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RESEARCH

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Characteristics and outcomes of diffuse non-midline gliomas with H3F3A gene mutation in the Kansai Molecular Diagnosis Network for CNS Tumors (Kansai Network): multicenter retrospective cohort study

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Abstract

Diffuse gliomas with *H3F3A* gene mutation such as H3.3 K27M and G34R/V are infrequently found in the cerebral hemisphere. These tumors may be called histone H3 K27M-mutant diffuse non-midline gliomas (NDMG) or H3 G34-mutant diffuse hemispheric gliomas (DHG), respectively. We investigated the clinical, radiological and molecular characteristics and treatment outcomes of patients with H3 K27-mutant NDMG compared with those with H3 G34-mutant DHG. We collected cases of histone H3-mutant diffuse glioma at non-midline location that were enrolled in the Kansai Molecular Diagnosis Network for CNS Tumors. The clinical, radiological and pathological characteristics and DNA methylation were retrospectively analyzed and then compared between cases of NDMG and DHG. We evaluated various factors to examine their effects on overall survival. Included in this study were 16 patients with H3 K27M-mutant NDMG and nine patients with H3 G34R/V-mutant DHG. Patients with NDMG were older than those with DHG (median age: 45 vs. 25 years old). Overall survival times of patients with NDMG were comparable to those of DHG (median overall survival: 20.0 vs. 22.5 months). Female sex, preoperative Karnofsky performance status score ≥ 80 and extensive surgical resection tended be related to better prognoses in the patients with these tumors. H3 K27M-mutant NDMGs were shown to possess similar DNA methylation properties to H3 K27M-mutant DMGs. Diffuse gliomas harboring histone H3 K27M mutation can arise in the cerebral hemisphere in older adults. These findings suggest that H3 K27M-mutant NDMGs show different clinical manifestations to H3 K27M-mutant DMGs and H3 G34R/V-mutant DHGs, despite having similar molecular properties to H3 K27M-mutant DMGs.

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Keywords Diffuse non-midline glioma, H3K27-altered, H3G34-mutant, Diffuse hemispheric glioma, H3F3A gene mutation, DNA methylation

Introduction

A mutation in *H3F3A* gene, which encodes the histone H3 variant H3.3, results in amino acid substitutions at two critical positions within the histone tail (K27M, G34R/G34V). It is highly prevalent in glioblastomas of children and young adults [14]. H3 K27M-mutant diffuse midline glioma (DMG) occurs mainly in the brainstem region in children or in a midline location in older children and adults, such as in the thalamus or spinal cord [16]. We have previously reported on the clinical features of DMG [8].

H3 G34-mutant diffuse hemispheric glioma (DHG), meanwhile, is a rare, newly-recognized infiltrating glioma of the cerebral hemisphere [16]. However, diffuse glioma harboring H3.3 K27M mutation has sometimes occurred in non-midline structures, such as in the cerebral hemisphere, and has been pathologically diagnosed as 'pediatric-type diffuse high-grade glioma, H3.3 K27-mutant, NEC', which can be called 'diffuse non-midline glioma' (NDMG) [7, 10, 11, 17] In the cerebral hemisphere, diffuse gliomas with *H3F3A* gene mutation such as H3 K27M-mutant NDMG and H3 G34-mutant DHG can arise, but the difference between these hemispheric tumors, other than the different regions of histone H3 mutation (K27 and G34), is unknown.

To resolve this question, we conducted a comparative study between H3 K27M-mutant NDMG and H3 G34-mutant DHG based on the Kansai Molecular Diagnosis Network for CNS tumors (Kansai Network) dataset [8]. We focused upon clinical, radiological and molecular characteristics as well as treatment outcomes of patients with these hemispheric gliomas.

Methods and materials

Ethics

This study was carried out in accordance with the principles of the Declaration of Helsinki. Approval was obtained from the Seirei Hamamatsu General Hospital Institutional Review Board (No. 2453), the Wakayama Medical University Institutional Review Board (No. 98), the Osaka National Hospital Institutional Review Board (No. 713), and those of all collaborative institutes. Written informed consent was obtained from all patients.

Patient population

We collected 4128 samples including all kinds of primary and recurrent gliomas from the 72 hospitals participating in the Kansai Network from between May 2007 and July 2022 that had clinical information and analyzes of the molecular status of tumors for diagnosis and research [8]. Within this data bank, we found 118 cases of patients with *H3F3A* mutation, and distinguished between DMG, H3 K27-altered, and NDMG. NDMG was defined as lesions that were cerebral hemispheres or hemispheric lesions and within the basal ganglia, while DMG was lesions in the basal ganglia and the thalamus [8]. We diagnosed 16 patients (14.7%) as having NDMG and nine patients (8%) as having DHG, excepting 93 cases (92%) of patients with DMG [8].

Clinical information

Clinical information was retrospectively collected from the Kansai Network database including patient demographics, the extent of surgical resection (EOR), adjuvant radiation and chemotherapy regimens, and survival time. Plain magnetic resonance images (MRI) scans and enhanced scans were performed in 24 patients. Preoperative MRI was unavailable in the case of one patient with DHG. MRI features were compared using diffusion-weighted imaging, fluid-attenuated inversion recovery (FLAIR)-weighted imaging and enhanced scans for 16 patients with NDMG and eight patients with DHG. MRI feature characterizations were made with the four neurosurgeons (HN, HK, JF, NH) blinded to outcomes. They were recorded as eleven semantic descriptors from the VASARI (Visually AcceSAble Rembrandt Images) MRI feature set reflecting the tumor margin, the extent of surrounding edema, the degree of enhancement, the presence or absence of radiologic necrosis, the diffusion status, and the presence or absence of leptomeningeal metastatic dissemination [19].

Histopathological examination

All cases underwent central pathology review by a senior board-certified neuropathologist (YoKo). Integrated diagnosis and WHO grading were made based on the 2021 WHO Classification of Tumors of the CNS [16].

Genetic analysis

Genetic analysis was performed using DNA derived from tumor samples, as previously reported [8]. Tumor genomic DNA was extracted from frozen or fresh tumor samples with NucleoSpin Tissue kit (Macherey–Nagel, Inc., Bethlehem, PA, USA) or DNeasy Blood and Tissue Kit (Qiagen, Tokyo, Japan), according to the manufacturer's protocol. The presence of hotspot mutations in

H3F3A, HIST1H3B, IDH1 (R132), *IDH2* (R172), *TERT promoter*, *BRAF* (V600), *FGFR1* (exon12 and exon14) and *EGFR* (exon 7 and exon20) genes, and all exons of *TP53* were analyzed by Sanger sequencing [8]. Methylation status of O-6-methylguanine-DNA methyltransferase (*MGMT*) promoter was analyzed by quantitative methylation-specific PCR (qMSP) after bisulfite modification of genomic DNA [8]. Based on an outcome-based study to determine an optimal cut-off to judge MGMT promoter methylation in a series of newly diagnosed glioblastoma (GBM), we used a cut-off of ≥ 1% for MGMT methylation [8].

Next-generation sequencing analysis

A DNA library was constructed using the QIAseq Targeted DNA Pro Human Brain Cancer Research Panel (PHS-004Z, Qiagen, Venlo, Netherlands) which targets 50 genes relevant to brain tumors in accordance with the manufacturer's instructions. Libraries were quantified and sequenced on the NextSeq 550 system (Illumina, San Diego, CA, USA). Amplicon sequences were aligned to the human reference genome GRCh38 in the target region of the sequence, and data analysis was performed using the Qiagen web portal (https://geneglobe.qiagen.com/jp) [1].

DNA methylation array

DNA methylation profiles were examined by Rhelixa (Tokyo, Japan) using the Infinium® methylation EPIC BeadChip system (Illumina) according to the manufacturer's instructions. Raw methylation profile data (IDAT files) were uploaded onto the Molecular Neuropathology website (https://www.molecularneuropathology.org/mnp/), and analyzed using the Brain Tumor Classifier of the Deutsches Krebsforschungszentrum (v.12.8 for the EPICV2 BeadChip) [3]. The t-distributed stochastic neighbor embedding (t-SNE) plot for five NDMGs and three DHGs and 406 references from the previously published Deutsches Krebsforschungszentrum study (GSE109381) was made using version 0.17 of the Rtsne package [12]. Preprocessing for analysis was performed with the SeSAME package [6] using R software, version 4.3.2. Data were normalized using the SeSAME normalization method. The 15,000 most variable probes were determined using random Forest R package v4.7.1.1 [2] and used to generate t-SNE plots.

Statistical analysis

Statistical analyses were performed using the SAS package and JMP Pro version 16 (SAS Institute, Cary, NC, USA) and with EZR [9], a modified version of R commander designed to add statistical functions frequently used in biostatistics. Categorized data were compared between subgroups using Chi-square test. Overall survival (OS) curves were estimated by Kaplan–Meier method and compared with log-rank test. Univariate analyses of risk factors were performed using Cox proportional hazards model. p value < 0.05 was considered statistically significant.

Results

Clinical characteristics of patients with NDMG and patients with DHG

Clinical characteristics of the 16 patients with NDMG and nine patients with DHG in the study are shown in Table 1 and Fig. 1. The NDMG group was significant older (mean age: 45 years old, age range: 26–79 years) than the DHG group (mean age: 25 years old, age range: 11–45 years) ($p = 0.002$). Male predominance was seen in both groups (62.5% and 66.7%, respectively). Regarding tumor location, there was a statistically significant difference between the groups ($p = 0.0351$). About half of the patients in the NDMG and DHG groups had preoperative Karnofsky performance status (KPS) scores ≥ 80 (50.0% and 55.6%, respectively). Regarding EOR, there was a trend toward partial resection (PR) or biopsy compared with gross-total or subtotal resection (GTR or STR) in both groups (75.0% vs. 25.0% in NDMG and 66.6% vs. 33.3% in DHG, respectively). After surgery, adjuvant radiation therapy (RT) and temozolomide (TMZ) chemotherapy was likely to be performed in both groups (81.3% and 88.9%). About one third of cases in each group received bevacizumab (BEV) chemotherapy (31.3% and 33.3%) (Table 1).

Radiological characteristics of patients with NDMG and patients with DHG

Radiological characteristics of patients with NDMG and patients with DHG in the study are shown in Table 2 and Fig. 1. In most cases, both NDMG and DHG had radiologically poorly-defined enhancing margins (11/14, 78.6% and 3/6, 50.0%, respectively), but both groups included some well-defined cases. However, the definition of the non-enhancing margin was irregular in all cases (16/16, and 8/8, respectively). Approximately one-third of both types of tumors contained a part of thick/solid enhancement with a varied pattern (10/16, 62.5%; 5/8, 62.5%). Intra-tumoral necrosis (as defined by MRI) was seen in six cases of NDMG (6/16, 37.5%) and in two cases of DHG (2/7, 28.6%). Hemorrhage was not seen in any cases of either of the groups. Cysts were present in one patient with NDMG (1/16, 6.3%), although there were three cases among patients with DHG (3/7, 42.9%). Over 34% proportion of edema was found in the NDMG (10/16, 62.5%) and DHG (7/8, 87.5%) groups. Most tumors of both NDMG

Table 1 Clinical characteristics of patients with histone H3 K27-mutant diffuse non-midline glioma (n = 16) and H3 G34-mutant diffuse hemispheric glioma (n = 9) in the Kansai Network

Characteristic	H3 K27-mutant diffuse non-midline glioma (n = 16)	H3 G34-mutant diffuse hemispheric glioma (n = 9)	*p* value
Age (y)[†]	45 (26–79)	25 (11–45)	0.002*
Sex			0.8345
Female	6 (37.5%)	3 (33.3%)	
Male	10 (62.5%)	6 (66.7%)	
Location			0.0351*
Frontal	4 (25.0%)	1 (11.1%)	
Front-temporal	1 (6.3%)	0 (–)	
Front-parietal	1 (6.3%)	0 (–)	
Temporal	5 (31.3%)	2 (22.2%)	
Parietal	1 (6.2%)	6 (66.7%)	
Basal ganglia	3 (18.8%)	0 (–)	
Multiple	1 (6.3%)	0 (–)	
Preoperative KPS score			0.7894
80–100	8 (50%)	5 (55.6%)	
– 70	8 (50%)	4 (44.4%)	
Extent of surgical resection			0.3222
GTR	2 (12.5%)	3 (33.3%)	
STR	2 (12.5%)	0 (–)	
PR	4 (25.0%)	3 (33.3%)	
Biopsy	8 (50%)	3 (33.3%)	
BCNU wafer			0.2488
Yes	1 (6.3%)	2 (22.2%)	
No	15 (93.8%)	7 (77.8%)	
Adjuvant treatment			0.5749
RT + TMZ	13 (81.3%)	8 (88.9%)	
RT alone	1 (6.3%)	0 (–)	
TMZ alone	1 (6.3%)	0 (–)	
None	1 (6.3%)	1 (11.1%)	
BEV therapy			0.9148
Yes	5 (31.3%)	3 (33.3%)	
No	11 (68.8%)	6 (66.7%)	
OS (mo)[†]	20	22.5	0.2958

Unless otherwise specified, data are numbers of cases. LGG, diffusely infiltrative glioma without histological features of anaplasia, which displays no/low mitotic activity; HGG, diffusely infiltrative glioma with histological features of anaplasia and displays significant mitotic activity; GBM features, microvascular proliferation or necrosis

GTR, Gross total resection; PFS, Progression-free survival; NA, Not available; OS, Overall survival; PR, Partial resection; RT, Radiation therapy; TMZ, Temozolomide; STR, Subtotal resection; WHO, World Health Organization

[†] Data are medians, with ranges in parentheses

*$p < 0.05$, statistically significant difference

and DHG groups showed a part of diffusion restriction (11/12, 91.7% and 6/6, 100%, respectively). Leptomeningeal dissemination before surgery was not present in either NDMG or DHG groups. Typical cases of NDMG and DHG are shown in Fig. 2; they were both located in the hemispheric lobe.

Histological and molecular characteristics of NDMG and DHG

Histological and molecular characteristics and the frequency of each genetic status are shown in Table 3 and Fig. 1. Regarding histopathological findings, high-grade histology such GBM and high-grade glioma without

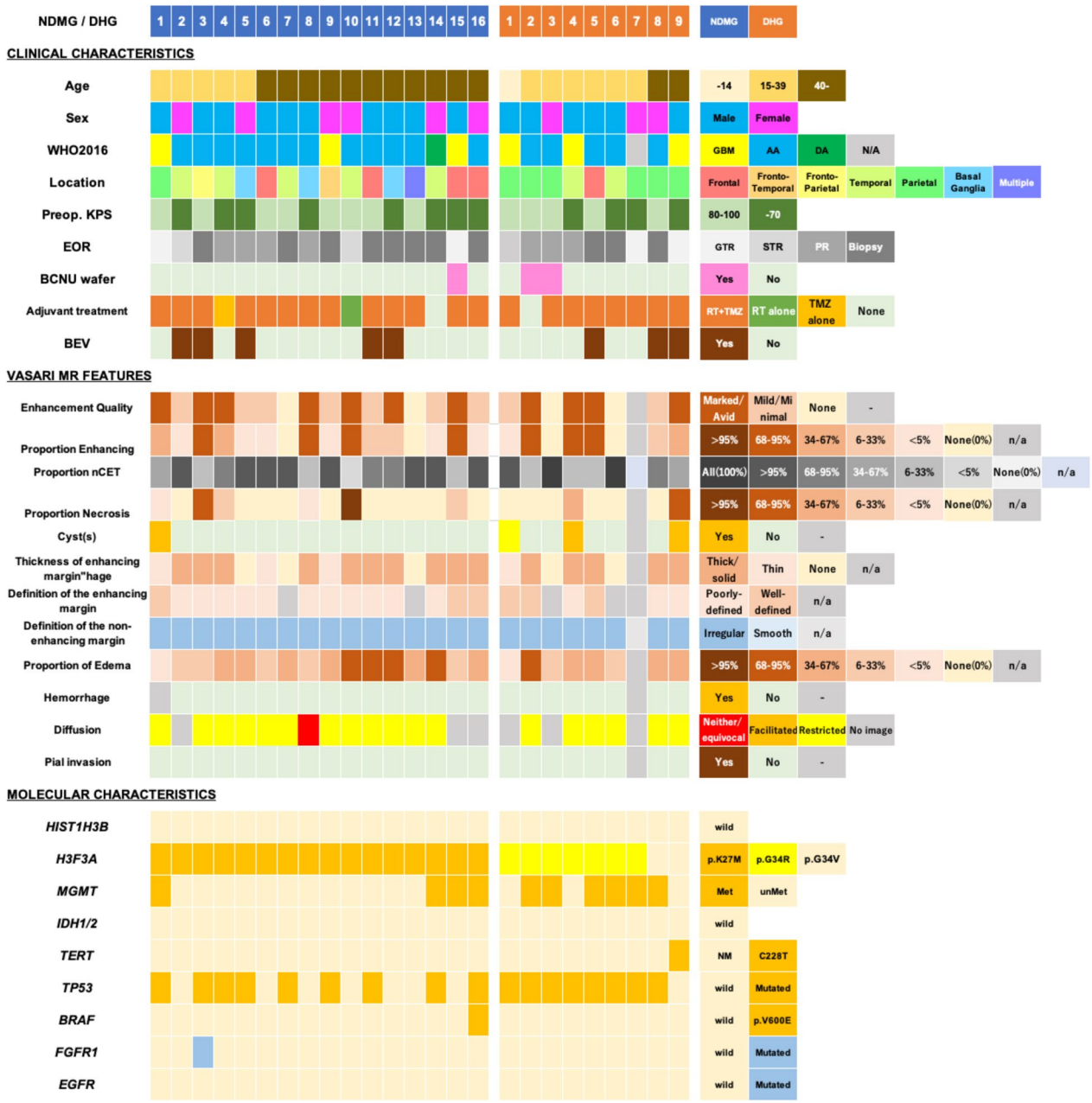

Fig. 1 Tile panel demonstrating clinical, radiological and molecular characteristics of patients with histone H3 K27-mutant diffuse non-midline glioma (n = 16) and H3 G34-mutant diffuse hemispheric glioma (n = 9) in the Kansai Network

GBM features was commonly observed in both NDMG and DHG groups. On the other hand, there were some differences of genetic status between the groups. By Sanger sequencing, one *TERT* promoter mutation was observed in only one patient of the DHG group (11.1%), but not in the NDMG group. *MGMT* promoter methylation in the cohort was present in four tumors in the NDMG group (25.0%) and in six tumors in the DHG group (66.7%), but there was no statistical difference between them ($p = 0.053$). In the cohort, *TP53* mutation was detected in the NDMG group less frequently than in the DHG group (56.3% vs. 88.9%). *BRAF* p.V600E or *FGFR1* hotspot mutation was observed in only one patient of the NDMG group (6.3%), but not at all in the DHG group. *EGFR* hotspot mutation was not seen in either of the groups.

To further analyze molecular properties of NDMGs, we additionally examined the genetic status of 50 genes

Table 2 Radiologic characteristics of patients with histone H3 K27-mutant diffuse non-midline glioma (n = 16) and H3 G34-mutant diffuse hemispheric glioma (n = 9) in the Kansai Network

Characteristic	H3 K27-mutant diffuse non-midline glioma (n = 16)	H3 G34-mutant diffuse hemispheric glioma (n = 9)	*p* value
VASARI MR features			
Enhancement quality			0.4137
–	0 (–)	1 (11.1%)	
None	2 (12.5%)	2 (22.2%)	
Mild/minimal	7 (43.8%)	2 (22.2%)	
Marked/avid	7 (43.8%)	4 (44.4%)	
Proportion enhancing			0.5916
–	0 (–)	0 (–)	
n/a	0 (–)	1 (11.1%)	
None (0%)	2 (12.5%)	2 (22.2%)	
< 5%	6 (37.5%)	1 (11.1%)	
6–33%	2 (12.5%)	1 (11.1%)	
34–67%	2 (12.5%)	1 (11.1%)	
68–95%	4 (25.0%)	3 (33.3%)	
> 95%	0 (–)	0	
All (100%)	0 (–)	0 (–)	
Indeterminate	0 (–)	0 (–)	
Proportion nCET			0.1623
–	0 (–)	0	
n/a	0 (–)	1 (11.1%)	
None (0%)	0 (–)	0 (–)	
< 5%	1 (6.3%)	0 (–)	
6–33%	3 (18.8%)	3 (33.3%)	
34–67%	1 (6.3%)	1 (11.1%)	
68–95%	3 (18.8%)	1 (11.1%)	
> 95%	8 (50.0%)	1 (11.1%)	
All (100%)	0 (–)	2 (22.2%)	
Indeterminate	0 (–)	0 (–)	
Proportion necrosis			0.4956
–	0 (–)	0 (–)	
n/a	0 (–)	1 (11.1%)	
None (0%)	10 (62.5%)	6 (66.7%)	
< 5%	2 (12.5%)	0 (–)	
6–33%	2 (12.5%)	0 (–)	
34–67%	1 (6.3%)	1 (11.1%)	
68–95%	1 (6.3%)	1 (11.1%)	
> 95%	0 (–)	0 (–)	
All (100%)	0 (–)	0 (–)	
Indeterminate	0 (–)	0 (–)	
Cyst(s)			0.0649
–	0 (–)	1 (11.1%)	
No	15 (93.8%)	5 (55.6%)	
Yes	1 (6.3%)	3 (33.3%)	
Thickness of enhancing margin			0.5582
–	0 (–)	0 (–)	
n/a	0 (–)	1 (11.1%)	
None	3 (18.8%)	2 (22.2%)	
Thin	3 (18.8%)	1 (11.1%)	

Table 2 (continued)

Characteristic	H3 K27-mutant diffuse non-midline glioma (n = 16)	H3 G34-mutant diffuse hemispheric glioma (n = 9)	*p* value
Thick/solid	10 (62.5%)	5 (55.6%)	
Definition of the enhancing margin			0.2176
–	0 (–)	0 (–)	
n/a	2 (12.5%)	3 (33.3%)	
Well-defined	3 (18.8%)	3 (33.3%)	
Poorly-defined	11 (68.8%)	3 (33.3%)	
Definition of the non-enhancing margin			0.1736
–	0 (–)	0 (–)	
n/a	0 (–)	1 (11.1%)	
Smooth	0 (–)	0	
Irregular	16 (100%)	8 (88.9%)	
Proportion of edema			0.6021
–	0 (–)	0 (–)	
n/a	0 (–)	1 (11.1%)	
None (0%)	0 (–)	0 (–)	
< 5%	1 (6.3%)	1 (11.1%)	
6–33%	5 (31.3%)	2 (22.2%)	
34–67%	6 (37.5%)	4 (44.4%)	
68–95%	4 (25.0%)	1 (11.1%)	
> 95%	0 (–)	0 (–)	
All (100%)	0 (–)	0 (–)	
Indeterminate	0 (–)	0 (–)	
Hemorrhage			0.6672
–	1 (6.3%)	1 (11.1%)	
No	15 (93.8%)	8 (88.9%)	
Yes	0 (–)	0 (–)	
Diffusion			0.7015
–	0 (–)	0 (–)	
No image	4 (25.0%)	3 (33.3%)	
Facilitated	1 (6.3%)	0 (–)	
Restricted	11 (68.8%)	6 (66.7%)	
Neither/equivocal	0 (–)	0 (–)	
Pial invasion			0.1736
–	0 (–)	1 (11.1%)	
No	16 (100%)	8 (88.9%)	
Yes	0 (–)	0 (–)	

Unless otherwise specified, data are numbers of cases

n/a, Not available; VASARI, Visually AcceSAble Rembrandt Images

(Fig. 3) using next-generation sequencing (NGS). In total, 14 cases (nine cases in NDMG [56.3%] and five cases in DHG [55.6%]) were analyzed using NGS, and four single nucleotide variants (SNVs) were newly identified; pathogenic SNVs of *PTEN* (NDMG and DHG) and *PIK3CA* (DHG), and variant of uncertain significance (VUS) of *APC* (NDMG) (Fig. 3). We also found a case with *EGFR* amplification, two cases with *PDGFRA* amplification, and a case with *PTEN* homozygous deletion which were all from NDMG group, respectively (Fig. 3).

Treatment outcomes and prognostic factors of patients with NDMG and patients with DHG

OS curves of the NDMG and DHG groups are shown in Fig. 4. Median OS (mOS) of the NDMG group was 20 months and not significantly different from that of the DHG group, which was 22.5 months. Regarding sex,

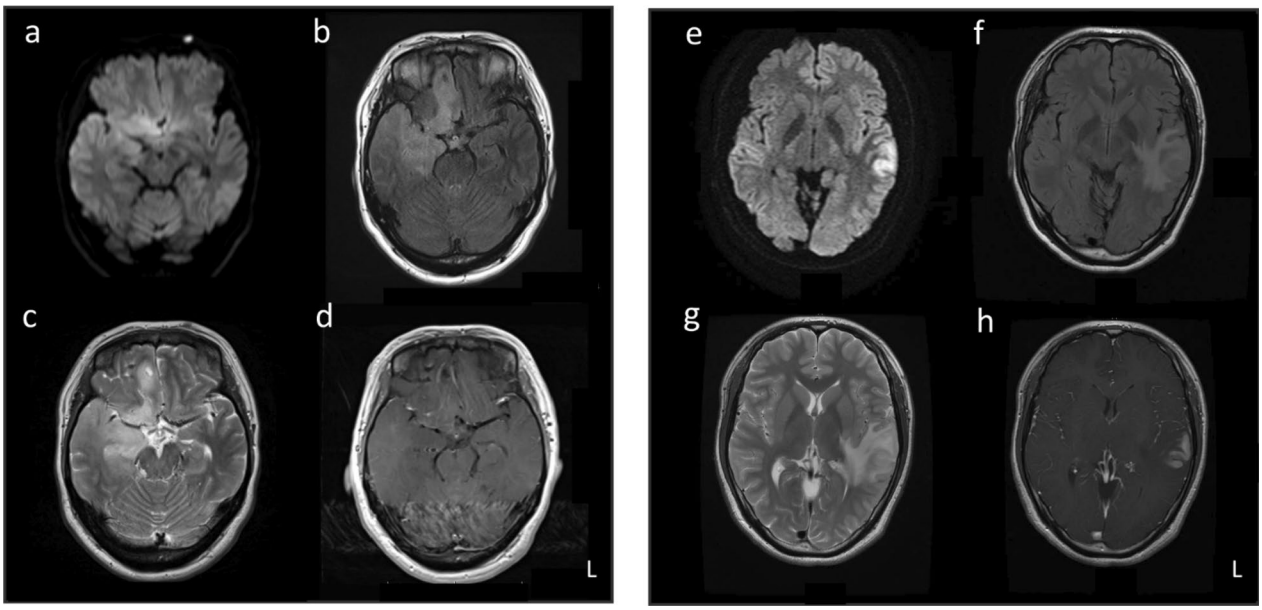

Fig. 2 Brain MRI of an illustrative case of diffuse non-midline glioma with H3 K27M-mutant shows ill-defined tumor margin in fronto-temporal region. **a** Diffusion-weighted image. **b** FLAIR image. **c** T2-weighted image. **d** Contrast enhancement T1-weighted image. Brain MRI of an illustrative case of diffuse hemispheric glioma with H3 G34-mutant shows ill-defined tumor margin in temporal region. **e** Diffusion-weighted image. **f** FLAIR image. **g** T2-weighted image. **h** Contrast enhancement T1-weighted image

the mOS of female patients was longer than that of male patients in both the NDMG (20.0 vs. 16.0 months) and the DHG (45.0 vs. 14.0 months) groups (Supplementary Fig. 1), but without significant difference in sex in either of the groups ($p=0.4468$ and 0.272, respectively). Regarding preoperative KPS score, mOS of patients with KPS scores of ≥ 80 was longer than that of ≤ 70 patients in both NDMG (33.0 vs. 16.0 months) and DHG (not reached vs. 14.0 months) groups, but there was no significant difference between patients with KPS scores of ≥ 80 and ≤ 70 in both groups ($p=0.0555$ and 0.0933, respectively) (Supplementary Fig. 2). Regarding EOR, mOS of GTR or STR was longer than that of PR or biopsy in both NDMG (27.5 vs. 16.0 months) and DHG (45.0 vs. 22.0 months) groups, but there was no significant difference between GTR or STR and PR or biopsy in either of the groups ($p=0.1447$ and 0.4925, respectively) (Supplementary Fig. 3). OS curves of patients treated with adjuvant RT and TMZ chemotherapy are shown in Supplementary Fig. 4. There was no significant difference between the groups. According to each clinical status, survival differences between the subgroups were examined in the cases of NDMG and the cases of DHG, respectively. Regarding BEV chemotherapy for the NDMG cases, mOS in patients with BEV chemotherapy tended to be longer than that in patients without BEV chemotherapy, but without statistical significance (20.0 vs. 16.0 months, $p=0.6687$); however, mOS in patients with and without BEV chemotherapy were similar in the cases of DHG (22.0 vs. 23.0 months, $p=0.8497$) (Supplementary Fig. 5).

Molecular characteristics and prognostic factors of patients with NDMG and patients with DHG

According to each molecular status, we examined survival differences between subgroups within the cases of NDMG and DHG, respectively. Regarding *MGMT* promoter methylation status, among the cases of NDMG, we found no significant difference in OS time between methylated and unmethylated subgroups (16.0 months vs. 20.0 months, $p=0.5673$). Among the cases of DHG, on the other hand, the methylated subgroup had a longer OS time (23.0 months) than the unmethylated subgroup (14.0 months), but the difference did not reach statistical significance ($p=0.9888$) (Supplementary Fig. 6). Regarding *TP53* mutation status, the mutated subgroup had a longer OS time than the wildtype subgroup in both the NDMG and DHG groups (23.0 vs. 14.0 months and 20.0 vs. 16.0 months, respectively), but the difference did not reach statistical significance ($p=0.515$ and 0.293, respectively) (Supplementary Fig. 7). In this study, survival analyses identified no independent prognostic factors in either the NDMG group or in the DHG group.

Table 3 Pathologic and molecular characteristics of patients with histone H3 K27-mutant diffuse non-midline glioma (n = 16) and H3 G34-mutant diffuse hemispheric glioma (n = 9) in the Kansai Network

Characteristic	H3 K27-mutant diffuse non-midline glioma (n = 16)	H3 G34-mutant diffuse hemispheric glioma (n = 9)	*p* value
Histopathologic findings			
Histologic diagnosis (CNS WHO 2021)			0.289
LGG	1 (6.3%)	0 (–)	
HGG without GBM features	12 (75%)	5 (55.6%)	
GBM features	3 (18.8%)	3 (33.3%)	
NA	0 (–)	1 (11.1%)	
Microvascular proliferation			0.0824
Present	1 (6.3%)	3 (33.3%)	
Absent	5 (31.3%)	4 (44.4%)	
NA	10 (62.5%)	2 (22.2%)	
Necrosis			0.0736
Present	2 (12.5%)	3 (33.3%)	
Absent	3 (18.8%)	4 (44.4%)	
NA	11 (68.8%)	2 (22.2%)	
Ki-67 (%)			0.5958
≤ 5	1 (6.3%)	0 (–)	
> 5	4 (25.0%)	3 (33.3%)	
NA	11 (68.8%)	6 (66.7%)	
Genetic status			
Histone H3			< 0.0001*
K27M	16 (100%)	0 (–)	
G34R	0 (–)	7 (77.8%)	
G34V	0 (–)	2 (22.2%)	
TERT promoter			0.174
Wild	16 (100%)	8 (88.9%)	
C228T/C250T	0 (–)	1 (11.1%)	
MGMT promoter			0.053
Methylated	4 (25.0%)	6 (66.7%)	
Unmethylated	12 (75.0%)	3 (33.3%)	
TP53			0.093
Wild	7 (43.7%)	1 (11.1%)	
Mutated	9 (56.3%)	8 (88.9%)	
BRAF			0.444
Wild	15 (93.7%)	9 (100%)	
p.V600E	1 (6.3%)	0 (–)	
FGFR1			0.444
Wild	15 (93.7%)	9 (100%)	
Mutated	1 (6.3%)	0 (–)	
EGFR			–
Wild	16 (100%)	9 (100%)	
Mutated	0	0	
PDGFRA			–
Wild	9 (56.3%)	5 (55.6%)	
Mutated	0 (–)	0 (–)	
NA	7 (43.8%)	4 (44.4%)	
CDK4/6			–
Wild	9 (56.3%)	5 (55.6%)	
Mutated	0 (–)	0 (–)	

Table 3 (continued)

Characteristic	H3 K27-mutant diffuse non-midline glioma (n = 16)	H3 G34-mutant diffuse hemispheric glioma (n = 9)	*p* value
NA	7 (43.8%)	4 (44.4%)	
ACVR1			–
Wild	9 (56.3%)	5 (55.6%)	
Mutated	0 (–)	0 (–)	
NA	7 (43.8%)	4 (44.4%)	
MYC			–
Wild	9 (56.3%)	5 (55.6%)	
Mutated	0 (–)	0 (–)	
NA	7 (43.8%)	4 (44.4%)	
ATRX			–
Wild	9 (56.3%)	5 (55.6%)	
Mutated	0 (–)	0 (–)	
NA	7 (43.8%)	4 (44.4%)	
CDKN2A/B			–
Wild	9 (56.3%)	5 (55.6%)	
Mutated	0 (–)	0 (–)	
NA	7 (43.8%)	4 (44.4%)	
PIC3CA			0.788
Wild	9 (56.3%)	4 (44.4%)	
Mutated	0 (–)	1 (11.1%)	
NA	7 (43.8%)	4 (44.4%)	
PTEN			0.733
Wild	8 (50.0%)	4 (44.4%)	
Mutated	1 (6.3%)	1 (11.1%)	
NA	7 (43.8%)	4 (44.4%)	
APC			0.757
Wild	8 (50.0%)	5 (55.6%)	
Mutated	1 (6.3%)	0 (–)	
NA	7 (43.8%)	4 (44.4%)	

Unless otherwise specified, data are numbers of cases

AA, Anaplastic astrocytoma; DA, Diffuse astrocytoma; EGFR, Epidermal growth factor receptor; GBM, Glioblastoma; NA, Not available; WHO, World Health Organization

*$p < 0.05$, statistically significant difference

Fig. 3 Tile panel demonstrating molecular characteristics by next-generation sequence analysis of patients with histone H3 K27-mutant diffuse non-midline glioma (n = 16) and H3 G34-mutant diffuse hemispheric glioma (n = 9) in the Kansai Network

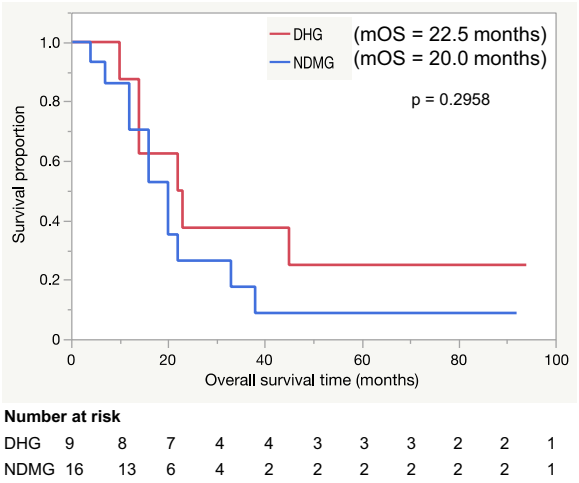

Fig. 4 Kaplan–Meier survival curves of patients with histone H3 K27-mutant diffuse non-midline glioma (n = 16) and H3 G34-mutant diffuse hemispheric glioma (n = 9) in the Kansai Network

Methylation analysis

Data regarding DNA methylation was available for eight patients (KNBTG_144, 663, 223, 833, 658, 239, 813 and 786) (Fig. 5). The DNA methylation-based CNS brain tumor classifier (v.12.8) of five NDMGs (KNBTG_144, 663, 223, 833, and 658) yielded scores of 0.99774, 0.95398, 0.18381, 0.86607 and 0.99983 for the diagnosis of DMG H3 K27-altered, respectively (Fig. 5). The t-SNE plot for five NDMGs and references showed that the NDMG group clustered close to the reference DMG_K27 group (Fig. 5). On the other hand, the DNA methylation-based CNS brain tumor classifier (v.12.8) of three DHGs (KNBTG_239, 813, and 786) yielded a score of 0.99894, 0.99733 and 0.96369 for the diagnosis of DHG H3 G34-mutant, respectively (Fig. 5). The t-SNE plot for three DHGs and references showed that the DHG group clustered close to the reference GBM_G34 group (Fig. 5).

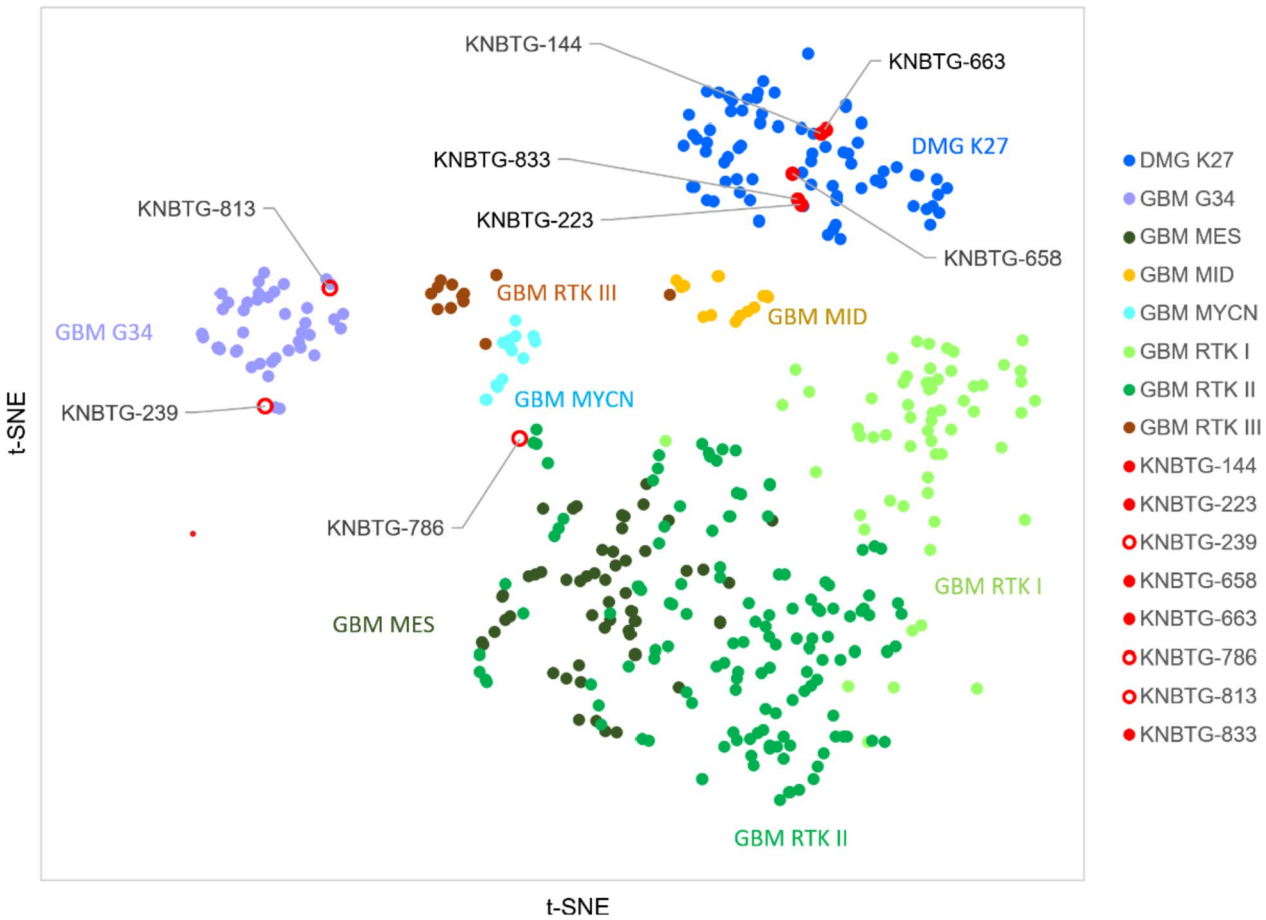

Fig. 5 t-SNE analysis of genomic DNA methylome of eight patients (KNBTG_144, 663, 223, 833, 658, 239, 813 and 786) with histone H3 K27-mutant diffuse non-midline glioma and H3 G34-mutant diffuse hemispheric glioma in the Kansai Network

Discussion

Summary of the present study

Diffuse gliomas of cerebral hemisphere could possess *H3F3A* gene mutation such as K27M as well as G34R/V. Both H3 K27M-mutant NDMG and G34-mutant DHG are rare high-grade gliomas that correspond to WHO CNS grade 4, but we consider the difference between these non-midline tumors to be the main area of investigation. In this comparative study, we revealed characteristics and outcomes of these tumors. DHG mostly arose in adolescents and young adults (AYA), while NDMG mostly arose in older adults. There could be a male predominance in both tumors. Frequent tumor locations were the frontal and temporal lobes in the patients with NDMG and the temporal and parietal lobes in the patients with DHG. In the present cohort, modest surgical resection followed by radiation and TMZ chemotherapy was commonly undertaken for both types of tumors (Table 1). We found the overall survival times of patients with NDMG were comparable to those of DHG. Although independent prognostic factors of NDMG or DHG could not be identified in the study cohort, female sex, preoperative KPS score $\geq$ 80 and extensive surgical resection were related to improved prognoses of these tumors (Supplementary Figs. 1, 2, 3).

H3 K27M-mutant NDMG in the literature

In the Kansai Network dataset, we found 16 cases of H3 K27M-mutant NDMG. To the best of our knowledge, only 40 cases of H3 K27M-mutant non-midline gliomas have previously been reported in the literature. Although one study stated that these tumors tend to involve the adult population (age range: 20–76 years old, mean age: 47 years old), [4] another reported four pediatric cases (age range: 5–19 years old) [13]. Age distribution of the cases in the Kansai Network was thought to be consistent with that in the former report (age range: 26–79 years old, mean age: 45 years old). As for sex, there might be a male predilection considering the reported case series (5 women and 7 men) and the results of our study (6 women and 10 men). Tumor locations generally include the frontal, temporal and parietal lobes, the insula, and the corpus callosum. The basal ganglia, in which the tumor was found in three patients in our cohort, was not included in the previous reports. As in our cohort, there are a few case reports of multifocal lesions with or without midline structures [13].

Molecular analysis revealed that all cases were proven IDH wildtype and H3K27M-mutant. p53 immunostaining and/or *TP53* mutation was not infrequently observed in the present cohort. Although a previous study reported that the tumors expressing the *TP53* mutation had a poorer OS in young patients with DMG, we did not find significantly poorer OS in those with *TP53* mutation in our cohort [5]. Interestingly, DNA methylation analysis showed these H3 K27M-mutant NDMGs possess similar DNA methylation property to H3 K27M-mutant DMGs (Fig. 4). Our study therefore suggests that H3 K27M-mutant NDMG had equivalent molecular characteristics to the H3 K27M-mutant DMG.

Survival data was available in four of 12 reported cases (follow-up period: 3–42 months, median 10.8 months); these four patients died with a mOS of 7.8 months (range: 3–21.9 months) [4]. In the Kansai Network, mOS was 20.0 months and thus longer than that reported in the literature. Corpus callosum involvement has been suggested to be a negative prognostic factor and potentially related to the limited resectability of tumors involving the corpus callosum [4]. Difference of survival data between reported cases and our cohort may be due to the limited resectability of tumors, so further study in a larger cohort is necessary.

Comparison between NDMG and DMG with H3 K27M mutation

All things considered, our retrospective analysis and recent reports may suggest that diffuse gliomas with H3 K27M mutation have the potential to arise in non-midline area (NDMG) rather than selectively arising in midline lesion (DMG), although with low frequency. Consequently, there may be major concerns regarding clinical and molecular differences between DMG and NDMG. However, the literature, which includes the CNS WHO 2021 blue book, suggests that there may be a few different features between them [16]. As described above, NDMG may occur in an older population than DMG. A recent comprehensive genomic study revealed that *FGFR1* mutations were exclusively identified in H3K27M-mutant DMGs (64/304, 21%); these tend to occur in older patients (median age: 32.5 years) [17]. However, in the present study there was only one patient with NDMG (1/16, 6.3%); *FGFR1* mutations may be a rare event in NDMG. Independent of the anatomic location, neuropathological tumor grading (e.g., WHO grade III and IV) and EOR, the prognosis of H3 K27M-mutant diffuse gliomas is thought to be poor [10]. As shown in this study, however, overall survival time of NDMG, similar to DHG in our cohort, may be longer than that of DMG, which reportedly has a shorter survival time than DHG [15]. Additionally, the possible surgical options for NDMG could prolong the prognosis of the patients compared with that in patients with DMG. Another study based on a Kansai Network cohort is currently ongoing with the aim of clarifying the survival differences between NDMG and DMG seen in the Japanese cohort,

and in the near future we hope to address these clinical questions.

H3 G34R/V-mutant DHG in the literature

In addition to NDMG with H3 K27M mutation, we also found nine cases of H3 G34-mutant DHG in the Kansai Network dataset. According to CNS WHO 2021, DHG typically affect the AYA generation (median age: 15–19 years); in the Kansai Network cohort, the patients belong to a somewhat older AYA population [16]. As observed in the present study, there seems to be a male predominance, with a male:female ratio of 1.4:1 [16]. Histopathologically, DHG typically has a GBM features; in our cohort, all except for one case showed GBM-like high-grade glioma appearance regardless of necrosis or microvascular proliferation according to central pathology review. As molecularly defining diagnostic criterion, G34R is identified in > 94% of cases, and G34V in < 6%, although there are no reported differences between p.G34R and p.G34V mutations [16]. *MGMT* is often methylated, as also found in our study cohort. About 90% of tumors bear *TP53* mutations and approximately 95% have alterations in the *ATRX* gene [16]. *TP53* mutations were at the same frequency in the present study. As for *TERT*, only one case has the mutation, although there is no pertinent information in the CNS WHO 2021 blue book. The prognosis for patients with DHG is poor, with a mOS of 18–22 months, and mOS of the Kansai Network cohort was 23.1 months. The present study cohort is too small to confirm the survival difference, but *MGMT* methylation and absence of oncogene amplifications may be associated with longer OS [16].

Differences between K27M and G34R/V-mutant tumors

Several clinicopathological differences between K27M and G34R/V-mutant tumors have been reported in the literature [15, 16]. K27M-mutant tumors are located in the midline region, whereas G34R/V-mutant tumors are typically located in the hemispheric region [18]. Patients with K27M mutation tend to be younger and have clinically poorer prognosis than the patients with G34R/V alteration [18]. However, we demonstrated some of K27M-mutant tumors located in the hemispheric region, which cannot be defined by CNS WHO 2021 criteria. Based on the results of our comparative study, patients with this tumor tended to be older and had similar prognosis to those with G34R/V alteration. Our DNA sequencing and methylation analysis also suggested NDMGs with H3 K27M mutation have generally identical molecular properties to DMG with H3 K27M mutation, whereas they are different from features of H3 G34-mutant DHGs, although both tumors involve hemispheric regions. Clinicopathological features of 'diffuse hemispheric glioma with K27M mutation' will be elucidated in the further investigation.

Limitations

There are several limitations concerning this study; it uses a retrospective cohort design and unlike a complete survey, there could be some degree of selection bias, and the selection could affect the molecular and survival findings. The limited number of patients could explain the absence of statistical power to detect differences between groups. The heterogeneity in treatment regimens and data collection may introduce bias. Molecular characteristics might be different between those in the Kansai Network and those in Western cohorts, although the reason is unclear. Other than racial differences, the different techniques used in previous reports might be also be a consideration. Additionally, no independent prognostic factors were identified, potentially due to the limited cohort size and variability in clinical management. Addressing these limitations in future prospective multicenter studies with larger cohorts and standardized protocols will aim to strengthen the findings.

Conclusion

We reviewed the clinical and molecular characteristics and prognosis of diffuse hemispheric gliomas with histone H3 K27M mutation (NDMG with H3 K27M mutation) or G34R/V mutation (H3 G34-mutant DHG) in the Kansai Network cohort. H3 K27M-mutant NDMG may be an undetermined entity in the literature. Diffuse gliomas with H3 K27M mutation have the potential to arise in non-midline areas (NDMG) rather than selectively arising in midline lesion (DMG), although their molecular properties may be almost identical to H3 K27-mutant DMG. Both NDMG with H3 K27M mutation and H3 G34-mutant DHG are rare tumors and involve non-midline, hemispheric lesions. However, these two entities have different clinical and molecular characteristics. Even for older adult patients, histone H3 mutation should be examined for precise diagnostics of cerebral diffuse gliomas. Future comprehensive analysis is needed to better understand the biology and the clinical significance of these hemispheric gliomas with H3 mutations.

Abbreviations

BEV	Bevacizumab
DHG	Diffuse hemispheric glioma
DMG	Diffuse midline glioma
EOR	Extent of surgical resection
GBM	Glioblastoma
GTR	Gross total resection
HR	Hazard ratio
Kansai Network	Kansai Molecular Diagnosis Network for CNS Tumors
KPS	Karnofsky performance status
MGMT	O-6-methylguanine-DNA methyltransferase
mOS	Median overall survival

MRI	Magnetic resonance image
Mut	Mutated
NDMG	Diffuse non-midline glioma
OS	Overall survival
PR	Partial resection
SNVs	Single nucleotide variants
STR	Subtotal resection
TERT	Telomerase reverse transcriptase
TMZ	Temozolomide
TP53	Tumor protein p53
t-SNE	T-distributed stochastic neighbor embedding
VASARI	Visually AcceSAble Rembrandt Images

Supplementary Information

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Additional file 1.

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Author contributions
HN, JF, HK, NH, KM, YoKa: Study design. HN, JF, HK, NH, KN, TU, YA, SK, NK, KI, TO, YS, SO, HA, MM, TF, TT, AI, TA, HiKa: Data collection. YoKo: Pathology review. EY, DK, AK, MS, TS, MM: Sample analysis. HN, JF, NH, HK, KM, YoKa: Data analysis. HN, JF, YoKa: Interpretation. HN, JF, YoKa: Manuscript writing. CI, KM, YoKa: Supervision. All authors read and approved the final manuscript.

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Availability of data and materials
The datasets analyzed in the current study are available from the corresponding authors upon reasonable request.

Declarations

Ethics approval and consent to participate
All procedures performed in studies involving human participants were in accordance with the ethical standards of the institutional and/or national research committee and the 1964 Helsinki declaration and its later amendments or comparable ethical standards. The study was approved by the Institutional Review Boards of the Seiri Hamamatsu General Hospital (No. 2453), the Wakayama Medical University Hospital (No. 98), the Osaka National Hospital (No. 713), and those of all collaborative institutes. Written informed consent was obtained from all patients.

Consent for publication
Not applicable.

Competing interests
The authors declare that they have no competing interests.

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References

1. Bhagwate AV, Liu Y, Winham SJ, McDonough SJ, Stallings-Mann ML, Heinzen EP et al (2019) Bioinformatics and DNA-extraction strategies to reliably detect genetic variants from FFPE breast tissue samples. BMC Genom 20(1):689. https://doi.org/10.1186/s12864-019-6056-8
2. Breiman L (2001) Random forests. Mach Learn 45:5–32. https://doi.org/10.1023/A:1010933404324
3. Capper D, Jones DTW, Sill M, Hovestadt V, Schrimpf D, Sturm D et al (2018) DNA methylation-based classification of central nervous system tumours. Nature 555(7697):469–474. https://doi.org/10.1038/nature26000
4. Chia N, Wong A, Teo K, Tan AP, Vellayappan BA, Yeo TT et al (2021) H3K27M-mutant, hemispheric diffuse glioma in an adult patient with prolonged survival. Neurooncol Adv 3(1):vdab135. https://doi.org/10.1093/noajnl/vdab135
5. Damodharan S, Abbott A, Kellar K, Zhao Q, Dey M (2023) Molecular characterization and treatment approaches for pediatric H3 K27-altered diffuse midline glioma: integrated systematic review of individual clinical trial participant data. Cancers 15(13):3478. https://doi.org/10.3390/cancers15133478
6. Ding W, Kaur D, Horvath S, Zhou W (2023) Comparative epigenome analysis using Infinium DNA methylation BeadChips. Brief Bioinform 24:1–12. https://doi.org/10.1093/bib/bbac617
7. Guidara S, Seyve A, Poncet D, Leonce C, Bringuier PP, McLeer A et al (2024) Characteristics of H3K27M-mutant diffuse gliomas with

a non-midline location. J Neurooncol. https://doi.org/10.1007/s11060-024-04733-z
8. Hayashi N, Fukai J, Nakatogawa H, Kawaji H, Yoshioka E, Kodama Y et al (2024) Neuroradiological, genetic and clinical characteristics of histone H3 K27-mutant diffuse midline gliomas in the Kansai Molecular Diagnosis Network for CNS Tumors (Kansai Network): multicenter retrospective cohort. Acta Neuropathol Commun 12(1):120. https://doi.org/10.1186/s40478-024-01808-w
9. Kanda Y (2013) Investigation of the freely available easy-to-use software 'EZR' for medical statistics. Bone Marrow Transpl 48:452–458. https://doi.org/10.1038/bmt.2012.244
10. Karremann M, Gielen GH, Hoffmann M, Wiese M, Colditz N, Warmuth-Metz M et al (2018) Diffuse high-grade gliomas with H3 K27M mutations carry a dismal prognosis independent of tumor location. Neuro-oncology 20(1):123–131. https://doi.org/10.1093/neuonc/nox149
11. La Rocca G, Sabatino G, Altieri R, Signorelli F, Ricciardi L, Gessi M et al (2019) Significance of H3K27M mutation in "Nonmidline" high-grade Gliomas of cerebral hemispheres. World Neurosurg 131:174–176. https://doi.org/10.1016/j.wneu.2019.08.024
12. van der Maaten L (2014) Accelerating t-SNE using tree-based algorithms. J Mach Learn Res 15(93):3221–3245
13. Onishi S, Ohba S, Kuraoka K, Kurashige T, Sugiyama K, Yamasaki F (2022) Molecular and clinical characterization of H3 K27M-mutant "non-midline" glioblastoma: a case report and literature review. Neurocirugia (Astur: Engl Ed) 33(6):356–360. https://doi.org/10.1016/j.neucie.2021.06.008
14. Schwartzentruber J, Korshunov A, Liu XY, Jones DT, Pfaff E, Jacob K et al (2012) Driver mutations in histone H3.3 and chromatin remodelling genes in paediatric glioblastoma. Nature 482(7384):226–231. https://doi.org/10.1038/nature10833
15. Sturm D, Witt H, Hovestadt V, Khuong-Quang DA, Jones DTW, Konermann C et al (2012) Hotspot mutations in H3F3A and IDH1 define distinct epigenetic and biological subgroups of glioblastoma. Cancer Cell 22:425–437. https://doi.org/10.1016/j.ccr.2012.08.024
16. WHO Classification of Tumours Editorial Board: Central Nervous System Tumours; WHO Classification of Tumours, 5th ed, IARC, Lyon (2021)
17. Williams EA, Brastianos PK, Wakimoto H, Zolal A, Filbin MG, Cahill DP et al (2023) A comprehensive genomic study of 390 H3F3A-mutant pediatric and adult diffuse high-grade gliomas, CNS WHO grade 4. Acta Neuropathol 146:515–525. https://doi.org/10.1007/s00401-023-02609-6
18. Yoshimoto K, Hatae R, Sangatsuda Y, Suzuki SO, Hata N, Akagi Y et al (2017) Prevalence and clinicopathological features of H3.3 G34-mutant high-grade gliomas: a retrospective study of 411 consecutive glioma cases in a single institution. Brain Tumor Pathol 34:103–112. https://doi.org/10.1007/s10014-017-0287-7
19. Wegscjeid ML, Jenning JW (2024) VASARI Glioma Features: insights into their impact and performance. Radiol Imaging Cancer 6(5):e249018

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