlotinib/gefinitib in this study. These so-called activating mutations have more frequently been detected in adenocarcinoma in a certain phenotype – it is present in 50% of the never smoker Asian women - than in other populations. Approximately 90-95% of mutations are either small deletions on exon 19 (codons 746 -750) or arginine substituting leucine at position 858 of exon 21 (L858R). A further 3% of them are substitutions of monoacid variations on codon 719 of exon 18 (G719X) at the position of glycine and 3% are frame insertions on exon 20. The dominant oncogenic role of EGFR is supported by the fact that its occurrence and that of other oncogenic mutations (such as KRAS, ALK or BRAF) are mutually exclusive. The mutant EGFR has a lower ATP-binding capacity than the wild-type, therefore a lower concentration of it is required for inhibition. The most important paradigm shift of the previous 10 years in the treatment of NSCLC was the administration of EGFR TKIs as a first-line therapy in the case of EGFR activating mutations. The first breakthrough was the administration of gefitinib in patients of a Far East origin, subsequently in the EURTAC study dominantly Spanish and French patients

carrying the EGFR mutation were treated with similar results. The prevalence of the classic EGFR mutation was 15% among Spanish patients.

KRAS

KRAS mutation develops as a consequence of smoking and primarily occurs in adenocarcinoma. The presence of KRAS mutations seems to be mutually exclusive with EGFR mutations, is associated with the absence of response to EGFR TKIs and has been linked to a reduced sensitivity to chemotherapy in more and more studies. Based on the results of the BR 21 trial, erlotinib has become an approved medication in Hungary for KRAS mutation negative (wild-type) locally advanced or metastatic lung adenocarcinomas after failure of at least one prior chemotherapy regimeen. As a result, launching an observational cohort study, MOTIVATE, conducted to prospectively collect efficacy data on erlotinib administered to patients with the wild-type KRAS in routine clinical practice, was possible from 2008. The predictive role of classic EGFR mutations has been known since 2009, therefore subsequent determination of EGFR mutations from the tumor samples of 100 patients was carried out.

Tumor angiogenesis as a therapeutic target

It is essential for tumor growth that above a certain size, the tumor forms its own vascular network by activating and stimulating the migration of the endothelial cells of the host. One of the most important signal transduction pathways of tumor angiogenesis is determined by the vascular endothelial growth factor (VEGF) family and its receptors. Inhibiting tumor angiogenesis by medication has yielded major advances in the treatment of several types of solid tumors, most of our knowledge being related to the administration of bevacizumab (Avastin). In lung adenocarcinoma, administration of cytotoxic chemotherapy must be considered if an activating mutation cannot be detected. Bevacizumab administered in addition to a combination of carboplatin and paclitaxel has been shown to improve overall survival (OS) in patients with a performance status of 0 or 1 in a randomized clinical trial. Bevacizumab treatment was continued in patients exhibiting no progression following 4-6 cycles of chemotherapy until disease progression or the occurrence of unacceptable adverse events. Maintenance treatment with Avastin delayed progression of the disease and provided longer OS than induction therapy alone.

Bevacizumab combined with cisplatin/gemcitabine has been proved to increase objective response rate (ORR) and PFS compared with a treatment without bevacizumab in another phase 3 trial, however, no improvement in OS has been shown (AVAIL trial). ESMO and national guidelines recommend chemotherapy to patients with a performance status of 0 or 1 in KRAS/EGFR double wild-type adenocarcinomas. In the case of angiogenesis inhibitor therapy, no biomarker was found which could help to select the therapeutic option. Due to the controversial data we have launched several national multicenter studies (AVALANCHE, AVAMAIN) involving a real patient population with several co-morbidities. In addition to providing data on PFS and OS, these studies are expected to reveal the characteristics of patients eligible for maintenance bevacizumab treatment.

Objectives

a) Determination of the efficacy of erlotinib as a second- or third-line treatment in IIIB or IV-stage lung adenocarcinoma patients screened for KRAS mutation.
b) Investigation of the efficacy and adverse events of EGFR TKI administered as a second- or third-line treatment in advanced and metastatic lung adenocarcinoma.
c) Analysis of the prevalence of classic EGFR mutations in clinical practice. Assessment of the efficacy of EGFR TKIs in cases of lung adenocarcinoma positive for EGFR activating mutation.
d) Investigation of the efficacy of induction and maintenance therapy with bevacizumab in advanced and metastatic lung adenocarcinoma in relation to KRAS mutation status.

Patient population, methods and results

Efficacy of erlotinib as a second- or third-line treatment in IIIB or IV-stage lung adenocarcinoma patients screened for KRAS mutation

MOTIVATE is an open-label, non-randomized, multicenter, non-interventional trial of erlotinib monotherapy investigating the efficacy of erlotinib in routine clinical practice in Hungary. In each case codon mutations 12 and 13 of exon 2 of the KRAS gene were analysed in the histological or cytological samples at the pathological departments of the centers prior to enrollment. Study population included patients with advanced (stage IIIB/IV) KRAS mutation-negative lung adenocarcinoma previously treated with one or two lines of standard systemic chemotherapy. The primary endpoint was PFS, while secondary endpoints were OS and best tumor response. The protocol was approved by the local ethical committee (No: 882-0/2010-1018EKU). Patients were administered erlotinib 150 mg/day orally until disease progression or the development of unacceptable adverse events. Response grading was evaluated by two independent radiological expert investigators using CT, MRI, X-ray or US in every two months. Dose modification of erlotinib and administration of supplementary medication were based on the clinical practice and the discretion of investigators.

Investigation of EGFR mutation: 4 EGFR exons (18, 19, 20, 21) were analysed retrospectively in the tumor samples of 100 patients. PCR amplification and sequencing of the EGFR gene were performed with the Roche 454 FLX sequencing instrument. 47 different tumor mutations were found in 35 of the 100 tumor samples. 16 tumors contained the classic mutations: exon 19 was detected in 9 cases, exon 21 L858R in 5 cases, exon 21 L861Q in 1 case and exon 18 4G717A in 1 case. 18 tumors contained only rare mutations, the clinical significance of which is largely unkown.

Statistical analysis: Descriptive statistical methods were used for analyzing efficacy data. Kaplan-Meier curves were generated to display PFS and OS. Differences between groups were calculated with a Log Rank test. PFS was defined as the time from the start of administration of erlotinib to the first documented progression or death. Table 1 summarizes the demographic and clinical data of patients enrolled to the prospective study.

Table 1.Demographic and clinical data in the MOTIVATE study

Patient characteristics		*n (%)*
Total number of patients		327
Age	Median	60.8 years
Gender	Male	164 (50.2)
	Female	163 (49.8)
Ethnic origin	Caucasian	327 (100.0)
ECOG PS	0	118 (36.1)
	1	169 (51.7)
	2	36 (11.0)
	3	4 (1.2)
Stage	IIIB	101 (30.9)
	IV	226 (69.1)
Therapeutic line	Erlotinib second-line	214 (65.4)
	Erlotinib third-line	113 (34.6)
Smoking status	Never smoker	104 (31.8)
	Former smoker	128 (39.1)
	Current smoker	95 (29.1)

Median PFS was 3.3 months (95% CI 2.93-3.67) and median OS was 14.4 months (95% CI 9.46-19.34). Median PFS and OS were significantly longer in females when compared to males (3.8 vs. 3.2 months;$p< 0.01$ and 20.5 vs. 9.4 months; $p = 0.042$, respectively). Median OS was not significantly longer in patients receiving erlotinib second-line versus those who received erlotinib third-line (16.1 months vs. 9.3 months; $p = 0.631$). No difference was observed in OS when stratified to disease stage (IIIB vs. IV) or smoking status (current smoker, never smoker, former smoker). It should be noted, however, that in the third-line treated group never-smoker patients lived twice as long as current smokers (11 vs. 5.5 months), which was a significant difference ($p = 0.039$). A significant and clinically meaningful correlation was observed between ECOG PS status and survival. The longest median OS (20.5 months) was observed in patients with ECOG PS 0-1 receiving erlotinib in second-line, and in asymptomatic ECOG PS 0 patients (25.7 months), irrespective of the line of treatment (*Fig. 1.*).

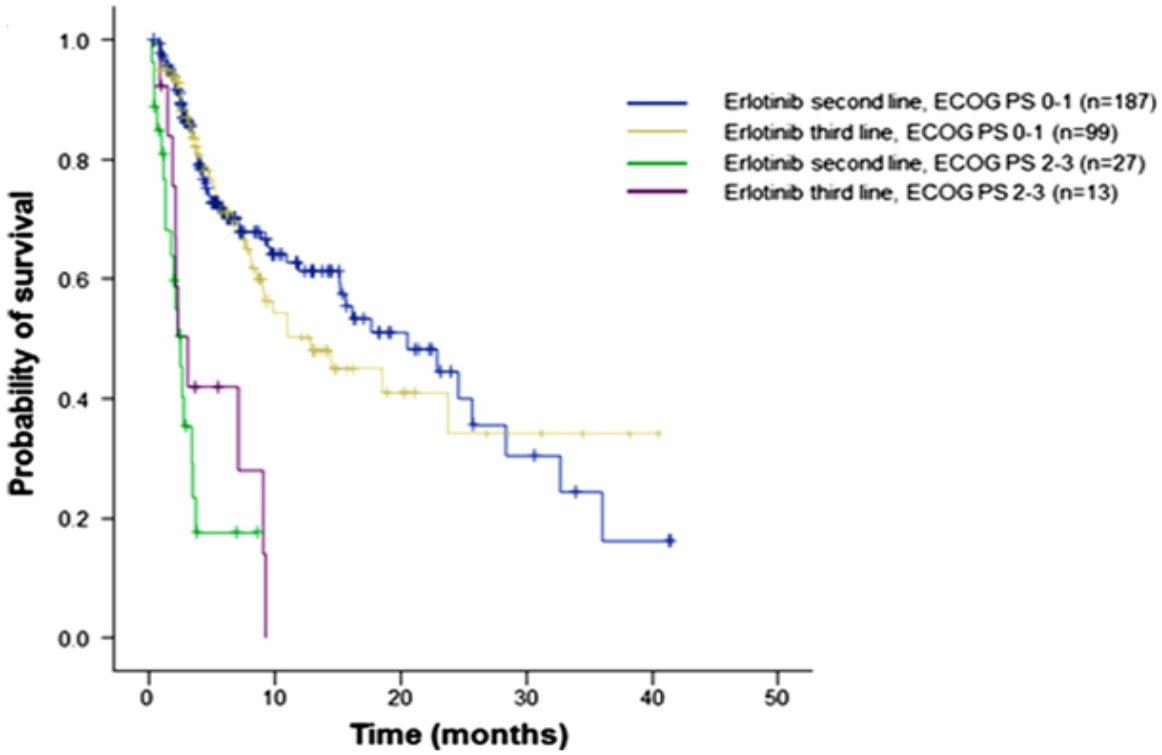

Figure 1. Overall survival of lung adenocarcinoma patients treated with erlotinib according to therapeutic line and performance status

The correlation of PFS and OS with demographic and clinical factors is summarized in Table 2. Male gender and poor ECOG status had a detrimental effect on therapeutic response.

Table 2. PFS and OS according to different baseline characteristics

Characteristic		PFS (months) (95% CI))	*p* value	OS (months) (95% CI))	*p* value
Gender	Male	3.2 (2.99-3.41)	**<0.01**	9.4 (6.29-12.45)	**0.02**
	Female	3,8 (2,85-4,75)		20,5 (10,36-30,71)	
Stage	IIIB	3.5 (2.75-4.19)	0.697	15.6 (9.70-2.57)	0.063
	IV	3.2 (2.72-3.74)		11.0 (5.48-16.46)	
Smoking status	Never smoker	3.5 (1.78-5.28)	0.467	15.2 (6.12-24.28)	0.085
	Former smoker	3.2 (2.89-3.58)		11.9 (4.79-19.02)	
	Current smoker	3.3 (2.73-3.87)		10.9 (3.28-18.52)	
ECOG PS	0	4.8 (2.96-6.57)	**<0.001**	25.7 (16.28-35.12)	**<0.001**
	1	3.1 (2.79-3.34)		10.9 (5.4-16.40)	
	2	2.1 (0.96-3.18)		2.5 (1.85-3.15)	
	3	1.7 (NA)		1.9 (1.61-2.25)	

The correlation of median PFS and OS with the results of EGFR mutation analysis is summarized in Table 3.

Table 3.The correlation of median PFS and OS with the results of EGFR mutation analysis

EGFR gene mutation status	PFS (months)	*p*value	OS (months)	*p*value
EGFR wild-type n=65	2.9		7.2	
Classic EGFR mutation n=16	13.2	**p<0.001**	29.0	**p<0.001**
Rare EGFR mutation n=18	3.0	p=0.432	5.2	p=0.68

The PFS and OS of patients positive for the classic EGFR mutation were significantly longer compared with those carrying the EGFR wild-type and rare mutations.

Best response data were available in the case of 252 patients. Complete response (CR) was found in 3 patients (1.2%), partial response was detected in 51 patients (20.2%) and stable disease (SD) was found in 123 patients (48.8%) as best response, all together resulting in a disease control rate of 70.2%. 75 patients (29.8%) progressed despite erlonitib treatment.

Dose modification was necessary for 48 patients, mainly due to skin lesions and diarrhea. Dose reduction due to skin lesions was required in 11% of the patients, while diarrhoea was responsible for the modification in 4.3% of the cases. A statistically longer PFS was detected in patients requiring dose modification due to adverse events (8.5 months vs. 3.1 months; p<0.001).

The occurrence of classic EGFR mutations in our clinical practice

The efficacy of EGFR TKIs in cases of EGFR activating mutation positive lung adenocarcinoma

In the past 6 years (Jan 2009 - Aug 2015), 446 patients with lung adenocarcinoma were screened for EGFR mutations and EGFR activating mutation was found in 44 cases (9.86%). The EGFR analysis was performed on both cytology samples (pleural fluid, bronchial brush biopsy, transbronchial needle aspiration and transthoracic aspiration specimens) and formalin fixed paraffin-embedded tissue blocks. Following DNA extraction, exon 19 and exon 21 of EGFR were amplified with polymerase chain reaction. The PCR conditions were set as described by Asano et al. The mutated (deleted) and wild-type exon 19 products were separated by fragment length analysis. Following purification, the PCR product was Sanger sequenced using the exon 21

primers to detect point mutations. An expert pathologist evaluated the tumor cell ratio and the sample was rejected for EGFR testing below a ratio of 30%.

Data of these patients were analysed retrospectively with respect to gender, smoking status, type of EGFR mutation and EGFR TKI treatment. In patients receiving EGFR TKI treatment, disease control rate (DCR), PFS, primary resistence and radiological progression were also determined. Statistical analysis was performed using the IBM SPSS Statistics Version 22.0 (SPSS, Inc., Chigaco, IL, USA). Kolmogorov- Smirnov test was used to determine the distribution of the data. Normally distributed data were presented as mean ± SE. Relationships between bimodal variables were analysed with Chi-square and Fischer's exact test. DCR, 6-month and 12-month PFS were investigated with binary logistic regression analysis. Results with p values <0.05 were considered significant.

The average age of the patients with EGFR-mutated lung adenocarcinoma was 69.6 ± 1.6 (40-89) years, while the female/male ratio was 31/13. Males were significantly younger than women (62.9 ± 24.4 versus 72.1 ± 2.0 years; p= 0.008). Never smokers accounted for 77.2% of the patient population, 4 patients were former smokers and 6 patients were smokers at the time of diagnosis. Focusing on EGFR mutations, exon 19 deletion was detected in 61.4 % of the patients, while L858R point mutation in exon 21 was observed in 34.1 % of them. In one case both exon 19 and 21 mutations were detected simultaneously. A rare mutation located in exon 21 was found in another patient. Until the end of the observation period, 38 patients received EGFR TKI treatment: 29 patients were treated with erlotinib and nine received gefitinib. TKI treatment was started as 1st, 2nd and 3rd line treatment in 22, 11 and 5 cases, respectively. The efficacy of TKI treatment was first evaluated after 2 months of therapy. Data were available in 35 cases, because 3 patients died within two months after starting the treatment. Primary resistence to TKI was determined if disease progression was detected at the 2-month visit; it occurred in 5 cases. The beneficial effect of TKI treatment was found in 30 patients, with a disease control rate of 85.7%. One patient responded with complete remission, while partial response and stable disease were detected in 16 and 13 cases, respectively. No difference was found between groups of patients responding with complete or partial remission and stable disease with respect to age, gender, mutation type, smoking status and TKI therapy. The progression free survival of the 35 patients was 12.4 ± 2.1 months, the longest PFS being 54 months and this patients is still on TKI treatment. At the time of closing the database, 6 patients received TKIs for less than 6 months, 21

patients exhibited PFS over 6 months (72.4%) and 11 out of 22 patients survived without progression over 12 months (50%).

PFS over 6 months was significantly more common in never smokers compared to current smokers and former smokers (p=0.005); age, gender, and the type of mutation did not impact on the efficacy of TKI treatment and the duration of PFS. The independent determinants of therapeutic response are depicted in regression models (*Table 4*). Age, gender, type of mutation, smoking status and the sequence and type of TKI therapy were included in these analyses. The sequence of TKI therapy was an independent determinant of therapeutic response (p=0.046); DCR was higher in patients treated in first line (94.7%) than in those treated in second or third line (81.8% and 60.0%). The detrimental effect of smoking and former smoking was confirmed at the 12-month PFS.

Table 4. Independent determinants of therapeutic response to EGFR TKI therapy in lung adenocarcinoma patients with mutated EGFR - binary linear logistic regression models

	Variables	Independent determinants	p value	Cox & Snell R^2	Nagelker-ke R^2
DCR	Age Gender Non-smoking Type of EGFR mutation Sequence of TKI therapy Type of TKI	Sequence of TKI therapy	0.045	0.100	0.179
12-month PFS	Age Gender Non-smoking Type of EGFR mutation Sequence of TKI therapy Type of TKI	Non-smoking Type of TKI	0.052 0.014	0.359	0.478

AVALANCHE prospective multicenter study

With regard to the results of pivotal clinical studies, we found it worthwhile to investigate the efficacy of platinum-based chemotherapy/bevacizumab combinations in Hungarian patients in a postmarketing observational study.

In the AVALANCHE study the efficacy of simultaneous administration of bevacizumab and a combination of third-generation cytotoxic agents and platinum was investigated in Hungarian clinical practice. The observational study was carried out at 28 sites and the period of enrollment lasted from June, 2008 to April, 2011. IIIB/IV-stage lung adenocarcinoma patients received a total of 6 cycles of a combination of platinum-based doublet chemotherapy and bevacizumab (7.5 mg/ body weight in kilograms). Patients without progression received maintenance treatment with bevacizumab until the occurrence of progression or unacceptable toxicity. PFS was the primary endpoint and OS and DCR were the secondary endpoints. Patients were also analysed regarding whether they exhibited progression or not during the initial therapy, i.e. whether they received maintenance therapy with bevacizumab or not. The groups were compared with a log rank method.

283 IIIB/IV-stage lung adenocarcinoma patients were enrolled to the study. Patient characteristics: average age was 58.2 (18-78) years, 55.5% of them were male, 18.4% were in IIIB stage, 79.9% were in IV stage, the adenocarcinoma/other ratio was 95:8/4.2 % and ECOG

performance statuses were 0:30.8%, I: 59.7%, II:2.6% and ≥ III:1.4%, respectively. 43% of the patients received maintenance treatment with bevacizumab in addition to induction treatment. Median PFS was 7.2 months and OS of the entire cohort was 15.2 months. Patients receiving maintenance treatment with bevacizumab had longer PFS and OS as well (Table 5).

Table 5. Median PFS and OS in the AVALANCHE study in relation to maintenance of Avastin treatment

	Without maintenance of Avastin	With maintainedAvastin	Log Rank p value
Median PFS (months)	5.8	9.1	<0.001
Median OS (months)	10.2	26.2	<00001

Best responses of patients enrolled to the study were: CR/PR/SD/PD: 1.5/29.9/26.9/9.1%.

Platinum-based doublet/bevacizumab in first-line in IIIB/IV-stage adenocarcinoma patients

At the 1st Department of Medicine, University of Pécs, Clinical Center, data of 174 IIIB/IV-stage lung adenocarcinoma patients having received first-line bevacizumab treatment combined with chemotherapy between 2 May, 2012 and 26 July, 2015 were analyzed. To determine the efficacy of the therapy, PFS and OS data of all patients receiving bevacizumab treatment were investigated. PFS and OS data were evaluated stratified to KRAS mutation status. PFS and OS values of patients having received maintenance treatment with bevacizumab and those of patients not enrolled to the maintenance phase were compared. All patients had an ECOG PS of 0 or 1 and 69 of them were administered combination treatment with bevacizumab. Sufficient data on PFS were available in 61 cases and in 58 cases there were data available to calculate the survival too.

Therapeutic regimees applied:	CBP-Paclitaxel-Avastin:	74.0% (51/69)
	CBP-Docetaxel-Avastin:	20.3% (14/69)
	Cisplatin-Docetaxel-Avastin:	2.9% (2/69)
	CBP-Gemcitabine-Avastin:	1.4% (1/69)
	Cisplatin-Gemcitabine-Avastin:	1.4% (1/69)

Forms of Avastin treastment: Administered only in the induction phase: 44.9% (31/69)
Administered in the maintenance phase too: 55.1% (38/69)

The average age of the patients was 60.93 years and 53.6% of patients included in the retrospective study were male and 46.4% were female. Prior to the therapeutic decision KRAS mutation status was determined for all patients. 36.8% of the patients were positive for KRAS mutation. The estimated PFS and OS values are summarized in *Table 6* stratified to KRAS mutation status and maintenance of Avastin treatment.

Table 6. Estimated average and median PFS and OS stratified to KRAS mutation status and maintenance of Avastin treatment

	estimated PFS			estimated OS		
	average	median	p value	average	median	p value
total	11.8	6.0		20.2	14.0	
KRAS wild	12.8	7.0	0.387	22.5	16.0	0.169
KRAS-mutated	7.4	4.0		10.7	11.0	
receiving maintained Avastin treatment	17.4	12.0	<0.001	27.9	21.0	<0.001
not receiving maintained Avastin treatment	3.8	3.0		9.3	7.0	

KRAS mutation status did not impact on progression-free survival and survival, however, PFS and survival of patients receiving maintenance treatment with bevacizumab were significantly better (*Figure 2*).

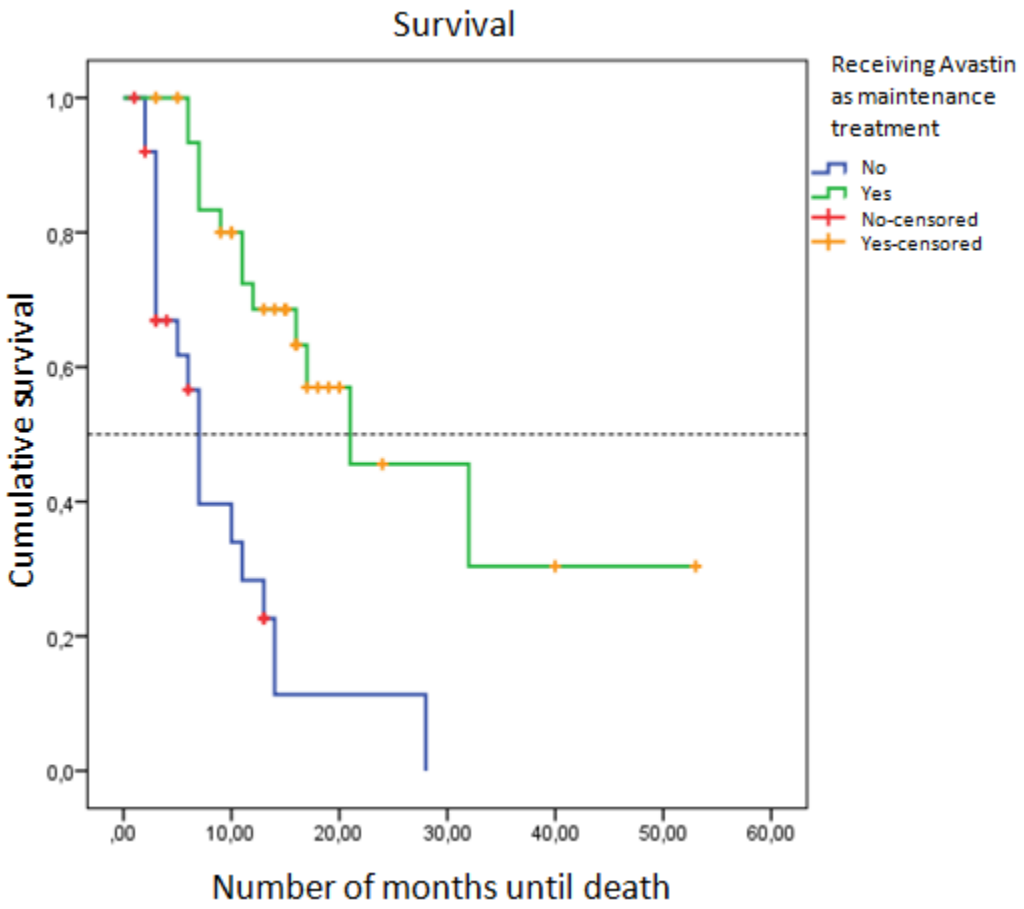

Figure 2. Survival of lung adenocarcinoma patients having received maintenance treatment with bevacizumab following platinum-based doublet bevacizumab induction treatment with regard to maintenance treatment with bevacizumab (N=69)

Discussion

Erlotinib was proved to be more effective with regard to PFS and OS compared to placebo in a retrospective analysis of subgroups of NSCLC patients carrying the wild-type EGFR in the BR 21 and SATURN studies in 2013. The results of MOTIVATE, a multicenter study launched in Hungary in 2008, confirmed the efficacy of erlotinib treatment administered in second and third line in lung adenocarcinoma patients carrying the KRAS wild-type in Hungarian routine clinical practice.

The longest survival (20.5 months) was detected in ECOG PS 0-1 patients screened for KRAS mutation receiving second-line erlotinib treatment. This finding draws the attention to the importance of assessing patients' performance statuses objectively. In patients carrying wild-type KRAS, long survival was found with a relatively low remission rate and, simultaneously, almost 50% of stable disease control. This result confirms the fact that the increased ability of tumor cells to multiply is in the background of tumorgenesis in EGFR wild-type tumors. This mechanism is inhibited by erlotinib, resulting in the stabilization of disease. Survival data of 100 patients analyzed retrospectively for EGFR (exons 18-21) mutation indicated that patients carrying the classic 19, 21 mutations had considerably longer PFS and OS values than those carrying rare mutations.

At our Division of pulmonology, the frequency of EGFR mutation is 9.86%, which is lower than it was previously published for Caucasian populations. The distribution of mutations was similar to literature data. TKI treatment of these patients is promising, since 85% of the patients benefited from TKI therapy. First-line treatment seems to be the best therapeutic option, which highlights the importance of early evaluation of EGFR status to determine first-line treatment.

Randomized, phase 3 clinical trials demonstrated that never smokers with EGFR mutated adenocarcinoma had the largest benefit from TKI treatment. Our retrospective analysis also revealed that none of the patients who were positive for EGFR mutation and were smokers at the time of the diagnosis had PFS over 6 months. PFS over 6 and 12 months was detected in 72.4% and 50 % of patients, respectively, irrespective of age, gender and type of mutation. It is important to note that male gender was not associated with disadvantages in our patient population. 20% of the patients required dose reduction due to adverse events.

The other new therapeutic approach is inhibition of tumor angiogenesis combined with chemotherapy to improve therapeutic response of lung adenocarcinoma patients. In the two pivotal clinical trials, 66% of the patients completed chemotherapy without progression and were administered maintenance treatment with Avastin. PFS and OS values of patients receiving maintenance treatment with bevacizumab were higher than those of patients not receiving maintenance therapy. In the national AVALANCHE study, 40% of the patient population received maintained Avastin treatment following induction treatment. Patients receiving maintenance treatment had a 2.5-times longer survival than patients not receiving maintenance treatment. It can be established that fewer patients reached the maintenance phase, however, these patients had a considerably long survival. Our own retrospective clinical study also confirmed that patients receiving maintenance Avastin treatment exhibit a 3-times longer overall survival compared to those who are enrolled only to the induction phase (21 vs. 7 months).

Comparing international and available Hungarian data, which show an occurrence of 25-29.55% for KRAS mutations, our study detected a higher occurrence rate of 36.8% for KRAS mutations.

International data support that KRAS codon mutations 12 and 13 had a negative predictive value with regard to platinum-based therapy. In our study KRAS mutation status did not have a significant impact on PFS and OS values given in response to platinum doublet bevacizumab combination therapy, although such a tendency was detectable, the difference did not prove to be significant, probably due to the small number of cases.

Summary of novel results

1. A lower number of lung adenocarcinoma patients positive for the classic EGFR mutations were diagnosed in the oncopulmonological center of the University of Pécs compared to available European data. The proportion of patients positive for EGFR mutations 19 and 21 was similar to that found in the database of the National Korányi Institute. The number of patients carrying the KRAS mutation was higher in our population.
2. EGFR TKI treatment had the best disease control when administered in first line. This result highlights the importance of determining KRAS mutation status prior to the start of the therapy. The treatment provided a progression-free survival of longer than 6 months in never smokers. Male gender was not associated with disadvantages with regard to TKI treatment.
3. Erlotinib treatment administered in second and third line taking KRAS negativity as a selection criterion proved to be considerably effective in adenocarcinoma patients carrying the wild-type KRAS in MOTIVATE, a national prospective, multicenter study.
4. A retrospective analysis in MOTIVATE study has also shown that patients with wild-type KRAS exhibited the best survival when they were carrying the classic EGFR mutation. Patients with the double wild-type of adenocarcinoma had a shorter survival.
5. AVALANCHE national multicenter prospective study showed a longer OS in patients receiving maintenance of Avastin treatment compared to those enrolled to the clinical studies.
6. In our retrospective studies, patients received maintained Avastin treatment in a high proportion owing to proper selection of patients. Patients receiving maintenance of Avastin treatment had significantly longer PFS and OS than those not receiving maintenance treatment. In our routine clinical practice we had similar PFS and OS values to those detected in clinical studies.

List of publications

Publications related to the topic of the thesis:

1. **Sárosi V**, Balikó Z. Erlotinib (Tarceva®) in Second/Third Line Treatment for Non-Small Cell Lung Cancer (NSCLC)-Case Description. HUN MED J 2007; 1(3):307-313.
2. **Sárosi V**, Ruzsics I, Enyezdi J, Grexa E, Balikó Z. Az Avastin kezelés biztonságossága egy tüdő-adenocarcinomás beteg kezelése kapcsán. TÜDŐGYÓGYÁSZAT 2009; 3 (11):24-25.
3. **Sárosi V**, Balikó Z, Albert B. Az EGFR tirozinkináz-gátlók gyakori mellékhatásainak kezelése. TÜDŐGYÓGYÁSZAT 2010; 4(6): 28-33.
4. **Sárosi V**, LosonczyG, Francovszky E, TolnayE, Torok S, Galffy G, Hegedus B, Dome B, Ostoros G. Effectiveness of erlotinib treatment in advanced KRAS mutation-negative lung adenocarcinoma patients: Results of a multicenter observational cohort study (MOTIVATE). LUNG CANCER 2014; 86(1):54-58.

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5. **Sárosi V**, Balikó Z.Az első vonalban alkalmazott afatinib versus kemoterápia hatásossága EGFR-mutációpozitív tüdőadenokarcinómában MAGYAR ONKOL 2014; 58 (4):325-329.
6. **Sárosi V**,Balikó Z,Mezősi E. Az afatinib aktivitása nem szokványos EGFR mutáció pozitív tüdőrákokban a LUX-Lung 2,Lux-Lung 3, és Lux-Lung 6 vizsgálatok alapján MED. THORAC 2015; 68(2):162-6.
7. **Sárosi V**, Balikó Z, Smuk G, Szabó M, Ruzsics I, László T, Mezősi E. The frequency of EGFR mutation in lung adenocarcinoma and the efficacy of tyrosine kinase inhibitor therapy in a Hungarian cohort of patients. PATHOL ONCOL RES. 2016. APR. DOI 10.1007/a12253-016-0063-8.

   IF: 1.855

Cumulative impact factors of publications related to the topic of the thesis: 5.813

Other publications:

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