Clinicopathological and genetic characteristics associated with brain metastases from lung adenocarcinoma and utility as prognostic factors

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Abstract. Brain metastases (BM) are common in patients with lung adenocarcinoma, and represent a significant cause of morbidity in the disease. A more comprehensive understanding of the clinicopathological characteristics that serve as prognostic factors for survival in patients with BM from lung adenocarcinoma may aid in informing treatment strategies for this patient population. In the present study, clinicopathological factors, including EGFR mutation status, were evaluated in 59 patients who were diagnosed with BM from lung adenocarcinoma, and underwent BM resection between January 1985 and December 2014 at Fukuoka University Hospital. The most frequent subtype of BM from lung adenocarcinoma was solid adenocarcinoma (57.6%), followed by papillary adenocarcinoma (22.0%) and acinar adenocarcinoma (18.6%). A total of 14 patients (23.7%) exhibited EGFR mutations, which were significantly associated with female sex (9/14, 64.3%), non-smoker status (8/14, 57.1%), BM in the frontal lobes (9/14, 64.3%) and papillary adenocarcinoma (5/14, 35.7%). Statistical analysis revealed a significant association between non-smoker status and BM in the frontal lobes, and more favorable disease prognosis. The results of the present study suggest that histological and genetic analysis of tissue from BM provides information useful for managing treatment of patients with resectable BM arising from lung adenocarcinoma.

Introduction

The frequency of brain metastases (BM) from non-small cell lung cancer (NSCLC) exceeds 20% at the time of diagnosis of the primary tumor, and a significant proportion of patients develop BM over the course of disease (1,2). In recent years, the incidence of BM in patients with NSCLC has increased, due to improved survival rates resulting from the molecular-targeting therapy and improvements in diagnostic neuroimaging modalities, including magnetic resonance imaging and positron emission tomography (3). Adenocarcinoma is the most common subtype of NSCLC, occurring in ~40% of all patients, and is characterized by rapid progression, early distant metastasis and a significantly higher incidence of BM compared with other subtypes of NSCLC (4-6). BM are considered to be a poor prognostic factor in NSCLC, and are associated with median overall survival (OS) times of 4-7 weeks in untreated patients (6-8). In patients with BM, recursive partitioning analysis (RPA) and the graded prognostic assessment (GPA) score are still the most widely accepted models for evaluation of disease prognosis (6,9).

In lung adenocarcinoma, mutations in the epidermal growth factor receptor (*EGFR*) gene have been identified as a driver mutation promoting tumor growth (10-12). Therapies targeting mutant EGFR protein are associated with relatively high overall response rates, and have been established as standard-of-care treatments (10-12). Furthermore, EGFR-targeted therapies have demonstrated efficacy in the treatment of BM from NSCLC (13-15).

Histological and genetic evaluation is essential in identifying patients with NSCLC that may benefit from EGFR-targeted therapies. In cases in which tissue sampling from the primary tumor is difficult, tissue samples from BM lesions may provide a useful alternative for evaluation. In the majority of previous reports, histological analysis and genetic evaluation in patients with NSCLC have been performed using tissue from the primary tumor only (10-15). There are a few reports describing a histopathological and genetic evaluation

using tissue samples from BM and the primary lung adeno-carcinoma together (16,17). Given evidence that the presence of *EGFR* mutations is a positive prognostic factor in patients with BM from lung adenocarcinoma (13-15), it is feasible that analysis of tissue from BM may be used to guide more aggressive and effective treatments for patients with *EGFR* mutation-positive NSCLC.

In the present study, the clinical features and histopatho-logical subtypes of BM from lung adenocarcinoma in a patient population from a single institution were retrospectively evaluated in order to identify prognostic factors. In addition, an evaluation of *EGFR* mutations and their association with disease prognosis was assessed. The results of the present study suggest that the analysis of tissue from BM may inform the treatment of patients with metastatic brain lesions arising from lung adenocarcinoma.

Materials and methods

Study design and patients. A total of 232 BM samples in the histopathological files of the Department of Pathology that had been surgically resected at the Department of Neurosurgery, Fukuoka University Hospital (Fukuoka, Japan), between January 1985 and December 2014 were reviewed. Tumor resection was performed to improve neurological symptoms and quality of life, and aimed to prolong the survival time of patients. Patients in which BM were resected included those in which: i) A single large (>3 cm) or symptomatic tumor was present; ii) the primary tumor was well-controlled; iii) life expectancy exceeded 3 months; and iv) written informed consent from the patient or family was obtained. A total of 68 patients with BM from lung adenocarcinoma were retro-spectively analyzed. Analysis data from eight patients lacking clinical data and specimens, and from one patient in which polymerase chain reaction (PCR) amplification of *EGFR* failed were excluded. Data from a total of 59 patients were included in the present study. The current study was reviewed and approved by the Ethics Committee of the Fukuoka University Hospital (approval no. 15-5-14).

Pathologic evaluation. Surgically-resected specimens were fixed in 10% formalin (room temperature for 24 h) and embedded in paraffin blocks. Tissue sections were cut at a thickness of 4 μm, and stained with hematoxylin and eosin for histopathological examination. Tissue sections were stained with hematoxylin for 10 min, and with eosin 3 min at room temperature. Histological subtyping was performed according to the 2015 World Health Organization criteria for classifica-tion of lung tumors (18). The clinicopathological parameters considered in this study included patient age, sex, location of BM, histological subtype of BM, smoking status, Karnofsky performance score (KPS), number of BM, presence of extra-cranial metastasis (ECM), GPA, timing of diagnosis, tyrosine kinase inhibitor (TKI) treatment prior to diagnosis of BM, presence of *RAS* mutations or anaplastic lymphoma kinase (*ALK*) rearrangements and re-operation.

Analysis of EGFR and KRAS mutations and ALK rear-rangement. *EGFR* and *RAS* gene mutations were analyzed in paraffin-embedded tissue sections from BM only. *EGFR* mutations (including G719X in exon 18, exon 19 deletion, T790M in exon 20, and L858R and L861Q in exon 21) and *RAS* mutations (G12C and G12A in exon 2 and Q61H in exon 3) were detected in matching formalin-fixed paraffin-embedded tissue samples using cycleave PCR (which was performed by SRL Inc., Tokyo, Japan, cobas® EGFR Mutation Test v2 kit and MEBGEN™ RASKET kit according to the manufacture's protocol). *ALK* rearrangement was analyzed by immunohis-tochemistry (ALK Detection kit; Nichirei Bioscience, Tokyo, Japan), according to the manufacturer's protocols. The slides were observed under x200 magnification using a BX50 fluo-rescence microscope (Olympus Corporation, Tokyo, Japan).

Statistical analysis. To identify clinical features associated with *EGFR* mutations, categorical variables were compared using Wilcoxon signed-rank test or Fisher's exact test. OS was defined as duration between the date of BM diagnosis and the date of mortality or last follow-up. The Kaplan-Meier estimator method was used for survival curve analysis, and differences between survival curves were analyzed using the log-rank test. Multivariate analysis with the Cox proportional hazard model was used to determine independent prognostic factors. P<0.05 were considered to indicate a statistically significant differ-ence. Statistical analysis was performed with SPSS software (version 21.0; IBM Corp., Armonk, NY, USA).

Results

Patient characteristics. Patient characteristics are summarized in Table I. A total of 59 patients with BM from lung adeno-carcinoma were included in the present study. The median patient age was 61.3±12.2 years old (range, 38-87 years). A total of 45 patients were male (76.3%) and 14 patients were female (23.7%). A total of 14 patients (23.7%) exhibited *EGFR* mutations (Table II), of which deletion of exon 19 was the most common genetic alteration (eight patients, 57.1%), followed by L858R mutation in exon 21 (five patients, 35.7%). In addi-tion, one patient (7.1%) had a L861Q mutation in exon 21. *EGFR* mutations occurred significantly more often in female compared with male patients (64.3 vs. 35.7%; P=0.0001). *EGFR* mutations also occurred significantly more frequently in non-smokers compared with current or ex-smokers (57.1 vs. 42.9%; P=0.04). BM within the frontal lobe were significantly more common compared with other locations in patients with *EGFR* mutations [64.3 (frontal lobe) vs. 21.4 (cerebellum) vs. 7.1% (posterior lobe); P=0.03]. The rate of surgery for tumor recurrence occurred significantly more often in patients with wild-type compared with mutated *EGFR* (37.8 vs. 7.1%; P=0.04). The two groups did not significantly differ in GPA measures of age, KPS, number of BM, presence of ECMs, timing of diagnosis, and frequency of *RAS* mutation and *ALK* rearrangements.

All patients received craniotomy followed by total resec-tion of the BM. In 32 patients (54.2%), ECMs were detected at the time of BM diagnosis. The sites at which ECMs occurred in the patients with *EGFR* mutation were similar to those in which ECMs occurred in patients with WT. In patients with *EGFR* mutations, six patients had ECMs (one patient had multiple lesions). Of these ECMs, six occurred in the lymph nodes (100%), one occurred in bone (16.7%), and one occurred

Table I. Patient baseline characteristics (n=59).

Characteristic	Total (n=59)	EGFR MT (n=14)	WT (n=45)	P-value MT vs. WT
Age, years[a]	61.3±12.2	61.6±12.0	61.1±12.4	0.80
Sex (%)				0.0001
Male	45 (76.3)	5 (35.7)	40 (88.9)	
Female	14 (23.7)	9 (64.3)	5 (11.1)	
Location (%)				
Frontal lobe	23 (39.0)	9 (64.3)	14 (31.1)	0.03
Cerebellum	11 (18.6)	3 (21.4)	8 (17.8)	0.76
Posterior lobe	8 (15.6)	1 (7.1)	7 (15.6)	0.67
Histological subtype (%)				
Solid	34 (57.6)	4 (28.6)	30 (66.7)	0.03
Papillary	13 (22.0)	5 (35.7)	8 (17.7)	0.26
Acinar	11 (18.6)	4 (28.6)	7 (15.6)	0.43
Smoking status (%)				0.04
Never	19 (32.2)	8 (57.1)	11 (24.4)	
Current or past	40 (67.8)	6 (42.9)	34 (75.6)	
KPS (%)				
<70	19 (32.2)	6 (42.9)	13 (28.9)	0.51
70-80	15 (25.4)	5 (35.7)	10 (22.2)	0.48
81-100	25 (42.4)	3 (21.4)	22 (48.9)	0.12
Number of BM (%)				
1	31 (52.5)	5 (35.7)	26 (45.8)	0.22
2-3	17 (28.8)	5 (35.7)	12 (26.7)	0.74
>3	11 (18.6)	4 (28.6)	7 (15.6)	0.43
ECM (%)				0.80
Absent	27 (45.8)	6 (42.9)	21 (46.7)	
Present	32 (54.2)	8 (57.1)	24 (53.3)	
GPA (%)				
0-1.0	13 (22.0)	4 (28.6)	9 (20.0)	0.71
1.5-2.0	24 (40.7)	7 (50.0)	17 (37.8)	0.54
2.5-3.0	18 (30.5)	3 (21.4)	15 (33.3)	0.52
3.5-4.0	4 (6.8)	0 (.0)	4 (8.9)	0.56
Timing of diagnosis				
BM diagnosed before PT (%)	27 (45.8)	5 (35.7)	22 (48.8)	0.54
TKI (prior to BM) (%)	2 (3.4)	2 (14.3)	0 (.0)	0.05
RAS (%)	5 (8.5)	0 (.0)	5 (11.1)	0.16
ALK (%)	3 (5.0)	0 (.0)	3 (6.7)	0.57
Re-operation (%)	18 (30.5)	1 (7.1)	17 (37.8)	0.04

[a]Data presented as the mean ± standard deviation. EGFR, epidermal growth factor receptor; MT, mutation; WT, wild type; KPS, Karnofsky performance score; BM, brain metastasis; ECM, extracranial metastasis; GPA, graded prognostic assessment; PT, primary tumor; TKI, tyrosine kinase inhibitor; ALK, anaplastic lymphoma kinase.

in liver (16.7%). In patients with WT, 21 patients had ECMs (4 patients had multiple lesions). Of these lesions, 15 occurred in the lymph nodes (71.4%), six occurred in bone (28.6%), two occurred in liver (9.5%) and two occurred in the adrenal glands (9.5%). There was no statistically significant difference in the sites at which ECMs occurred in comparing patients with these two conditions. Regarding the number of BM present, one in 31 patients (52.5%), two in 13 patients (22.0%), three in four patients (6.8%), four in two patients (3.4%) and ≥five in nine patients (15.3%). Following surgical resection of BM, the majority of patients received local therapy (38 patients, 64.4%), consisting of whole brain radiation therapy (WBRT; 24 patients,

Table II. EGFR mutation analysis with brain metastasis from lung adenocarcinoma (n=59).

EGFR mutation	No. of patients (%)
Exon 19 deletion	8 (57.1)
Exon 21 L858R	5 (35.7)
Exon 21 L861Q	1 (7.1)
Exon 18 G719X	0 (0.0)
Exon 20 T790M	0 (0.0)

A total of 14/59 patients had EGFR mutations. EGFR, epidermal growth factor receptor.

40.7%) or stereotactic radiosurgery (SRS; 14 patients, 23.7%). Following local therapy, 18 patients (30.5%) received systemic chemotherapy. A total of five patients (8.5%) received systemic chemotherapy only, while two patients (3.4%) with *EGFR* mutations were treated with the EGFR-TKI gefitinib prior to the diagnosis of BM. Patients with poor performance status and patients that declined treatment (14 patients, 23.7%) were provided with best supportive care.

Pathological subtypes. The predominant subtypes of BM associated with lung adenocarcinoma were solid adenocarcinoma (34 patients, 57.6%), followed by papillary adenocarcinoma (13 patients, 22.0%), acinar adenocarcinoma (11 patients, 18.6%) and micropapillary adenocarcinoma (one patient, 1.8%) (Table I). *EGFR* mutations occurred most commonly in papillary adenocarcinoma (five patients, 35.7%), followed by acinar adenocarcinoma (four patients, 26.7%), solid adenocarcinoma (four patients, 26.7%) and micropapillary adenocarcinoma (one patient, 7.1%). No significant association was identified between the occurrence of an *EGFR* mutation and histological subtype. However, the wild-type *EGFR* gene was significantly more common in patients with solid adenocarcinoma compared with other histological subtypes. Fig. 1 shows representative histological subtypes of BM from lung adenocarcinoma.

Tissue from the primary lung adenocarcinoma together with BM were available for 20 patients. Characteristics of primary lung adenocarcinoma and BM are summarized in Table III. The most common histological subtype in patients with primary lung adenocarcinoma who also had BM was solid adenocarcinoma (10 patients, 50.0%), followed by acinar adenocarcinoma (seven patients, 35.0%), and papillary adenocarcinoma (three patients, 15.0%). Histopathological subtypes of lung adenocarcinoma and BM matched each other in 75% of all of cases.

Patient survival. After a mean follow-up duration of 17.9 months, 39/59 patients (66.1%) had succumbed, 10 patients (16.9%) survived and 10 patients (16.9%) were lost to follow-up. The median OS was 19.2 months. Table IV presents the results of the OS univariate analysis. The OS time was significantly increased in patients with *EGFR* mutations (median OS of 31.5 months vs. 15.6 months; P=0.02; Fig. 2A). No significant difference was observed in OS in patients with

exon 19 deletion compared with those with L858R mutations in exon 21 (P=0.31, data not shown). In addition, the OS time was significantly increased in patients with BM in the frontal lobe compared with patients with BM the cerebellum or in other regions [median OS, 28.2 (BM in the frontal lobe) vs. 13.7 (BM in the cerebellum) vs. 12.7 months (BM in other brain regions); P=0.004] (Fig. 2B) and in non-smokers (33.1 months in non-smokers vs. 12.5 months in current or ex-smokers; P=0.001; Fig. 2C).

Patients diagnosed with BM following initial diagnosis of the primary lung adenocarcinoma primarily had a longer OS, compared with patients with an initial diagnosis of BM prior to lung cancer (median OS, 23.4 vs. 12.5 months; P=0.079), but this did not reach statistical significance. Patient age, histological subtype of BM, KPS and re-operation were not identified to be associated with OS. Statistical analysis of patients with *ALK* rearrangement was not feasible as only three patients with this genetic change were present in the patient population.

Multivariate analysis suggested that BM located in the frontal lobe and a non-smoker status were independent and significant prognostic factors (P=0.007 and P=0.008, respectively; Table V).

Discussion

In the present study, the clinical and histological features of patients that developed BM from lung adenocarcinoma were evaluated in order to identify prognostic factors for such patients. Previous studies have investigated the incidence of *EGFR* mutations in a large population of patients with lung cancer (19,20). *EGFR* mutations have been detected in 30% of patients with adenocarcinoma, 38% of female patients, 47% of non-smokers and 32% of patients of East Asian origin in the patients with lung adenocarcinoma (19,21). It has also been reported to be easier to detect *EGFR* mutations in smaller primary tumors (22). Deletion of exon 19 and L858R mutations in exon 21 account for ~85% of *EGFR* mutations and have been recognized as important markers in determining the appropriate treatment strategy for patients with advanced lung cancer (21). BM from lung cancer were detected at the time of diagnosis of the primary tumor in 8-23% of patients with *EGFR* mutations, and in 24% of patients with NSCLC during follow-up care (23,24). In patients with NSCLC in which *EGFR* mutations were present, 34.2% were identified to have developed BM 1 year after diagnosis, 38.4% at 2 years, 46.7% at 3 years, 48.7% at 4 years and 52.9% at 5 years in the USA (25).

At the time of the initial diagnosis of primary lung adenocarcinoma, the incidence of *EGFR* mutation is reported to vary according to the site of BM (24,26). Patients with lung adenocarcinoma with *EGFR* mutations have been suggested to have a higher incidence rate of BM compared with patients without mutations (24,26). However, another study reported that the timeline of development of BM was not significantly longer in *EGFR*-mutated NSCLC compared with wild-type *EGFR* NSCLC (27). Therefore, the association between the presence of *EGFR* mutations and BM is controversial. Multiple BM (≥2) have been reported to occur more frequently in patients with *EGFR* mutations, but the size of the largest

Table III. Pathological subtype of primary lung adenocarcinoma and metastatic brain tumor (n=20).

No.	Stage	Size (mm)	Primary	BM
1	T1aN0Mx	11x10x15	Solid (solid + acinar)	Solid
2	T1bN0M0	20x15x20	Solid (solid + acinar)	Acinar
3	T2aNxMx	40x30x25	Acinar	Acinar
4	T1cN0M0	28x26x24	Acinar (acinar + solid)	Acinar
5	T1bN0Mx	20x15x15	Acinar (acinar + papillary)	Papillary
6	T1bNxMx	20x10x8	Solid	Solid
7	T1bNxMx	20x8x8	Solid (acinar + papillary + MPP)	Solid
8	T2aNxMx	30x38x35	Solid (solid + acinar + papillary)	Acinar
9	T4N2M1	40x35x35	Acinar (acinar + MPP)	Solid
10	T1cN0Mx	30x25x20	Solid (solid + acinar)	Solid
11	T2NxM1	15x10x10	Solid (solid + acinar + MPP)	Solid
12	T3N0M1	40x35x35	Solid (solid + acinar)	Solid
13	T4N0Mx	70x70x55	MPP (MPP + acinar + solid)	Solid
14	T1bN0Mx	16x15x15	Solid (solid + acinar)	Papillary
15	T2aN2M0	18x17x24	MPP (MPP + acinar + papillary)	Solid
16	T4N2Mx	85x60x50	Solid (solid + MPP + acinar)	Solid
17	T1cNxMx	22x18x10	Acinar (acinar + MPP + papillary)	MPP
18	T2bN2Mx	50x50x30	Acinar (acinar + solid + papillary)	Solid
19	T1cNxMx	30x20x27	Acinar (acinar + papillary)	Papillary
20	T2aNxMx	35x40x25	MPP (MPP + acinar)	Papillary

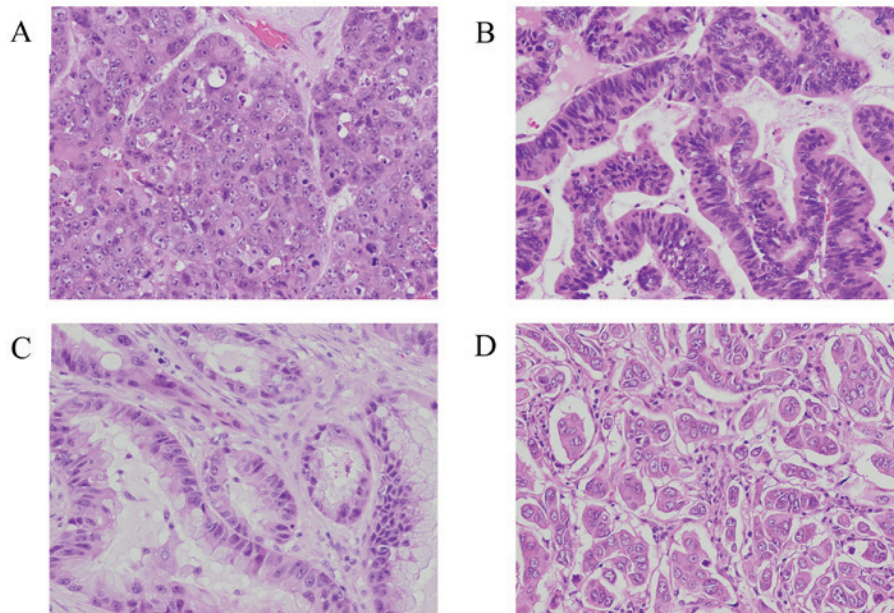BM, brain metastasis; MPP, micropapillary.

Figure 1. Histopathological subtypes of brain metastases from lung adenocarcinoma (hematoxylin and eosin staining; magnification, x200). (A) Solid predominant, (B) papillary predominant, (C) acinar predominant and (D) micropapillary predominant.

BM does not appear to be associated with the presence of *EGFR* mutations (28).

Several studies have compared *EGFR* status in the primary tumor and BM in patients with NSCLC (26,29). These results suggested high concordance rates in *EGFR* mutation status (93.3-100%) between the primary tumor and BM (26,29). In the current study, 5/20 patients examined had *EGFR* mutations in the primary lung adenocarcinoma and BM. Five patients had the same *EGFR* mutation in both sites (three patients had exon 19 deletions and one patient each had had L858R and L861Q substitutions in exon 21). In the present study, the frequency of *EGFR* mutations in patients with BM (23.7%) was
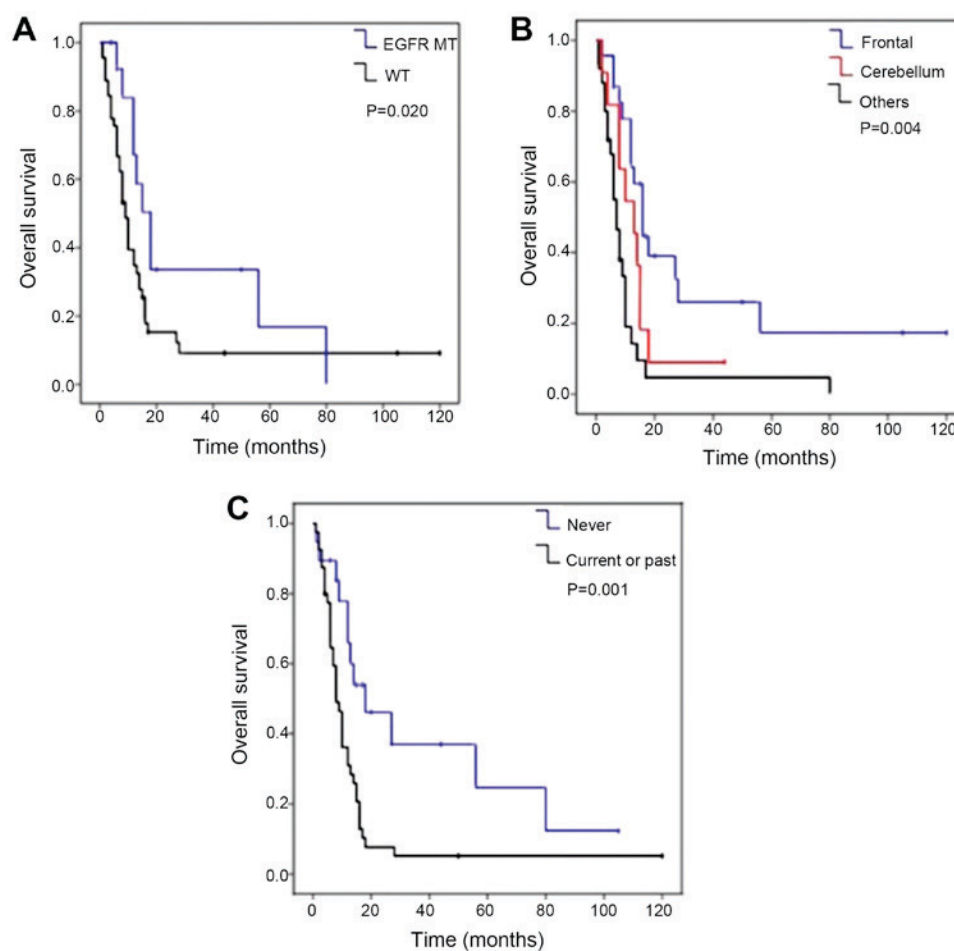
Figure 2. Kaplan-Meier curves of OS. Curves of OS were analyzed in accordance with (A) EGFR mutation status, (B) location of BM and (C) smoking. BM, brain metastases; EGFR, epidermal growth factor receptor; MT, mutation; WT, wild type; OS, overall survival.

lower compared with previously reported (19,21). This discrepancy may be due to the characteristics of the patient population in the current study. *EGFR* mutations in lung adenocarcinoma are most common in female patients and non-smokers, but the present study cohort included predominantly male patients (76.3% of all patients).

The International Association for the Study of Lung Cancer/American Thoracic Society/European Respiratory Society criteria for lung adenocarcinoma classification (30), and the 2015 WHO criteria lung tumor classification (18) recommend the identification of pathological patterns. Primary invasive lung adenocarcinoma has five architectural growth patterns, including lepidic, acinar, papillary, solid and micropapillary patterns. Tsuta *et al* (31) reported that the most prevalent subtype of adenocarcinoma was papillary-predominant (37.1%), followed by lepidic-predominant (15.1%), solid with mucin production-predominant (13.7%), acinar-predominant (10.8%) and micropapillary-predominant (6.7%) in a study of 757 cases of invasive lung adenocarcinoma (31). In a previous study of 119 patients with primary lung adenocarcinoma, the most common histopathological subtype reported was papillary-predominant (58%), followed by acinar-predominant (16%), micropapillary-predominant (9%), solid-predominant (4%) and lepidic-predominant (2%) (32). *EGFR* mutation have been reported to occur significantly more often in adenocarcinoma papillary predominant, compared with all other subtypes (31).

To the best of our knowledge, only one report has demonstrated the clinical relevance of pathological subtypes in metastatic lung adenocarcinoma (16). In this study of 100 patients with metastatic lung adenocarcinoma, the most frequent histological pattern was solid with mucin (50%), followed by acinar (29%), micropapillary (20%) and papillary (1%). This study included 30 tissue samples from BM. The study authors hypothesized that this difference in predominant components in primary and metastatic sites was based on the function of the metastatic potential of each adenocarcinoma subtype. The major solid histopathological subtype of metastatic lung adenocarcinoma was associated with poorer OS compared with other histopathological subtypes (16). Furthermore, this histopathological subtype was less likely to harbor *EGFR* mutations and occurred less frequently in never smokers (16). In a separate study, Amin *et al* (17) reported micropapillary predominant adenocarcinoma may be more likely to metastasize. The present study also identified that the most frequent subtype of BM from lung adenocarcinoma was solid predominant (57.6%). OS did not vary as a function of histological subtype. However, wild type *EGFR* was significantly more common in patients with the solid predominant subtype. This data is consistent with previous reports (30,31), but these results require validation in larger cohorts.

In recent years, TKIs targeting mutated EGFR proteins have been established as the standard treatment for NSCLC.

Table IV. Univariate analysis of overall survival with brain metastasis from lung adenocarcinoma.

Variable	No. of patients	Median OS (m)	P-value
Age			0.729
<70	45	22.7 (11.8-33.7)	
≧70	14	19.4 (9.1-29.7)	
Sex			0.131
Male	45	16.9 (11.4-22.4)	
Female	14	27.8 (15.7-39.9)	
Location			0.004
Frontal	23	28.2 (19.0-37.5)	
Cerebellum	11	13.7 (7.4-20.0)	
Others	25	12.7 (5.9-19.5)	
Smoking			0.001
Never	19	33.1 (22.1-33.1)	
Current or past	40	12.5 (8.3-16.7)	
Histological subtype			0.236
Solid	34	17.4 (11.4-23.5)	
Papillary	13	26.9 (13.0-40.8)	
Aciner	11	11.2 (5.9-16.6)	
EGFR			0.020
Mutation	14	31.5 (19.3-43.8)	
Wild type	45	15.6 (10.3-20.8)	
KPS			0.551
<70	19	18.2 (8.8-27.6)	
70-80	15	17.1 (8.3-25.9)	
81-100	25	21.9 (13.4-30.4)	
GPA			0.180
0-1.0	13	22.8 (10.0-35.6)	
1.5-2.0	24	12.8 (7.8-17.8)	
2.5-3.0	18	20.2 (11.5-28.8)	
3.5-4.0	4	37.0 (12.7-61.2)	
Timing of diagnosis			0.079
BM diagnosed before PT			
Yes	27	12.5 (8.3-16.9)	
No	32	23.4 (15.6-31.3)	
RAS			0.074
Mutation	5	7.8 (4.7-10.8)	
Wild type	54	23.7 (13.8-33.5)	
Re-operation			0.712
Present	18	17.7 (8.8-26.7)	
Absent	41	19.7 (13.3-26.1)	

EGFR, epidermal growth factor receptor; OS, overall survival; KPS, Karnofsky performance score; GPA, graded prognostic assessment; BM, brain metastasis; PT, primary tumor.

This treatment is associated with longer progression-free survival and OS compared with treatment with conventional chemotherapy (11,33), particularly in certain populations, including Eastern Asian, female and non-smoker patients with adenocarcinoma (14). In addition to demonstrating improved efficacy, TKI treatment is associated with less toxicity relative to conventional chemotherapy. Several studies have reported that the TKI treatment of patients with NSCLC with BM results in intracranial response rates ranging between 26.7 and 83% (14,15), and rates of 64-87% when combined with WBRT (34). TKI treatment for the patients with BM resulted in reduced tumor size and significantly improved OS (18.8

Table V. Multivariate analysis of overall survival with brain metastasis of lung adenocarcinoma.

Variable	HR	95% CI	P-value
Location, frontal	0.552	0.215-0.781	0.007
Never smoking	2.654	1.295-5.442	0.008
EGFR mutation	0.552	0.260-1.171	0.121

EGFR, epidermal growth factor receptor; HR, hazard ratio; CI, confidence interval.

vs. 11.1 months) (13-15). In the present study, only two patients received TKI treatment prior to surgical resection, as the majority of patients were treated prior to the development of TKIs. Excluding these two patients, there was no statistically significant difference in OS between patients with and without *EGFR* mutations (17.1 vs. 15.6 months). The presence of an *EGFR* mutation prolonged OS, but improved outcome may be due to the efficacy of treatment with TKIs. In agreement with the current results, for patients with BM from lung adenocarcinoma, the presence of *EGFR* mutations has been associated with improved survival from the time of development of BM (15-25 vs. 7 months) (35-37). However, no significant difference was observed in the timeline of BM development in patients treated with EGFR-TKI compared with those receiving conventional chemotherapy (24).

To the best of our knowledge, no previous study has reported an association between *EGFR* mutation and the site of BM. Thus, the findings in the present study of a significant association between *EGFR* mutations and BM in the frontal lobe is novel. In low-grade gliomas, lesions localized in the frontal lobe may be vigorously resected without causing persistent functional deficits (38). The frontal lobe, which does not contain vital brain regions controlling speech and motor areas, is a brain region in which relatively aggressive surgical treatment may be pursued with lower risk of causing lasting and serious functional deficits.

In patients with BM, RPA and GPA scores are still the most widely accepted variables considered in evaluating disease prognosis (6,9). However, the present study indicated that the presence of *EGFR* mutations, BM within the frontal lobe and non-smoker status are additional independent prognostic factors.

A long time period was investigated for retrospective analysis to evaluate the clinicopathological and genetic changes of BM from lung adenocarcinoma. The diagnostic neuroimaging and the treatment modalities, including WRRT, SRS and chemotherapy have been developed. Nonetheless, these results remain valid. The most common histological subtype in patients with primary lung adenocarcinoma was the papillary subtype. However, the predominant subtype of BM from lung adenocarcinoma was solid adenocarcinoma in this study. The clinicopathological characteristics of patients are not affected by the development of treatment. While the interval between detection of the primary cancer and detection of BM increased over the period covered in this study, the interval elapsed between detection of BM and the date of patient mortality or last follow-up did not change significantly.

Therefore, the prognostic factors identified in this study are reliable.

The limitations of the current study include its retrospective nature, and the potential bias in treatment strategy following resection of BM cannot be excluded. Furthermore, other clinically important genetic changes, including *ALK* rearrangement, were not assessed due to the limited number of patients with this genetic alteration in the present cohort. Despite these limitations, to the best of our knowledge, this is the first report to investigate the histopathological subtypes of BM from lung adenocarcinoma, and the association between these subtypes, *EGFR* mutations and disease prognosis in a large patient cohort.

In the present study, univariate analysis was used to demonstrated that three factors, EGFR mutation, BM localized to the frontal lobes and non-smoker status, may be used as prognostic indicators. In addition, multivariate analysis confirmed these results, and demonstrated that the presence of BM in the frontal lobes and non-smoker status were the most informative prognostic factors. However, these conclusions are primarily applicable to postoperative assessment of BM. The present study suggests that tissue from the BM may be useful as an alternative tissue for analysis, as resectable BM from lung adenocarcinoma cases exhibit characteristics that are similar to those of the primary tumor tissue. TKI treatment for BM from lung adenocarcinoma should be considered if indicated by histopathological and genetic examination.

Availability of data and materials

All data generated and/or analyzed during this study are included in this published article.

Authors' contributions

HK and MH analyzed statistics, and drafted the manuscript. HK, MH, MY HA and MN analyzed and interpreted the patient data. TM TI and KN contributed to the conception of the study, critically reviewed the manuscript and supervised the study. All authors read and approved the final manuscript.

Ethics approval and consent to participate

Written informed consent was obtained, and the study design was approved by our institutional review board (IRB protocol #15-5-14).

Patient consent for publication

Patients provided written informed consent for the publication of any data and associated images.

Competing interests

The authors declare that they have no competing interests.

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