Original Article

Expression and prognostic significance of microsomal triglyceride transfer protein in brain tumors: a retrospective cohort study

Soo Min Son[1,2#], Hye Sun Lee[1#], Jeongsu Kim[3,4*], Ryuk Jun Kwon[1,2*]

[1]Family Medicine Clinic and Research Institute of Convergence of Biomedical Science and Technology, Pusan National University Yangsan Hospital, Yangsan, Korea; [2]Department of Family Medicine, Pusan National University School of Medicine, Yangsan, Korea; [3]Division of Cardiology, Department of Internal Medicine, Pusan National University Yangsan Hospital, Yangsan, Korea; [4]Division of Cardiology, Department of Internal Medicine, Pusan National University School of Medicine, Yangsan, Korea

Contributions: (I) Conception and design: SM Son, J Kim, RJ Kwon; (II) Administrative support: None; (III) Provision of study materials or patients: SM Son, HS Lee, RJ Kwon; (IV) Collection and assembly of data: J Kim, HS Lee; (V) Data analysis and interpretation: SM Son, J Kim, RJ Kwon; (VI) Manuscript writing: All authors; (VII) Final approval of manuscript: All authors.

[#]These authors contributed equally to this work as co-first authors.

[*]These authors contributed equally to this work.

Correspondence to: Jeongsu Kim, MD, PhD. Division of Cardiology, Department of Internal Medicine, Pusan National University Yangsan Hospital, 20, Geumo-ro, Mulgeum-eup, Yangsan 50612, Korea; Division of Cardiology, Department of Internal Medicine, Pusan National University School of Medicine, 20, Geumo-ro, Mulgeum-eup, Yangsan 50612, Korea. Email: jeongsukim@lycos.co.kr; Ryuk Jun Kwon, MD, PhD. Family Medicine Clinic and Research Institute of Convergence of Biomedical Science and Technology, Pusan National University Yangsan Hospital, 20, Geumo-ro, Mulgeum-eup, Yangsan 50612, Korea; Department of Family Medicine, Pusan National University School of Medicine, 20, Geumo-ro, Mulgeum-eup, Yangsan 50612, Korea. Email: brain6@hanmail.net.

Background: Glioblastoma (GBM) is the most common malignant brain tumor and has poor survival. An elevated cholesterol level is involved occurrence and progression of brain tumors. Microsomal triglyceride transfer protein (MTTP) is a target for lowering lipids, and its inhibition helps to improve hyperlipidemia. However, whether the altered expression of *MTTP* affects the development and prognosis of brain tumors is currently unidentified. The purpose of this study is to determine *MTTP* as a prognostic marker for brain tumors.

Methods: Data for patients with brain cancers and control brain tissue were acquired from The Cancer Genome Atlas (TCGA) and Gene Expression Omnibus (GEO). The datasets were analyzed using Mann-Whitney *U*-test or *t*-test to compare the expression of *MTTP* in normal and brain tumor tissues. To examine whether *MTTP* affected the prognosis of patients with brain tumors, log-rank test and multivariable Cox proportional hazard regression were conducted.

Results: The expression of *MTTP* was significantly upregulated in brain tumors and was correlated with age, tumor stage, and isocitrate dehydrogenase (*IDH*) mutation. Importantly, increased *MTTP* expression in brain tumors is associated with poor patient survival.

Conclusions: High *MTTP* expression is associated with brain tumor development, tumor stage, and prognosis. Therefore, *MTTP* is an independent prognostic indicator for brain tumors, which can serve as one of the possible targets for adjuvant treatment of GBM.

Keywords: Brain neoplasm; cholesterol; glioblastoma (GBM); lomitapide; microsomal triglyceride transfer protein (MTTP)

Submitted Dec 12, 2023. Accepted for publication Apr 10, 2024. Published online May 28, 2024.
doi: 10.21037/tcr-23-2286
View this article at: https://dx.doi.org/10.21037/tcr-23-2286

Introduction

Glioblastoma (GBM), which originates from astrocytes, is the most common central nervous system (CNS) tumors, explaining 55% of all gliomas and 45.2% of malignant brain tumors (1,2). GBM is associated with significant mortality (3) and the 5-year survival rate of GBM is only 3–4% despite therapeutic advances (4). The fact indicates that understanding the pathogenesis of GBM is important for effective treatment. The significance of molecular markers in diagnosis has been emphasized by the World Health Organization (WHO) CNS5 classification of 2021 (5). The most significant alterations are related to diffuse gliomas, in which isocitrate dehydrogenase (*IDH*) status have gained importance (6,7). *IDH* wild type (WT) is referred to grade 4 in 2021 WHO CNS5 classification. Even though it has low-grade features histologically, *IDH*-WT glioma is considered grade 4 GBM. *IDH*-WT GBM has a lower survival rate compared to *IDH*-mutant type (15 *vs.* 31 months) (8). Therefore, new approaches to GBM are needed to better understand its molecular properties and improve survival.

Cholesterol has received increasing attention because of its function in cancer. Studies have shown that the altered cholesterol metabolism is associated with cancer development and progression (9). High serum cholesterol levels have been reported to increase the incidence of developing cancers, and cholesterol-lowering agents have beneficial effects by reducing the risk of mortality from colorectal, breast, and prostate cancers (10,11). It has been found that the cholesterol pathway is upregulated in patients with GBM and that cholesterol biosynthesis-related genes are downregulated in densely plated normal astrocytes (12). David *et al.* showed that the hazard ratios for death due to GBM were significantly reduced by using high-intensity statins, the most common lipid-lowering drugs (13). Therefore, targeting the regulation of cholesterol metabolism may be a potential strategy for GBM treatment.

Microsomal triglyceride transfer protein (MTTP), a target of lomitapide which was approved by Food and Drug Administration [2012], is a principal cellular protein that transfers neutral lipids between membrane vesicles and is a target molecule to treat diseases that produce high apolipoprotein B (apoB) lipoproteins such as familial combined hyperlipidemia, hypertriglyceridemia, and atherosclerosis (14,15). The main site of MTTP expression is the small intestine and the liver and MTTP is expressed in neurons as well (15). A study reported that *Mttp* intestinal knock-out mice showed increased tumor formation (16). However, it is unidentified if the change of *MTTP* expression associates with the occurrence of CNS tumors and prognosis of patients with CNS tumors.

In this study, the comparison of *MTTP* expression between normal and brain tumors was evaluated, and the correlation between the *MTTP* expression and clinical information in CNS tumors was also examined. In addition, the effects of *MTTP* on the survival rate of patients with brain tumors were assessed, and the impact of *MTTP* as a prognostic marker of brain tumors was determined when compared with other prognostic factors. We present this article in accordance with the REMARK reporting checklist (available at https://tcr.amegroups.com/article/view/10.21037/tcr-23-2286/rc).

Highlight box

Key findings

- The expression of microsomal triglyceride transfer protein (*MTTP*) was significantly increased in brain tumors and was associated with age, tumor stage, and the presence of an isocitrate dehydrogenase (*IDH*) mutation. Increased expression of *MTTP* in brain tumors is notably linked to poorer patient survival.

What is known and what is new?

- *MTTP* plays a pivotal role in shuttling neutral lipids between membrane vesicles and *IDH* enzymes are essential for the oxidative decarboxylation of isocitrate.
- MTTP is mainly expressed in the small intestine and liver, and is also expressed in neurons. *MTTP* knock-out mice showed increased colonic tumor formation.
- *MTTP* is overexpressed in brain tumors and that high *MTTP* expression is correlated with a poor prognosis, suggesting that *MTTP* is an independent prognostic marker in brain tumors.

What is the implication, and what should change now?

- MTTP, the target of lomitapide, may be used as a screening marker to individually manage brain tumor patients with poor prognosis.
- The results of the current study can be used to investigate potential therapeutic targets for the adjuvant therapy of brain tumors.

Methods

Patients and data collection

This study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). The Cancer Genome Atlas (TCGA) database includes clinical information of a large number of patients and also contains information on RNA expression, DNA methylation, and miRNAs in several

Table 1 Clinical characteristics of patients with brain tumors

Patient characteristics	Value (N=665)
Age, years	
≤60	505 (75.9)
>60	110 (16.5)
Unknown	50 (7.5)
Sex	
Male	346 (52.0)
Female	269 (40.5)
Unknown	50 (7.5)
Race	
Caucasian	567 (85.3)
Others	37 (5.6)
Unknown	61 (9.2)
Tumor stage	
LGG	513 (77.1)
GBM	152 (22.9)
Overall survival months	27.62±29.34

Data are presented as n (%) or mean ± SD. LGG, low-grade glioma; GBM, glioblastoma; SD, standard deviation.

types of cancers. TCGA data of 1,099 patients with brain tumors were downloaded, as previously described (17). Among them, 434 patients were excluded owing to a lack of information on *MTTP* expression in 432 patients and clinical data of two patients. Finally, clinical data corresponding to 665 patients with brain tumors was included for prognosis analysis in this study.

Age, sex, race, histological type, overall survival (OS), progression-free survival (PFS), disease-specific survival (DSS), and *MTTP* expression were selected as the clinical attributes. Histological types were divided into low-grade glioma (LGG) and GBM based on the tumor stage. OS refers to the period from the initial day of diagnosis of brain tumors to death from any cause or the last follow-up. PFS refers to the time from the initiation of the treatment to the initial stage of cancer acceleration or relapse for any reason. DSS refers to the period from the day of diagnosis to death due to brain tumors compared to other causes.

MTTP expression datasets of normal tissues corresponding to brain tumor tissues were extracted from the database of the Genome Data Analysis Center. The Gene Expression Omnibus (GEO) is a public genomics database that includes array- and sequence-based data. The GSE50161 dataset was retrieved from GEO to investigate whether the expression of *MTTP* is higher in brain tumors than in normal tissues and the GSE4271 dataset was used to confirm the prognostic significance of *MTTP* in brain tumors.

Definition of brain tumors in TCGA

Brain tumor samples included in TCGA were collected between 1989 and 2013. All cases were diagnosed according to WHO guidelines and were classified into only two groups based solely on histopathological characteristics in this study. Low-grade glioma (LGG) was defined as grades II and III and GBM was defined as grade IV according to the histopathological classification of primary brain tumors.

Statistical analyses

The comparison of the *MTTP* expression values between normal and brain tumors (LGG and GBM) was examined as the Mann-Whitney *U*-test or the Student's *t*-test after applying the Shapiro-Wilk normality test. A Chi-squared formula was used to assess the relation between *MTTP* expression and patient's characteristics. For Kaplan-Meier (KM) survival plots, subjects with brain tumors were classified into two groups (low *MTTP* and high *MTTP* expression) depending on the median of *MTTP* expression. To calculate P values, a log-rank test was performed. Univariable and multivariable Cox proportional hazard regression analyses were conducted to investigate whether *MTTP* was a significant marker of OS, PFS, and DSS.

Box and whisker plots were generated using Microsoft Excel. IBM SPSS Statistics for Windows version 21 (IBM Corp., Armonk, NY, USA) was used for KM plots and statistical analyses. MedCalc (version 22.016) was conducted for obtaining number at risk. P value less than 0.05 is considered significant. To determine the association between immune cell infiltration and *MTTP* expression, TIMER 2.0 was used (http://timer.cistrome.org/).

Results

Clinical characteristics

The clinical information of patients with brain tumors was represented (*Table 1*). Of the 665 participants, 505 (75.9%)

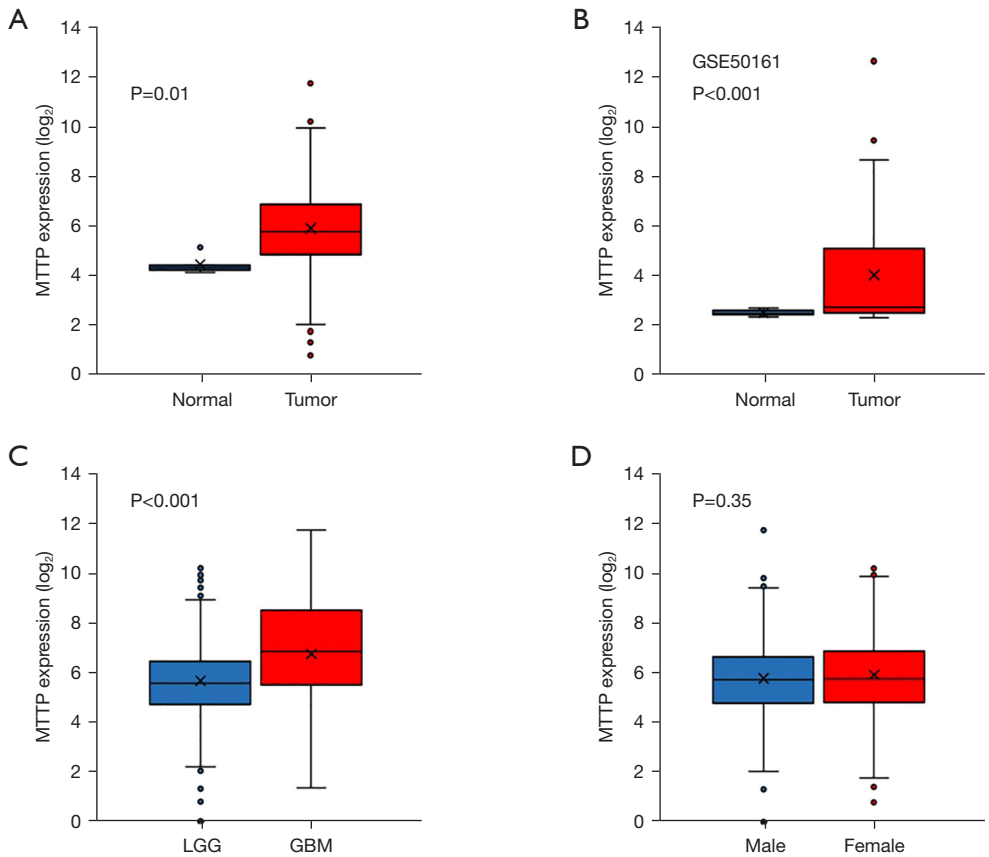

Figure 1 *MTTP* expression in normal and brain tumor tissues. (A) The mean ± SD of *MTTP* expression in normal (n=5) and brain tumor (n=665) was 4.45±0.406 and 5.92±1.670, respectively (P=0.01). (B) *MTTP* expression in normal (n=13) and brain tumor (n=117) tissues (GSE50161). The mean ± SD of *MTTP* expression in normal and brain tumor tissues was 2.54±0.114 and 4.06±2.127, respectively (P<0.001). (C) *MTTP* expression level in LGG (n=513) and GBM (n=152). The mean ± SD of *MTTP* expression in LGG and GBM was 5.67±1.477 and 6.75±1.995, respectively. (D) *MTTP* expression in males and females. The number of males and females was 346 and 269, respectively. The mean ± SD of *MTTP* expression in males and females was 5.81±1.628 and 5.94±1.665, respectively (P=0.35). *MTTP*, microsomal triglyceride transfer protein; LGG, low-grade glioma; GBM, glioblastoma; SD, standard deviation.

were ≤60 years old, 110 (16.5%) were >60 years old, and the age of 50 (7.5%) patients was unknown. The sex proportion was 52.0% for males and 40.5% for females, and the sex of 7.5% of the patients was unknown. The proportion of white people was higher (n=567, 85.3%) than non-white people (5.6%). A higher number of patients had LGG (77.1%) than GBM (22.9%). The mean OS of patients with brain tumors was 27.62±29.34 months.

Expression level of MTTP in normal brain tissue, LGG, and GBM

MTTP expression was higher in brain cancers than in normal samples (normal tissues: 4.45±0.406, brain tumors: 5.92±1.670, P=0.01) (*Figure 1A*). The GEO dataset (GSE50161) was analyzed to determine if *MTTP* expression levels corresponding to normal tissues and brain tumors were consistent with the results obtained using TCGA database (*Figure 1B*). As a result, *MTTP* expression in brain tumors was significantly elevated than that in normal samples. Importantly, the expression of *MTTP* in GBM was significantly higher than that in LGG (LGG: 5.67±1.477, GBM: 6.75±1.995, P<0.001) (*Figure 1C*). However, *MTTP* expression levels did not differ between males and females (males: 5.81±1.628, females: 5.94±1.665, P=0.35) (*Figure 1D*).

Table 2 Correlation between *MTTP* expression and clinical characteristics of patients with brain tumors

Characteristic	*MTTP* expression		
	Low	High	P value
Age (years) (n=615)			0.003
≤60	272	233	
>60	42	68	
Sex (n=615)			0.83
Male	178	168	
Female	136	133	
Race (n=604)			0.31
Caucasian	294	273	
Others	16	21	
Tumor stage (n=665)			<0.001
LGG	287	226	
GBM	45	107	

MTTP, microsomal triglyceride transfer protein; LGG, low-grade glioma; GBM, glioblastoma.

Correlation between MTTP expression and clinical information

To assess the relation between *MTTP* expression and patient information, patients with brain tumors were classified into the low- and the high-expression groups based on the level of *MTTP* expression. High expression of *MTTP* was significantly associated with older age and higher tumor stage but not with sex or race (*Table 2*).

OS, PFS, and DSS for MTTP expression groups

To investigate the value of *MTTP* expression as a prognostic factor for GBM, KM survival analysis was conducted for the *MTTP*-high and *MTTP*-low expression groups against OS (*Figure 2*, *Table 3*). The median OS of the high *MTTP* expression was lower than that of the low *MTTP* expression in the total cohort (*MTTP*-high: 36.82±6.612 months, *MTTP*-low: 75.02±8.096 months, P<0.001) (*Figure 2A*). In LGG, the OS was 98.24±21.862 months for low *MTTP* expression group and was 73.48±17.560 months for high *MTTP* expression group (P=0.03) (*Figure 2B*). Similarly, the median OS of the high *MTTP* expression group was lower than that of the low *MTTP* expression group in GBM (*MTTP*-high: 11.28±1.113 months, *MTTP*-low: 14.93±0.847 months, P=0.008) (*Figure 2C*). To determine whether *MTTP* expression had a more significant effect on the prognosis of LGG and GBM, KM plots were plotted for the high and low *MTTP* expression group against PFS and DSS. The PFS of high *MTTP* expression group was less than that of low *MTTP* expression group (*MTTP*-high: 18.15±2.335 months, *MTTP*-low: 39.62±4.909 months, P<0.001) (*Figure 2D*). There was no difference between the high and low *MTTP* expression group in LGG (P=0.08) (*Figure 2E*). The PFS of high *MTTP* expression group was significantly decreased, as compared to that of low *MTTP* expression group in GBM (*MTTP*-high: 5.16±0.663 months, *MTTP*-low: 7.59±0.739 months, P=0.02) (*Figure 2F*). The median DSS of the high *MTTP* expression group was lower than that of the low *MTTP* expression group (*MTTP*-high: 40.54±7.345 months, *MTTP*-low: 78.21±10.468 months, P=0.02) (*Figure 2G*). In LGG, the DSS of the high and low *MTTP* expression groups was estimated to be 87.45±14.980 months and 98.24±21.099 months, respectively (P=0.02) (*Figure 2H*). Similarly, the median DSS of high *MTTP* expression group was less than that of low *MTTP* expression group in GBM (*MTTP*-high: 11.28±1.113 months, *MTTP*-low: 15.78±0.795 months, P=0.02) (*Figure 2I*). Next, GSE4271 was analyzed to confirm whether patients with high *MTTP* expression in brain tumors had poorer survival (*Figure 3*). The result revealed that the median OS of the high *MTTP* expression group was lower than that of the low *MTTP* expression group (*MTTP*-high: 62.00±4.151 months, *MTTP*-low: 150.00±60.325 months, P=0.001).

Relationship between MTTP expression and prognosis

To identify the prognostic importance of *MTTP*, the Cox proportional hazard regression were conducted. With the *MTTP* expression, factors known to affect prognosis, such as matrix metallopeptidase 2 (*MMP2*), insulin-like growth factor binding protein 2 (*IGFBP2*), phosphatase and tensin homolog (*PTEN*), and marker of proliferation Ki-67 (*MKI67*) were evaluated. Univariable analysis showed that age, tumor stage, expression levels of *IGFBP2*, *MKI67*, *PTEN*, and *MTTP* were significantly related with OS (*Table 4*). Similarly, the multivariable analyses showed that age, tumor stage, expression levels of *IGFBP2*, *MKI67*, *PTEN*, and *MTTP* were significantly related with OS (*Table 4*). The hazard ratios for age, stage, *IGFBP2*, *MKI67*, *PTEN*, and *MTTP* were 2.875, 2.773, 2.947, 1.536, 0.556, and 1.369, respectively.

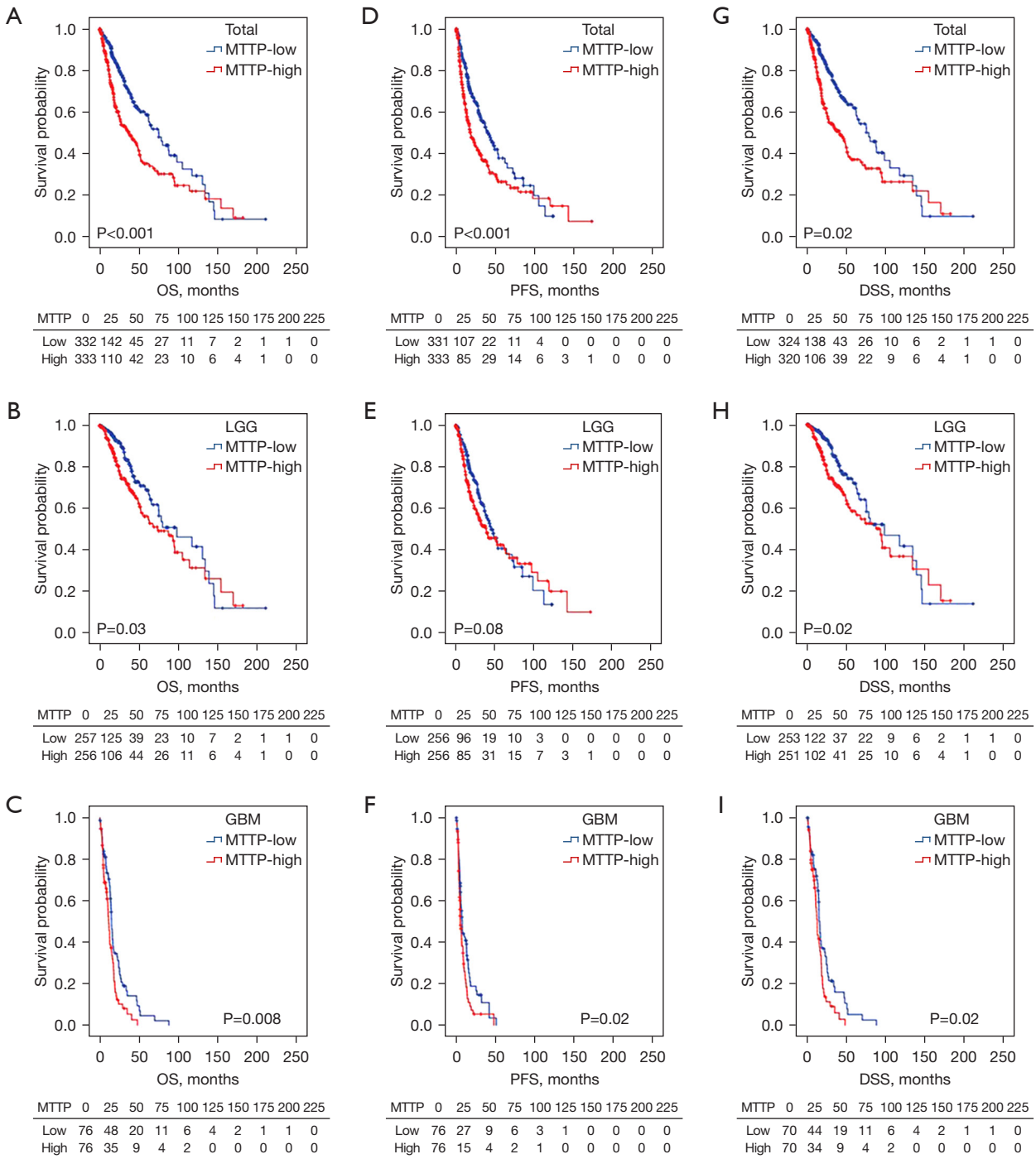

Figure 2 OS, PFS, and DSS based on *MTTP* expression. Kaplan-Meier analysis was performed for *MTTP* low- and high-expression groups for OS, PFS, and DSS. Blue line: low *MTTP* expression; red line: high *MTTP* expression. (A) The median OS of *MTTP* high and low. (B) In LGG, the median OS of *MTTP* high and low. (C) In GBM, the median OS of *MTTP* high and low. (D) The median PFS of *MTTP* high and low. (E) In LGG, the median PFS of *MTTP* high and low. (F) In GBM, the median PFS of *MTTP* high and low. (G) The median DSS of *MTTP* high and low. (H) In LGG, the median DSS of *MTTP* high and low. (I) In GBM, the median DSS of *MTTP* high and low. *MTTP*, microsomal triglyceride transfer protein; OS, overall survival; PFS, prognostic free survival; DSS, disease-specific survival; LGG, low-grade glioma; GBM, glioblastoma.

Table 3 Median OS, PFS, and DSS based on *MTTP* expression in total cohort, LGG, and GBM

Cohort	*MTTP*	OS			PFS			DSS		
		N	Median OS (mean ± SD), months	P value	N	Median PFS (mean ± SD), months	P value	N	Median DSS (mean ± SD), months	P value
Total	Low	332	75.02±8.096	<0.001	331	39.62±4.909	<0.001	324	78.21±10.468	<0.001
	High	333	36.82±6.612		333	18.15±2.335		320	40.54±7.345	
LGG	Low	257	98.24±21.862	0.03	256	45.17±5.133	0.08	253	98.24±21.099	0.02
	High	256	73.48±17.560		256	38.93±7.759		251	87.45±14.980	
GBM	Low	76	14.93±0.847	0.008	76	7.59±0.739	0.02	70	15.78±0.795	0.02
	High	76	11.28±1.113		76	5.16±0.663		70	11.28±1.113	

OS, overall survival; PFS, prognostic free survival; DSS, disease-specific survival; *MTTP*, microsomal triglyceride transfer protein; LGG, low-grade glioma; GBM, glioblastoma; SD, standard deviation.

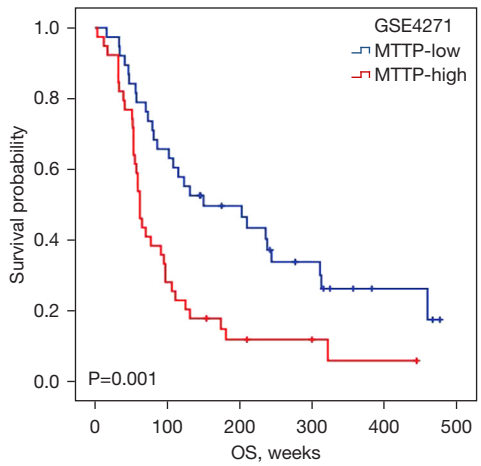

Figure 3 OS based on *MTTP* expression (GSE4271 dataset). Kaplan-Meier analysis of the *MTTP* low- and high-expression groups for OS. The median OS of *MTTP* high- (red, n=39) and low- (blue, n=38) groups was 62.00±4.151 and 150.00±60.325 weeks (P=0.001), respectively. OS, overall survival; *MTTP*, microsomal triglyceride transfer protein.

Survival analysis of the combined prognostic factors

KM analysis was performed using a combination of age and *MTTP* expression in the total cohort and the LGG and GBM patient sub-groups (*Figure 4*, *Table 5*). For the total cohort, the OS was decreased significantly with older age (median OS of aged patients >60/≤60 years: 15.95±1.996/78.21±10.865 months, P<0.001) (*Figure 4A*). Survival curves produced by a combination of *MTTP* expression and age, showed that the median of older age with high *MTTP* expression was 12.56±0.876 months, and these patients showed the worst prognosis (P<0.001) (*Figure 4B*). In addition, the mixed group, i.e., younger patients with high *MTTP* expression or older patients with low *MTTP* expression showed moderate prognosis (median OS: 50.14±5.349 months, P<0.001). In the LGG group, the median OS was 94.52±10.984 months in younger patients, and 23.90±6.085 months in older patients (P<0.001) (*Figure 4C*). Survival curves produced by a combination of age and *MTTP* expression showed that the median OS of older age with high *MTTP* expression was 18.44±5.585 months, and these patients showed the worst prognosis in LGG (P<0.001) (*Figure 4D*). Similarly, in patients with GBM, median OS was decreased with increasing age, as in LGG (median OS of older/younger patients: 11.01±2.080/16.64±1.392 months, P=0.006) (*Figure 4E*). After combination with the level of *MTTP* expression, the median OS of older age with high *MTTP* expression was 8.84±3.190 months, which was the worst prognosis among all groups, similar to the pattern above (P=0.02) (*Figure 4F*).

MTTP expression in IDH-WT and mutant

MTTP expression was significantly higher in *IDH*-WT than that in *IDH*-mutant (262.20±334.932 *vs.* 56.47±52.671, P=0.01) (*Figure 5*).

Discussion

MTTP was first discovered as a major molecule that transfers neutral lipids from ER membranes to apoB-lipoproteins and is a target of lomitapide, a medicine used to treat patients with abetalipoproteinemia. However, the

Table 4 Univariable and multivariable analyses of prognostic factors for overall survival in patients with brain tumors (N=615)

Factors	Univariable analysis		Multivariable analysis	
	HR (95% CI)	P value	HR (95% CI)	P value
Age >60 years (*vs.* ≤60)	5.349 (3.916–7.306)	<0.001	2.875 (2.049–4.033)	<0.001
Stage GBM (*vs.* LGG)	9.897 (7.142–13.716)	<0.001	2.773 (1.848–4.159)	<0.001
MTTP high (*vs.* low)	1.834 (1.379–2.441)	<0.001	1.369 (1.011–1.854)	0.04
IGFBP2 high (*vs.* low)	5.704 (4.099–7.938)	<0.001	2.947 (2.017–4.305)	<0.001
MMP2 high (*vs.* low)	2.509 (1.870–3.367)	<0.001	0.992 (0.701–1.402)	0.96
MKI67 high (*vs.* low)	2.722 (2.008–3.690)	<0.001	1.536 (1.103–2.140)	0.01
PTEN high (*vs.* low)	0.349 (0.260–0.467)	<0.001	0.556 (0.401–0.771)	<0.001

HR, hazard ratio; CI, confidence interval; GBM, glioblastoma; LGG, low-grade glioma; *MTTP,* microsomal triglyceride transfer protein; *IGFBP2*, insulin-like growth factor binding protein 2; *MMP2*, matrix metallopeptidase 2; *MKI67*, marker of proliferation Ki-67; *PTEN*, phosphatase and tension homolog.

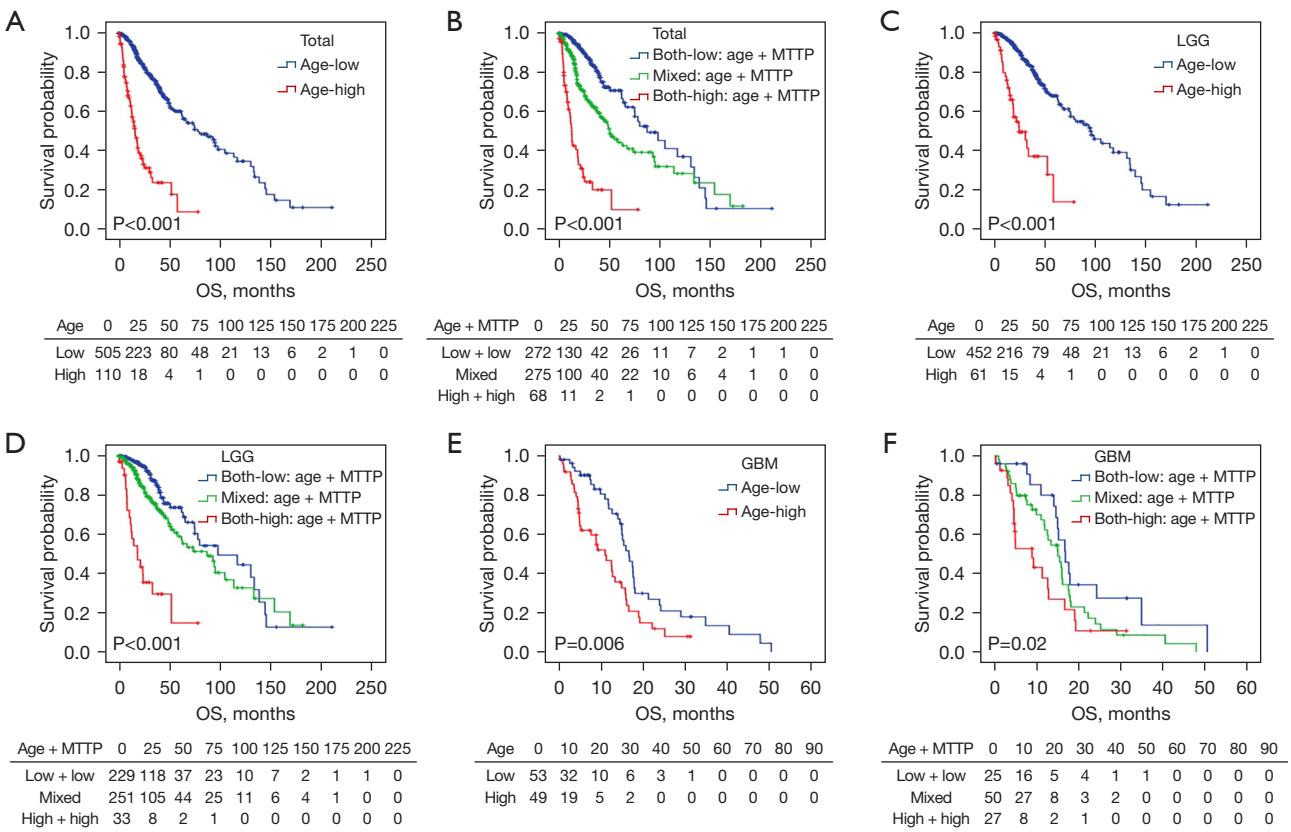

Figure 4 Kaplan-Meier analysis after a combination of age and *MTTP* expression level in LGG and GBM. Survival analyses were performed after age (low: ≤60 years *vs.* high: >60 years) and *MTTP* (low *vs.* high expression) groups were divided into three subgroups, blue line: low age + low *MTTP*; green line (mixed): low age + high *MTTP* or high age + low *MTTP*; red line: high age + high *MTTP*. (A) The median OS of the low- and high-aged groups. (B) The median OS of the low-age + low *MTTP*, high-age + high *MTTP*, and mixed groups. (C) In LGG, the median OS of the low- and high-aged groups. (D) In LGG, the median OS of the low-age + low *MTTP*, high-age + high *MTTP*, and mixed groups. (E) In GBM, the median OS of the low- and high-aged groups. (F) In GBM, the median OS of the low age + low *MTTP*, high age + high *MTTP*, and mixed groups. *MTTP*, microsomal triglyceride transfer protein; LGG, low-grade glioma; GBM, glioblastoma; OS, overall survival.

Table 5 Median OS after a combination of age and *MTTP* expression level in total cohort, LGG, and GBM

Cohort	Age				Age + *MTTP*			
	Level	N	Median OS (mean ± SD), months	P value	Level	N	Median OS (mean ± SD), months	P value
Total	Low	505	78.21±10.865	<0.001	Low + low	272	87.45±12.541	<0.001
	High	110	15.95±1.996		Mixed	275	50.14±5.349	
	–				High + high	68	12.56±0.876	
LGG	Low	452	94.52±10.984	<0.001	Low + low	229	87.24±24.631	<0.001
	High	61	23.90±6.085		Mixed	251	87.45±15.734	
	–				High + high	33	18.44±5.585	
GBM	Low	53	16.64±1.392	0.006	Low + low	25	16.77±1.694	0.02
	High	49	11.01±2.080		Mixed	50	14.96±1.751	
	–				High + high	27	8.84±3.190	

Age (low: ≤60 years *vs.* high: >60 years); *MTTP* (low *vs.* high expression). OS, overall survival; *MTTP*, microsomal triglyceride transfer protein; LGG, low-grade glioma; GBM, glioblastoma; SD, standard deviation.

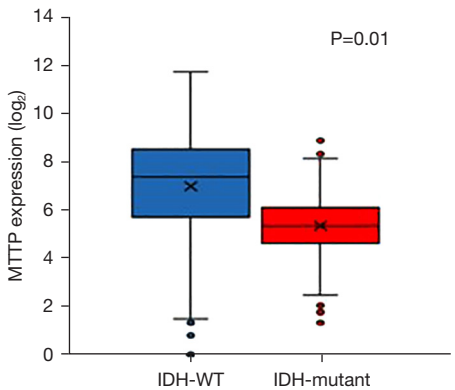

Figure 5 *MTTP* expression in *IDH*-WT and *IDH*-mutant brain tumors. *MTTP* expressions in *IDH*-WT *vs. IDH*-mutant brain tumors are shown as a box and whisker plot. The mean ± standard deviation of *MTTP* expression in WT (n=197) and *IDH* mutant (n=419) was 262.20±334.932 and 56.47±52.671, respectively (P=0.01). *IDH*, isocitrate dehydrogenase; *IDH*-WT, *IDH*-wild type; *MTTP*, microsomal triglyceride transfer protein.

expression of *MTTP* and its prognostic significance in brain tumors have been poorly characterized.

Intestine-specific *Mttp* deletion in mice increased the tumor burden of colorectal cancer, which was associated with increased colorectal inflammation as well as changes in cytokine expression (16). Moreover, low *MTTP* expression has been correlated with poor recurrence-free survival in breast cancer (18). In contrast, in this study, *MTTP* expression was increased in the order of normal, LGG, and GBM and also was associated with the stage and age of patients with brain tumors (*Figure 1*, *Table 2*), suggesting that the function of *MTTP* was altered depending on kinds of cancer. For instance, high Notch signaling is associated with tumor grade and metastasis, but it has an inhibitory effect on GBM (19,20). Therefore, the role of *MTTP* in the occurrence and progression of cancers may be dependent on the cancer type.

Abnormal lipid metabolism plays important roles in the proliferation, migration, invasion, and angiogenesis of cancers (21,22). The brain, the most cholesterol-rich organ, consists of 23% total body cholesterol, and GBM also requires lipids for cell survival (23,24). GBM accumulates more fatty acids than the surrounding normal brain tissue and uses lipids as energy reservoirs (25,26). These lipid stores promote GBM proliferation and are maintained to avoid oxidative damage and lipotoxicity (27). Thus, it is worthwhile to investigate the association between the molecular targets involved in lipid metabolism and the prognosis of GBM. In this study, patients with high *MTTP* expression showed a significantly shorter OS, PFS, and DSS than those with low *MTTP* expression, except for median PFS in LGG (*Figure 2*, *Table 3*). Those results suggest that *MTTP* expression has a prognostic importance in brain tumors, especially GBM.

IGFBP2, which acts as the antagonist of the insulin-like growth factor (IGF) signal involved in tumor suppression,

increases the proliferation and invasiveness of gliomas (28,29). In a study, the median survival of patients with low *IGFBP2* mRNA expression was 16.9 months while that of patients with high *IGFBP2* mRNA expression was only 11.6 months (30). KM analysis revealed a shorter survival time with a higher level of *MMP2*, which was also associated with brain tumors and metastasis (31). MKI67 is a non-histone nuclear protein that enters the mitotic cycle and is positively related to histological tumor grade in gliomas (32,33). Chen *et al.* reported that high levels of MKI67 expression are associated with poor OS in gliomas (34). PTEN is a tumor suppressor whose expression in glioma cells suppresses growth and inhibits migration and dissemination (35,36). Phillips *et al.* showed that low *PTEN* mRNA expression is associated with poor survival (37). Consistent with the reported studies, the present study reported *IGFBP2*, *MMP2*, *MKI67*, and *PTEN* as significant indicators of OS (*Table 4*). Importantly, *MTTP* expression level was also significantly associated with OS (*Table 4*). Thus, these findings suggest that *MTTP* is a prognostic factor in brain tumors.

Age and tumor stage are known as prognostic factors correlated with survival in LGG and GBM (38). According to the literature, the median survival of patients with LGG ranged from 5.6 to 13.3 years depending on specific histological and molecular characteristics, and the median survival of GBM was reported to be only 6 to 10 months (39,40). Since the survival period of patients with GBM is short, it is important to identify and manage patients with poor prognoses. Moreover, genes related to lipid metabolism have recently been identified as potential targets for the treatment of GBM and brain metastasis (25). For example, the activation of the liver X receptor (LXR) by LXR-623 (LXR agonist) decreased intracellular cholesterol levels and selectively killed GBM cells with improved survival in a mouse model (41). The survival period of mice treated with both temozolomide (TMZ) and lomitapide, which is supposed to pass through the blood-brain barrier, was increased by 1.34 times compared to the control group and by 1.14 times compared to mice treated with only TMZ (42). In this study, high *MTTP* expression is correlated with a higher brain tumor stage and higher *MTTP* expression with older patients showed the worst prognosis in all the groups (the total cohort, LGG, and GBM) (*Tables 2*,*3*,*5*, *Figures 2*,*4*). Those results suggest that *MTTP* can serve as a screening marker for GBM patients with poorer prognosis and may be a possible target for improving the prognosis of GBM patients.

The WHO classification of CNS tumors published in 2021, relied more heavily on molecular tests for diagnosis and grading than in 2016 with certain molecular markers providing strong prognostic information (8). Since the data used in the analysis were prepared before the 2021 WHO classification of brain tumors was announced, there may be differences from the current staging results. Nevertheless, *IDH* type is still an important factor in brain tumor prognosis. Previous studies showed that *IDH*-WT had a significantly poorer prognosis than *IDH*-mutant in CNS tumors (median survival of *IDH*-WT: 15 months, *IDH*-mutant: 31 months) (43). IDH plays several roles in cellular function such as lipogenesis, glucose sensing, and regulation of cellular redox status (44). Contrary to *IDH*-mutant, IDH1 and IDH2 (IDH-WT) increase NADPH/$NADP^+$ which facilitates lipid biosynthesis and promotes cellular defense against reactive oxidative stress, and reduces the effect of chemotherapy and radiotherapy in GBM (45). In the present research, *MTTP* expression in brain tumors was significantly higher in the *IDH*-WT than in the *IDH*-mutant (*Figure 5*). In general, more than 90% of GBM is *IDH*-WT although there are several GBM with poor prognosis despite being *IDH*-mutant (46). Taken together, these results suggest that the role of MTTP in the lipid metabolism for cell proliferation of brain tumors may be associated with *IDH* status.

Strengths and limitations

The present study has some strengths. First, this is the first research to identify *MTTP* as an independent prognostic marker of patients with brain tumors. Thus, clinicians may use *MTTP* expression as a marker of poor prognosis for clinical management. Second, this is a large-scale study including *MTTP* expression and clinical data of patients with CNS tumors. Third, the research provides a basis for future investigation to improve the survival rate of patients with GBM using lomitapide.

Despite its strength, the present study has a few limitations. First, there was no patient information on risk factors including obesity, diabetes, history of smoking, and alcohol consumption. Second, this dataset does not contain information about spatial distribution, characteristics of *MTTP* expressing cells and the origin of brain tumor cells. Tumor purity is defined as the percent of cancer cells in the admixture. TCGA claims that 60% purity is sufficient to distinguish cancer signals from other cells and the purities of LGG and GBM in the TCGA database were more than 80% according to a systematic pan-cancer analysis of tumor

purity (47). Thus, in this study, *MTTP* expression in LGG and GBM may be distinct from other cells. In addition, interestingly, it was found that the association between mRNA and protein expression was high in cancer cell lines (GBM, osteosarcoma, and epidermoid carcinoma) from 0.58 to 0.63 (48). If there is a high relation between mRNA and protein expression in cancers, MTTP protein expression in brain tumors may be high relative to the transcript of *MTTP*. Finally, there was no experimental evidence that high *MTTP* expression affects brain tumor development and progression. Therefore, further experimental investigation is needed to prove the findings of this study.

Conclusions

In conclusion, the present study showed that high *MTTP* expression in brain tumors including GBM is correlated with poor survival. Thus, *MTTP* is an independent prognostic indicator in brain tumor patients which may serve as a predictor for managing patients with brain tumors, thereby improving their OS. In addition, the study showed that *MTTP* is upregulated in brain tumors and is correlated with the tumor stage and *IDH* status. These findings provide a framework for experimental studies on the function of *MTTP* in the occurrence and progression of brain tumors and for studying the possibility of using lomitapide as adjuvant treatment for GBM through drug repositioning.

Acknowledgments

The authors wish to acknowledge the 'The Cancer Genome Atlas' (TCGA) database and the tools provided by the TCGA database.

Funding: This study was supported by a National Research Foundation of Korea (NRF) grant funded by the Korean government (MSIT) (No. NRF-2022R1F1A1074769; R.J.K.) and the Research Institute for Convergence of Biomedical Science and Technology (No. 20-2023-002), Pusan National University Yangsan Hospital. This study was supported by 2024 research grant from Pusan National University Yangsan Hospital. The funding bodies did not have a role in the design of the study and collection, analysis, and interpretation of data and in writing the manuscript.

Footnote

Reporting Checklist: The authors have completed the REMARK reporting checklist. Available at https://tcr.amegroups.com/article/view/10.21037/tcr-23-2286/rc

Peer Review File: Available at https://tcr.amegroups.com/article/view/10.21037/tcr-23-2286/prf

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at https://tcr.amegroups.com/article/view/10.21037/tcr-23-2286/coif). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. This study was conducted in accordance with the Declaration of Helsinki (as revised in 2013).

References

1. Carrano A, Juarez JJ, Incontri D, et al. Sex-Specific Differences in Glioblastoma. Cells 2021;10:1783.
2. Hanif F, Muzaffar K, Perveen K, et al. Glioblastoma Multiforme: A Review of its Epidemiology and Pathogenesis through Clinical Presentation and Treatment. Asian Pac J Cancer Prev 2017;18:3-9.
3. Sung H, Ferlay J, Siegel RL, et al. Global Cancer Statistics 2020: GLOBOCAN Estimates of Incidence and Mortality Worldwide for 36 Cancers in 185 Countries. CA Cancer J Clin 2021;71:209-49.
4. Poon MTC, Sudlow CLM, Figueroa JD, et al. Longer-term (≥2 years) survival in patients with glioblastoma in population-based studies pre- and post-2005: a systematic review and meta-analysis. Sci Rep 2020;10:11622.
5. Louis DN, Wesseling P, Aldape K, et al. cIMPACT-NOW update 6: new entity and diagnostic principle recommendations of the cIMPACT-Utrecht meeting on future CNS tumor classification and grading. Brain Pathol

2020;30:844-56.
6. Louis DN, Perry A, Wesseling P, et al. The 2021 WHO Classification of Tumors of the Central Nervous System: a summary. Neuro Oncol 2021;23:1231-51.
7. Melhem JM, Detsky J, Lim-Fat MJ, et al. Updates in IDH-Wildtype Glioblastoma. Neurotherapeutics 2022;19:1705-23.
8. Pasquini L, Napolitano A, Tagliente E, et al. Deep Learning Can Differentiate IDH-Mutant from IDH-Wild GBM. J Pers Med 2021;11:290.
9. Ding X, Zhang W, Li S, et al. The role of cholesterol metabolism in cancer. Am J Cancer Res 2019;9:219-27.
10. Murai T. Cholesterol lowering: role in cancer prevention and treatment. Biol Chem 2015;396:1-11.
11. Hu J, La Vecchia C, de Groh M, et al. Dietary cholesterol intake and cancer. Ann Oncol 2012;23:491-500.
12. Debinski WSassi K, Nury T, et al. Cholesterol Derivatives as Promising Anticancer Agents in Glioblastoma Metabolic Therapy. In: Debinski W, editor. Gliomas. Brisbane (AU): Exon Publications; April 30, 2021.
13. Gaist D, Hallas J, Friis S, et al. Statin use and survival following glioblastoma multiforme. Cancer Epidemiol 2014;38:722-7.
14. Alonso R, Cuevas A, Mata P. Lomitapide: a review of its clinical use, efficacy, and tolerability. Core Evid 2019;14:19-30.
15. Hussain MM, Rava P, Walsh M, et al. Multiple functions of microsomal triglyceride transfer protein. Nutr Metab (Lond) 2012;9:14.
16. Xie Y, Matsumoto H, Nalbantoglu I, et al. Intestine-Specific Mttp Deletion Increases the Severity of Experimental Colitis and Leads to Greater Tumor Burden in a Model of Colitis Associated Cancer. PLoS One 2013;8:e67819.
17. Kwon RJ, Kim YH, Jeong DC, et al. Expression and prognostic significance of zinc fingers and homeoboxes family members in renal cell carcinoma. PLoS One 2017;12:e0171036.
18. Mamoor S. MTTP is differentially expressed in brain metastatic human breast cancer. OSF 2021;1-10. doi: 10.31219/osf.io/ajns7.
19. Bazzoni R, Bentivegna A. Role of Notch Signaling Pathway in Glioblastoma Pathogenesis. Cancers (Basel) 2019;11:292.
20. Vinson KE, George DC, Fender AW, et al. The Notch pathway in colorectal cancer. Int J Cancer 2016;138:1835-42.
21. Fernández LP, Gómez de Cedrón M, Ramírez de Molina A. Alterations of Lipid Metabolism in Cancer: Implications in Prognosis and Treatment. Front Oncol 2020;10:577420.
22. Beloribi-Djefaflia S, Vasseur S, Guillaumond F. Lipid metabolic reprogramming in cancer cells. Oncogenesis 2016;5:e189.
23. Taïb B, Aboussalah AM, Moniruzzaman M, et al. Lipid accumulation and oxidation in glioblastoma multiforme. Sci Rep 2019;9:19593.
24. Dietschy JM. Central nervous system: cholesterol turnover, brain development and neurodegeneration. Biol Chem 2009;390:287-93.
25. Shakya S, Gromovsky AD, Hale JS, et al. Altered lipid metabolism marks glioblastoma stem and non-stem cells in separate tumor niches. Acta Neuropathol Commun 2021;9:101.
26. Wu X, Geng F, Cheng X, et al. Lipid Droplets Maintain Energy Homeostasis and Glioblastoma Growth via Autophagic Release of Stored Fatty Acids. iScience 2020;23:101569.
27. Kou Y, Geng F, Guo D. Lipid Metabolism in Glioblastoma: From De Novo Synthesis to Storage. Biomedicines 2022;10:1943.
28. Lindström MS. Expanding the scope of candidate prognostic marker IGFBP2 in glioblastoma. Biosci Rep 2019;39:BSR20190770.
29. Hsieh D, Hsieh A, Stea B, et al. IGFBP2 promotes glioma tumor stem cell expansion and survival. Biochem Biophys Res Commun 2010;397:367-72.
30. Yuan Q, Cai HQ, Zhong Y, et al. Overexpression of IGFBP2 mRNA predicts poor survival in patients with glioblastoma. Biosci Rep 2019;39:BSR20190045.
31. Sincevičiūtė R, Vaitkienė P, Urbanavičiūtė R, et al. MMP2 is associated with glioma malignancy and patient outcome. Int J Clin Exp Pathol 2018;11:3010-8.
32. Alkhaibary A, Alassiri AH, AlSufiani F, et al. Ki-67 labeling index in glioblastoma; does it really matter? Hematol Oncol Stem Cell Ther 2019;12:82-8.
33. Nielsen LAG, Bangsø JA, Lindahl KH, et al. Evaluation of the proliferation marker Ki-67 in gliomas: Interobserver variability and digital quantification. Diagn Pathol 2018;13:38.
34. Chen WJ, He DS, Tang RX, et al. Ki-67 is a valuable prognostic factor in gliomas: evidence from a systematic review and meta-analysis. Asian Pac J Cancer Prev 2015;16:411-20.
35. Hayati N, Tabriz HM, Nazar E, et al. Relationship between phosphatase and tensin homolog (PTEN) expression in high grade glioma and histopathologic

findings. Ro J Neurol 2021;20:452.
36. Tang L, Li X, Gao Y, et al. Phosphatase and tensin homolog (PTEN) expression on oncologic outcome in renal cell carcinoma: A systematic review and meta-analysis. PLoS One 2017;12:e0179437.
37. Phillips HS, Kharbanda S, Chen R, et al. Molecular subclasses of high-grade glioma predict prognosis, delineate a pattern of disease progression, and resemble stages in neurogenesis. Cancer Cell 2006;9:157-73.
38. Walid MS. Prognostic factors for long-term survival after glioblastoma. Perm J 2008;12:45-8.
39. Brown NF, Ottaviani D, Tazare J, et al. Survival Outcomes and Prognostic Factors in Glioblastoma. Cancers (Basel) 2022;14:3161.
40. Brown TJ, Bota DA, van Den Bent MJ, et al. Management of low-grade glioma: a systematic review and meta-analysis. Neurooncol Pract 2019;6:249-58.
41. Villa GR, Hulce JJ, Zanca C, et al. An LXR-Cholesterol Axis Creates a Metabolic Co-Dependency for Brain Cancers. Cancer Cell 2016;30:683-93.
42. Wilson TM. Identifying Novel Targets in Chemo-Resistant Cell Populations in Glioblastoma Multiforme [Internet]. Institute of Medical Science University of Toronto; 2020. Available online: https://tspace.library.utoronto.ca/bitstream/1807/103133/1/Wilson_Taylor_Marie_202011_MSc_thesis.pdf
43. Han S, Liu Y, Cai SJ, et al. IDH mutation in glioma: molecular mechanisms and potential therapeutic targets. Br J Cancer 2020;122:1580-9.
44. Huang J, Yu J, Tu L, et al. Isocitrate Dehydrogenase Mutations in Glioma: From Basic Discovery to Therapeutics Development. Front Oncol 2019;9:506.
45. Waitkus MS, Diplas BH, Yan H. Isocitrate dehydrogenase mutations in gliomas. Neuro Oncol 2016;18:16-26.
46. Alzial G, Renoult O, Paris F, et al. Wild-type isocitrate dehydrogenase under the spotlight in glioblastoma. Oncogene 2022;41:613-21.
47. Aran D, Sirota M, Butte AJ. Systematic pan-cancer analysis of tumour purity. Nat Commun 2015;6:8971.
48. Lundberg E, Fagerberg L, Klevebring D, et al. Defining the transcriptome and proteome in three functionally different human cell lines. Mol Syst Biol 2010;6:450.

Cite this article as: Son SM, Lee HS, Kim J, Kwon RJ. Expression and prognostic significance of microsomal triglyceride transfer protein in brain tumors: a retrospective cohort study. Transl Cancer Res 2024;13(5):2282-2294. doi: 10.21037/tcr-23-2286

