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Molecular grouping and outcomes of young children with newly diagnosed ependymoma treated on the multi-institutional SJYC07 trial

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Abstract

Background. This report documents the clinical characteristics, molecular grouping, and outcome of young children with ependymoma treated prospectively on a clinical trial.

Methods. Fifty-four children (aged ≤3 y) with newly diagnosed ependymoma were treated on the St Jude Young Children 07 (SJYC07) trial with maximal safe surgical resection, 4 cycles of systemic chemotherapy, consolidation therapy using focal conformal radiation therapy (RT) (5-mm clinical target volume), and 6 months of oral maintenance chemotherapy. Molecular groups were determined by tumor DNA methylation using Infinium Methylation EPIC BeadChip and profiled on the German Cancer Research Center/Molecular Neuropathology 2.0 classifier.

Results. One of the 54 study patients had metastases (cerebrospinal fluid positive) at diagnosis. Gross or near-total resection was achieved in 48 (89%) patients prior to RT. At a median follow-up of 4.4 years (range, 0.2–10.3 y), 4-year progression-free survival (PFS) was 75.1% ± 7.2%, and overall survival was 92.6% ± 4.4%. The molecular groups showed no significant difference in PFS (4-year estimates: posterior fossa ependymoma group A [PF-EPN-A; 42/54], 71.2% ± 8.3%; supratentorial ependymoma positive for v-rel avian reticuloendotheliosis viral oncogene homolog A [ST-EPN-RELA; 8/54], 83.3% ± 17.0%; and supratentorial ependymoma positive for Yes-associated protein [4/54], 100%, $P = 0.22$). Subtotal resection prior to RT was associated with an inferior PFS compared with gross or near-total resection (4-year PFS: 41.7% ± 22.5% vs 79.0% ± 7.1%, $P = 0.024$), as was PF-EPN-A group with 1q gain ($P = 0.05$). Histopathologic grading was not associated with outcomes (classic vs anaplastic; $P = 0.89$).

Conclusions. In this prospectively treated cohort of young children with ependymoma, ST-EPN-RELA tumors had a more favorable outcome than reported from retrospective data. Histologic grade did not impact outcome. PF-EPN-A with 1q gain and subtotal resection were associated with inferior outcomes.

Key Points

1. Retrospective studies have identified molecular based high-risk ependymoma groups.
2. In our study, outcomes did not differ by molecular group or tumor histology grade.
3. Children with posterior fossa A ependymoma with tumor 1q gain had inferior survival.

Importance of the Study

Advances in ependymoma biology have identified 9 molecularly defined groups. Retrospective studies suggest that children with PF-EPN-A or ST-EPN-RELA tumors experience inferior outcomes. Additionally, gain of chromosome 1q in PF-EPN-A tumors has been associated with shorter PFS. We report the clinical characteristics, molecular grouping, and outcomes of young children (≤3 y at diagnosis) treated on the multi-institutional SJYC07 trial (NCT00602667). Our study prospectively validated (i) the presence of 3 molecular groups of ependymoma in this age group and (ii) the inferior outcomes of PF-EPN-A tumors with 1q gain. Patients with ST-EPN-RELA, though, have favorable outcomes with combined modality therapy in contrast to published retrospective data. Chemotherapy facilitated repeat tumor resection in those with a less than gross total resection prior to radiation therapy. Subtotal resection prior to radiation therapy was an adverse prognostic factor for the entire cohort, and for the PF-EPN-A group. Outcomes did not differ by histologic grading.

Ependymal tumors account for 5.1% of all CNS tumors in infants, children, and adolescents aged 0–19 years.[1] In children, 90% of ependymomas occur intracranially; approximately 75% are located in the posterior fossa (PF), and the remaining are located within the supratentorial (ST) region of the brain.[2–4] The 5-year overall survival (OS) is 65–85% among all children, with those aged 3–5 years or younger reported to have lower OS of approximately 55–60%.[2,3,5,6] Several studies have demonstrated favorable outcome in patients treated with gross total resection (GTR) and postoperative radiation therapy (RT).[2–5,7] Conversely, an increased risk of relapse and shorter survival have been noted in patients with subtotal resection (STR) of their tumor and a PF primary, and in those treated with postoperative chemotherapy only, without RT.[2–6,8,9] The benefit of adjuvant chemotherapy prior to or after postoperative RT in improving outcomes for children with ependymoma, though, remains undefined.

Tumor DNA-methylation profiling has identified 9 clinically and molecularly distinct groups of ependymoma arising from the 3 anatomic compartments of the CNS.[10] Retrospective studies indicate that certain molecular features, like the gain of chromosome 1q (1q+) in PF A-group ependymoma (PF-EPN-A), and the *C11orf95–RELA*-fused ST group ependymoma (ST-EPN-RELA), have inferior outcomes.[10–12] However, these molecular risk factors need validation in prospective clinical trials.

We report outcomes of children aged 3 years or younger at the time of diagnosis of ependymoma treated on the risk-adapted, multi-institutional St Jude Young Children 07 (SJYC07) trial. Molecular grouping and molecular and clinical risk factors associated with outcome were also identified.

Methods

Study Design

SJYC07 was a phase II, risk-adapted, multi-institutional clinical trial approved by the institutional review boards of St Jude Children's Research Hospital and the 6 other participating hospitals. Written informed consent was obtained from the parents or legal guardians of the study participants. The trial was conducted between November 2007 and April 2017.

Children 3 years of age or younger with newly diagnosed ependymoma (World Health Organization [WHO] grades II/III) and no prior anticancer therapy, other than surgical resection of the tumor, were eligible for the study. Other brain tumor diagnoses that were eligible for enrollment in this trial included medulloblastoma, supratentorial primitive neuroectodermal tumor, pineoblastoma, atypical teratoid/rhabdoid tumor, high-grade glioma, and choroid plexus carcinoma. The outcome data for the ependymoma cohort are presented here.

The diagnosis of ependymoma reported by a pathologist at the enrolling institution was centrally reviewed and classified as WHO grade II (classic) or WHO grade III (anaplastic) ependymoma by the study neuropathologists (D.W.E. and B.A.O.). Study subjects had to have normal organ function and a Lansky score of at least 30, and to begin treatment within 31 days of definitive surgery. Metastatic staging was completed for all patients via MRI of the brain and spine, along with lumbar puncture for cerebrospinal fluid (CSF) analysis (unless medically contraindicated) prior to study enrollment. Children with no evidence of CNS dissemination of their tumor (M0 disease) were enrolled in the

intermediate-risk arm of the study and those with evidence of metastases (M+ disease) were enrolled in the high-risk arm. If a lumbar puncture was not obtained at diagnosis due to medical reasons and there was no imaging evidence of metastases, those subjects were coded as MX (metastases unknown) and treated on the intermediate-risk arm.

Objectives

This report describes the progression-free survival (PFS) and OS of children with ependymoma, which was a secondary cohort of the SJYC07 study, treated with risk-adapted therapy. PFS was defined as the interval from date of treatment initiation to date of relapsed or progressive disease or date of last follow-up. No second malignancies or death before disease relapse or progression occurred in this cohort of patients, so event-free survival and PFS were identical. OS was defined as the time interval from date of treatment initiation to date of death from any cause or to the date of last follow-up for survivors. In this paper we also report results of additional secondary objectives, including the rates of local and/or distant disease progression in ependymoma patients treated with focal RT. The cumulative incidence of local failure was estimated and defined as the time interval from date of treatment start to date of local disease recurrence. Distant recurrence was considered a competing risk.

SJYC07 also included secondary biologic objectives of identifying molecular groups of ependymoma by tumor DNA-methylation analysis to describe outcome by molecular groups. We aimed to determine the prognostic or therapeutic significance of various molecular features.

DNA-Methylation and FISH Analyses

Genomic DNA was extracted from formalin-fixed paraffin embedded (FFPE) tissue samples using the Maxwell RSC DNA FFPE kit (#AS1450, Promega). Genome-wide DNA-methylation profiles were generated from Illumina Infinium MethylationEPIC BeadChip arrays according to the manufacturer's instructions.[13] Data files were uploaded to the German Cancer Research Center/Molecular Neuropathology 2.0 classifier (http://pediatric-neurooncology.dkfz.de/index.php/en/diagnostics/molecular-neuropathology) and results returned for copy number variants and ependymoma molecular group. Tumors were classified as ST-EPN-RELA, ST Yes-associated protein 1 (*YAP1*)–fused (ST-EPN-YAP), PF-EPN-A, or PF-EPN-B.

Multicolor interphase fluorescence in situ hybridization (iFISH) was performed on 5 μm FFPE sections as previously described.[14] Probes were derived from bacterial artificial chromosome (BAC) clones (BACPAC Resources), labeled with either Alexa Fluor 488 or Rhodamine fluorochromes. The following BACs were used to assess copy number abnormalities on chromosome 1: EXO1 at 1q43, RP11-610O24; 1p control, CTD-3241G19.

Treatment Plan

All study subjects underwent a maximal safe surgical resection at diagnosis. The extent of surgical resection was determined by the operating surgeon and postoperative MRI. The categories of surgical resection were defined by postoperative imaging as follows: GTR, when there was no residual tumor; near-total resection (NTR), when the size of residual tumor was <1 cm^2; and STR, when the size of residual tumor was ≥1 cm^2.

Following maximal safe surgical resection, all participants were to receive 4 cycles of induction chemotherapy that included i.v. high-dose methotrexate (5 g/m^2 for patients older than 31 days at enrollment or 2.5 g/m^2 for those younger than 31 days at enrollment given over 24 hours with leucovorin rescue), vincristine, cisplatin, and cyclophosphamide. In addition, patients enrolled in the high-risk arm received low-dose vinblastine (Figure 1 and Supplementary Figure 1). Audiograms for monitoring ototoxicity secondary to cisplatin and dose modifications in the presence of hearing loss were done as reported previously.[15] Patients with M0 disease whose resection at diagnosis was less than GTR were considered for a second-look resection after 2 or 4 cycles of induction chemotherapy to achieve GTR before starting RT.

Induction chemotherapy was followed by risk-adapted consolidation therapy:

1. Patients with intermediate-risk disease who were 1 year or older at the completion of induction therapy received focal RT to the tumor bed. The gross tumor volume was defined as the postoperative tumor bed. The clinical target volume (CTV) included an anatomically confined margin of 5 mm. The planning target volume included a 3 mm geometric expansion of the CTV. The protocol-specified dose to the planning target volume was 54 Gy/cobalt gray equivalent (CGE). Passively scattered proton therapy or 3D conformal or intensity-modulated photon RT methods were permitted.
2. Patients with intermediate-risk disease but younger than 1 year at the time of completion of induction chemotherapy received cyclophosphamide (1.5 g/m^2 i.v, day 1), carboplatin (area under the curve 5 mg/mL/min i.v., day 2), and etoposide (100 mg/m^2 i.v., days 1–2) for two 4-week cycles to delay focal RT until the patient was 1 year old.
3. Patients with high-risk disease were to receive either i.v. topotecan and cyclophosphamide or optional craniospinal irradiation, if they were at least 3 years old by the end of induction.

Maintenance chemotherapy consisted of oral cyclophosphamide and topotecan as low-dose (metronomic) anti-angiogenic therapy, alternating with erlotinib therapy against the ERBB family of transmembrane receptors (Supplementary Figure 1). Oral maintenance chemotherapy was to begin within 2 weeks of completing RT.

Treatment was to be continued until completion of regimen, a diagnosis of progressive or relapsed disease, unacceptable toxicity, or parental withdrawal of consent. Toxicities were graded per the Common Terminology Criteria for Adverse Events (CTCAE) v4.0, except hearing loss, which was graded by the Chang system.

Statistical Methods

As per the intent to treat design, all eligible patients with ependymoma who received at least one dose of

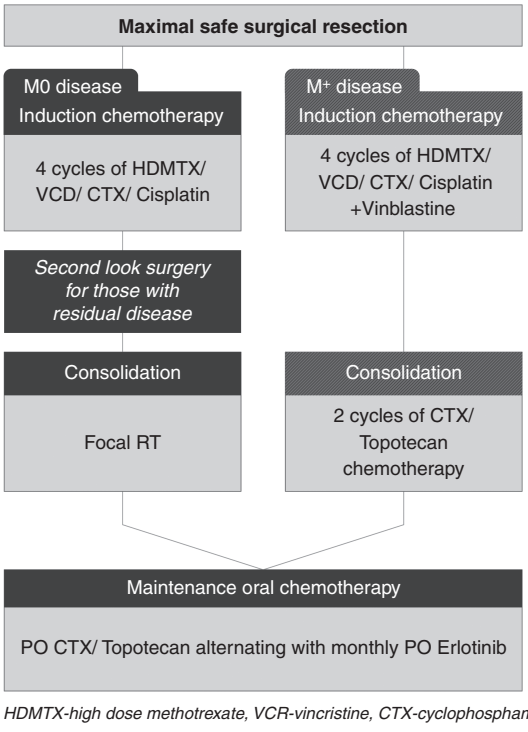

Fig. 1 Schematics of the study design.

the first induction chemotherapy were included in the outcome and safety analyses, and all subjects with adequate tissue for methylation profiling were included in the biological analyses. Outcome distributions were estimated using the Kaplan–Meier method. Mean outcome estimates are reported ±1 SE, which was obtained using the Peto and Pike method. Log-rank tests were used for outcome comparisons. Although the study permanently closed to accrual in April 2017, long-term follow-up is ongoing.

Results

Patient Characteristics

Fifty-four subjects with classic or anaplastic ependymoma were enrolled in the trial between November 2007 and April 2017. The median age at the time of diagnosis was 1.5 years (range, 0.4–3.0 y). One patient had evidence of CSF dissemination at diagnosis on 2 separate lumbar puncture analyses. CSF could not be obtained at diagnosis from 13 patients due to safety concerns. These 13 patients were coded as MX (metastatic state unknown) and treated on the intermediate-risk stratum. No patient had imaging evidence of tumor dissemination. Thus, 53 (98%) patients were treated on the intermediate-risk stratum and 1 on the high-risk stratum. Most patients had anaplastic tumors (76%) and a PF primary site (78%) (Table 1).

All patients were off treatment at the time of this analysis. Twenty-nine (54%) completed the entire treatment regimen on the trial. Twenty-five (46%) patients discontinued treatment early: 19 per family requests (3 during induction, 4 at the end of induction, 6 after RT, 2 after consolidation chemotherapy, and 4 during maintenance), 3 due to treating physician request (1 each after 2 or 4 cycles of induction, and 1 after 3 cycles of maintenance), and 3 due to progressive disease (2 during induction and 1 after 2 cycles of maintenance) (Supplementary Figure 2). Forty-eight patients received RT: proton beam therapy in 26 and photon in the remaining 22.

Tumor Molecular Features

Methylation group information was available for all patients. The majority (42; 77.8%) of ependymomas, all located in the posterior fossa, were classified as PF-EPN-A. Eight (14.8%) were classified as ST-EPN-RELA, and 4 (7.4%) were ST-EPN-YAP. All PF-EPN-A tumors demonstrated a loss of H3K27-trimethylation by immunohistochemistry, thus providing additional proof of their group affiliation.[16] Three of the 4 patients with an ST-EPN-YAP tumor were younger than 1 year at diagnosis (Supplementary Table 1).

Outcomes

Forty-six (85%) patients were alive at the time of analysis (November 2018), with a median follow-up of 4.4 years (range, 0.2–10.3 y). The 4-year estimates of PFS and OS for the cohort were 75.1% ± 7.2% and 92.6% ± 4.4%, respectively, and we continued to observe failures beyond this timepoint with 7-year PFS and OS estimates of 54.8% ± 11.7% and 79.5% ± 9.6% (Figure 2A). The median follow-up of patients with ST-EPN-RELA was 3.6 years (range, 1.1–10.3 y); for those with PF-EPN-A, 5.0 years (range, 0.2–9.4); and for those with ST-EPN-YAP, 4.5 years (range, 2.7–6.1). There were no significant differences in outcome by molecular group (Figure 2B and C). The single patient with a positive CSF at diagnosis was treated with 4 cycles of induction chemotherapy, after which the family opted out of the trial and pursued focal RT. This patient has not experienced treatment failure to date, approximately 6 years from the initial diagnosis. There was no difference in outcome between patients with M0 disease and those with MX disease ($P = 0.34$ for PFS and $P = 0.99$ for OS), and by type of radiation used (photon vs proton) ($P = 0.67$ for PFS and $P = 0.89$ for OS).

Outcomes by Histology and Chromosome 1q Status in the PF-EPN-A Group

PFS and OS of the entire cohort did not differ by tumor grade (Figure 2D and E). Additionally, outcomes did not differ by tumor grade in the PF-EPN-A group (Supplementary Figure 3). We identified 1q+ in tumors by iFISH at diagnosis in 5 of 42 (12%) patients with PF-EPN-A tumor. We found a difference in PFS but not in OS by tumor 1q status at diagnosis for the PF-EPN-A cohort (4-year PFS

Table 1. Demographic and clinicopathologic characteristics of 54 subjects at diagnosis

Characteristics	*n* (%)
Sex	
Female	23 (43)
Male	31 (57)
Age at enrollment	
<1 y	15 (28)
1–3 y	39 (72)
Race	
Native American/Inuit	1 (2)
Asian	2 (4)
Black	6 (11)
Mixed race	4 (7)
White	37 (69)
Other/unknown	4 (7)
Tumor location	
Supratentorial	12 (22)
Infratentorial (posterior fossa)	42 (78)
Metastatic status	
M0	40 (74)
M1	1 (2)
MX	13 (24)
Histology	
WHO grade II (classic)	13 (24)
WHO grade III (anaplastic)	41 (76)
Risk stratification	
Intermediate	53 (98)
High	1 (2)
Extent of resection at diagnosis	
Biopsy	1 (2)
GTR	29 (54)
NTR	12 (22)
STR	12 (22)

M1, metastatic disease—ie, CSF dissemination; MX, metastatic state unknown (ie, CSF not obtained).

60.0% ± 21.9% [1q+] vs 72.9% ± 8.7% [no 1q+], $P = 0.05$; and 4-year OS 80.0% ± 17.9% [1q+] vs 92.4% ± 5.3% [no 1q+], $P = 0.21$) (Figure 3A and B). Four of 5 patients with 1q+ at diagnosis experienced disease recurrence: 2 patients had recurrence relatively early (1.01 and 1.85 y from treatment initiation) and the other 2 at later timepoints (4.15 and 5.42 y from treatment initiation). Two of the 4 with 1q+ had a local failure, and the remaining 2 had metastatic failures. In addition, 3 patients whose tumors were not 1q+ at diagnosis, both by iFISH and copy number variation analysis on tumor DNA methylation, were found to be 1q+ by iFISH at relapse.

Outcomes by the Extent of Resection Prior to Focal Radiation Therapy

At the time of diagnosis, 29 (54%) patients had tumor GTR. Twelve patients each had NTR or STR, and 1 patient had a biopsy. Fifteen patients had repeat resections pre-RT (7 post-course # 2 of induction chemotherapy, 1 post-course # 3, and 7 post-course # 4). Twelve of the 15 procedures resulted in GTR. Effectively, 41 (76%) of the subjects had GTR; 7 (13%) had NTR; and 6 (11%) had STR prior to receiving focal RT (Table 1 and Supplementary Table 1). Patients who had GTR or NTR before focal RT had a better PFS (but not OS) than did those with STR ($P = 0.024$ for PFS and $P = 0.078$ for OS) (Figure 4A and B). We also observed this difference in PFS among the subset of patients with PF-EPN-A who underwent GTR/NTR versus STR but not in OS ($P = 0.034$ for PFS and $P = 0.073$ for OS) (Figure 4C and D). Of note, only 5 patients with PF-EPN-A had an STR.

Outcomes Among Children Who Were Younger Than 1 Year at Diagnosis

Fifteen (28%) patients in our cohort were younger than 1 year of age at the time of study enrollment. The group affiliations for these infant tumors were: 9 (60%) PF-EPN-A, 3 (20%) ST-EPN-RELA, and 3 (20%) ST-EPN-YAP. Ten patients received RT on protocol therapy. Three additional patients received RT off protocol prior to progression. The median age at the start of RT was 15.1 months (range, 12.9–17.2 mo). One patient received RT at disease progression, after the family initially refused RT and opted to treat with chemotherapy alone. Another patient who received only chemotherapy currently shows no evidence of disease. This patient's tumor demonstrated a *YAP1* fusion and clustered with the ST-EPN-YAP group by methylation. Four infants had progressive disease at 0.2, 1.0, 3.7, and 6.0 years from the start of treatment; 2 of them subsequently died. There was no difference in outcomes for children younger than 1 year versus 1 year or older at study enrollment (Supplementary Figure 4).

Treatment Failures

Eighteen patients experienced progressive or recurrent/relapsed disease. The median time to failure of therapy was 2.3 years (range, 1.7 mo–7.3 y). Disease recurrence was local ($n = 8$), distant ($n = 9$), or combined ($n = 1$). The cumulative incidence of local failure was 12.7% ± 5.0% at 4 years. There were no differences in the cumulative incidence of local failure ($P = 0.81$) or distant failure ($P = 0.99$) by type of radiation used (photon vs proton). The one patient with a combined local and distant recurrence was considered as having local recurrence for this analysis. There were a few late treatment failures in our cohort: 4 patients had events occur more than 5 years after the start of therapy. Eight patients died after treatment failure. The median time from progressive disease to last follow-up for the 10 patients alive after treatment failure was 1.1 years (range, 0.9 mo–5.8 y). The events and patterns of treatment failure are detailed in Table 2.

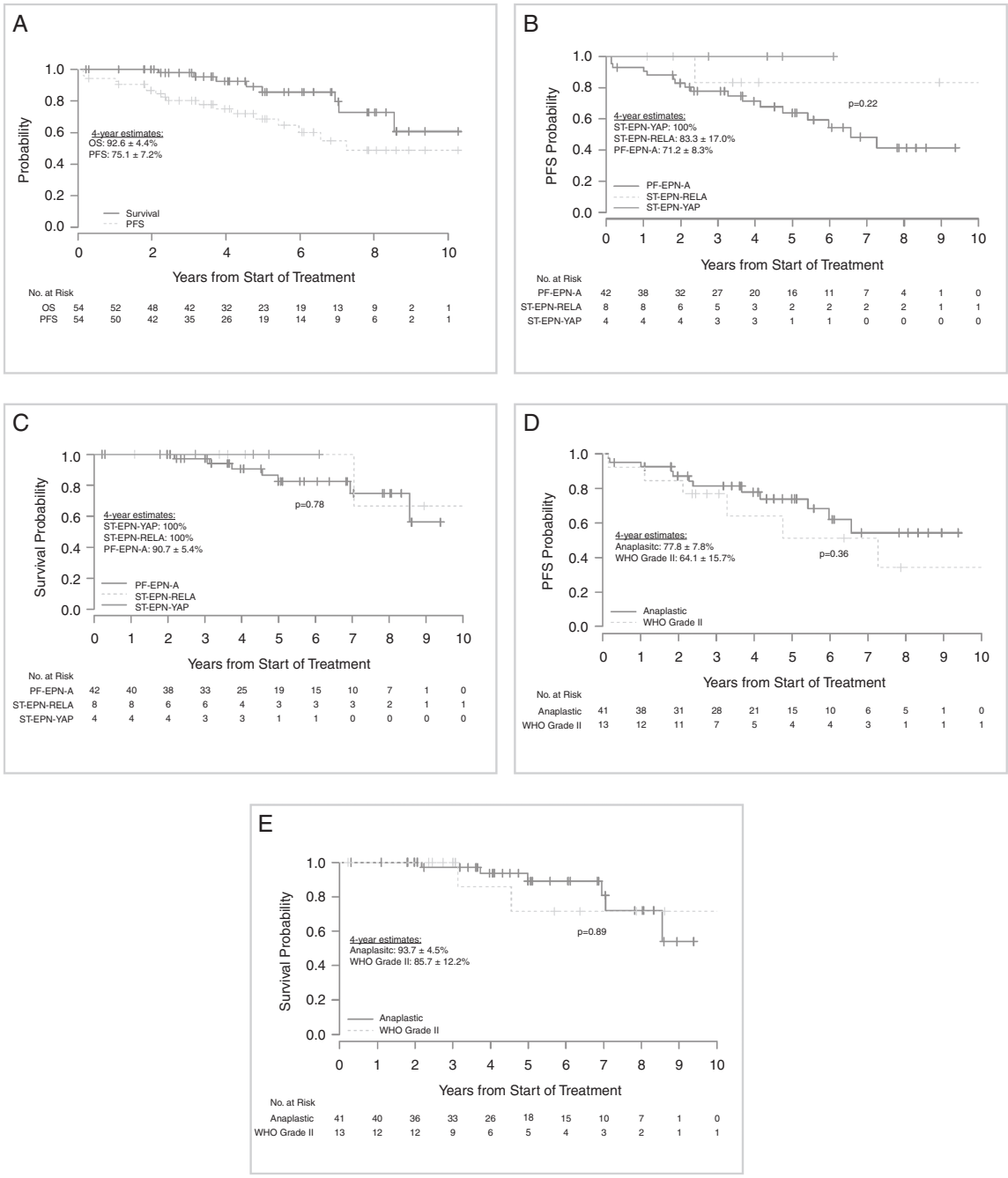

Fig. 2 (A) PFS and OS of all 54 study subjects. Four-year PFS (B) and OS (C) by ependymoma molecular groups. PFS (D) and OS (E) grouped by histology (classic vs anaplastic).

Toxicity

No deaths were caused by toxicity. Two patients suffered symptomatic radiation necrosis (grade 3 per CTCAE v4.0) at 2.4 and 4.3 months after the completion of proton beam RT; both were treated with hyperbaric oxygen therapy and had resolution of the imaging findings. The first patient had mild residual hemiparesis at the last follow-up visit, 3.5 years since the diagnosis of radiation necrosis, and the second patient had complete recovery of motor deficits at 4 months from the diagnosis of the necrosis. Febrile neutropenia and grades 3 and 4 myelosuppression were the most common adverse events (Supplementary Table 2). Forty-seven patients had baseline hearing evaluations prior to start of therapy, 1 of whom had Chang grade 1a and 5 had Chang grade 2b or higher unilateral hearing loss. Of the 39 patients who did not have any hearing loss at baseline and had hearing evaluation during and after

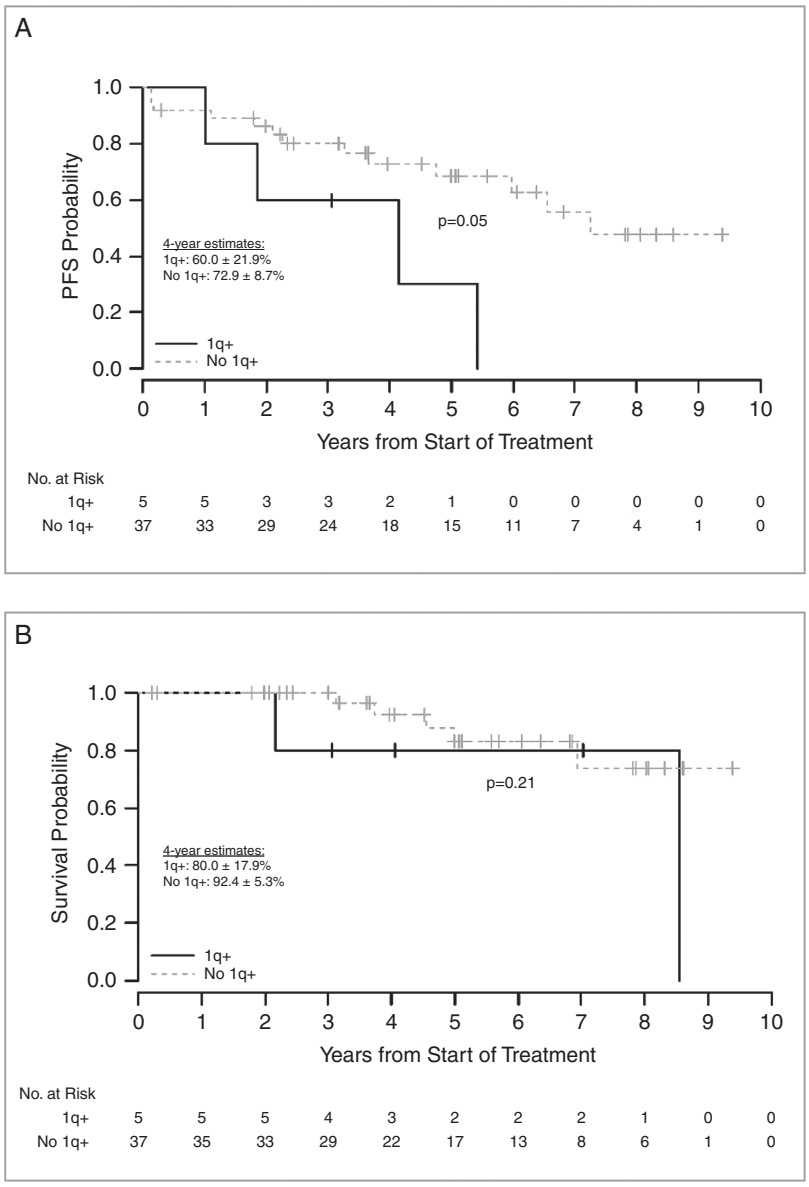

Fig. 3 (A) PFS and OS (B) of the PF-EPN-A group by tumor 1q status at diagnosis.

treatment, 11 (28%) developed Chang grade 2b or higher hearing loss.

Discussion

Our prospective multicenter study demonstrates no significant difference in outcomes for children with newly diagnosed ependymoma by molecular groups. Children with PF-EPN-A with 1q gain had inferior PFS. Based on the results of historical studies, maximal safe surgical resection followed by focal RT has emerged as the treatment of choice for most children with localized ependymoma.[17] Children who undergo GTR prior to RT have demonstrated the best outcomes in several studies.[2,3,6–8,18–20] Although adjuvant chemotherapy alone in the absence of RT can produce durable outcomes in a small percentage of children without residual disease, the role of chemotherapy in addition to surgery and RT remains to be determined.[5,6,21]

Pajtler and colleagues have described 9 molecularly distinct groups of ependymoma, with PF-EPN-A, PF-EPN-B, ST-EPN-RELA, and ST-EPN-YAP being the predominant groups in children.[10] Their study suggests that molecular stratification is a more reliable predictor of response to therapy than is histologic grading. A study by Ellison et al has demonstrated considerable interobserver variability in the pathologic classification of childhood intracranial ependymomas as WHO grade II or WHO grade III.[22] Pajtler et al also found molecular features (eg, ST-EPN-RELA and

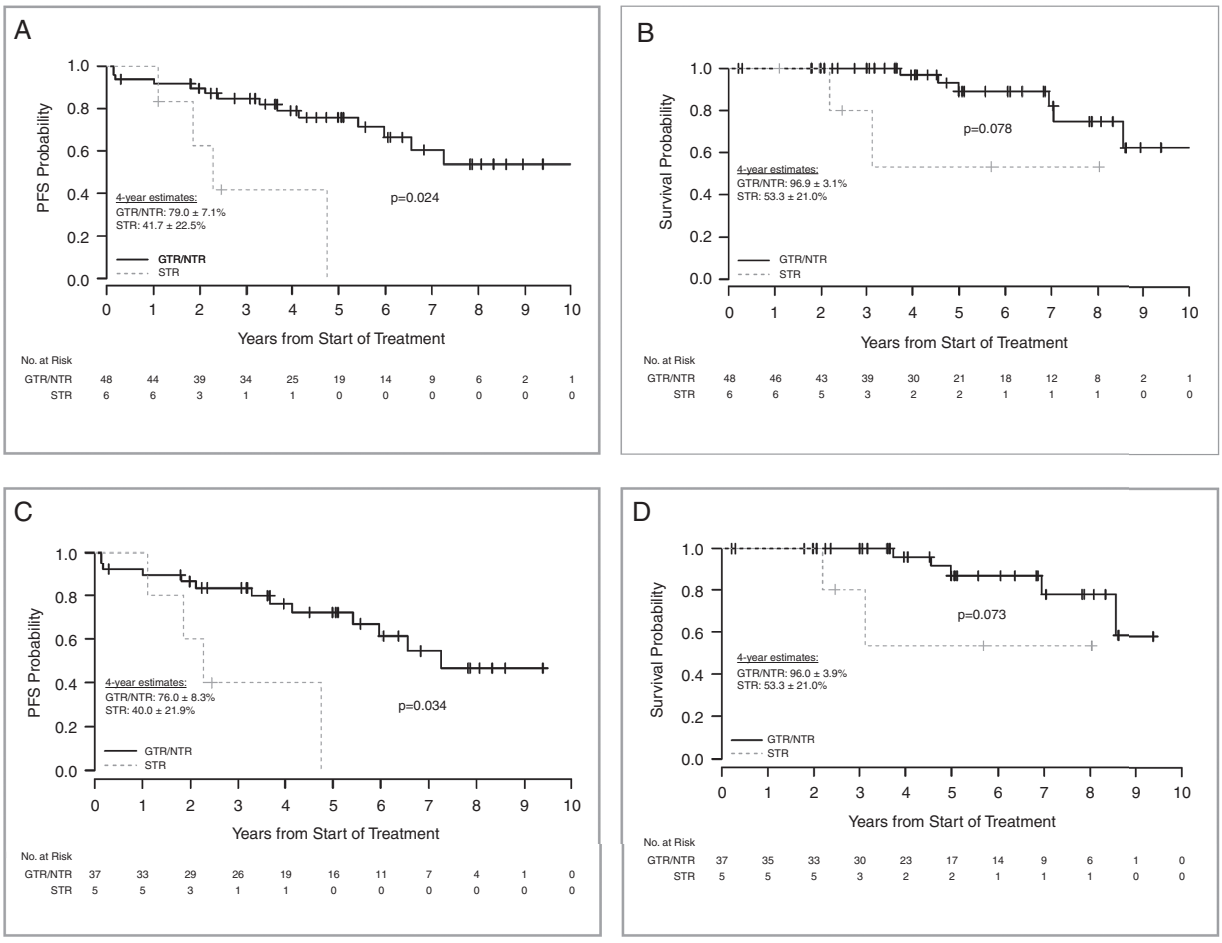

Fig. 4 PFS (A) and OS (B) of the 54 subjects by extent of surgical resection prior to RT. PFS (C) and OS (D) of the 42 subjects with PF-EPN-A by extent of surgical resection prior to RT.

tumor 1q+ in PF-EPN-A) that are prognostic, as indicated in some earlier studies.[11,12,23]

As expected, PF was the most common location for primary tumors in our cohort, and all PF tumors belonged to the PF-EPN-A group. This finding corroborates the observation that PF-EPN-A is the predominant group in infants and young children, and PF-EPN-B tumors are seen in older children and adolescents.[24–26] The results of our trial also affirmed the earlier finding that the extent of resection is one of the most important determinants of treatment success in children with localized ependymoma.[2,6–8,19,20] GTR of PF tumors, however, is challenging because permanent neurologic sequelae are possible, even in the hands of the most experienced pediatric neurosurgeons. The use of chemotherapy before surgical re-resection of any residual tumor has the potential to aid these efforts and achieve a more complete resection before administration of RT.[27] Our results demonstrate the utility of postoperative adjuvant chemotherapy in facilitating a repeated resection in subjects who received less than GTR. Approximately half of the subjects had a GTR at study enrollment. The administration of postoperative chemotherapy enabled 15 additional patients to undergo a second resection of the residual tumor prior to RT and resulted in a GTR in 12 of these patients. Thus, 89% of the total cohort either had a GTR or an NTR prior to planned RT. The induction chemotherapy regimen was administered safely and was well tolerated, with no therapy-related deaths. This favorable outcome associated with the extent of resection was also apparent in the PF-EPN-A group; children who had a GTR showed significantly improved PFS. This finding reinforces the need to consider a repeat resection of any residual tumor prior to RT, and administration of adjuvant chemotherapy could aid the re-resection. In the absence of necessary expertise at the treating hospital, due consideration should be given to referring children with less than GTR of the tumor to a larger center with an experienced pediatric neurosurgical team for repeat tumor resection.

Neither the PFS nor the OS differed by molecular grouping in our study. This result contrasts with the report of dismal outcomes among patients with ST-EPN-RELA by Pajtler and colleagues.[10] This difference can be attributed to several possible reasons. First, we had a small

Table 2 Treatment failure in 18 study subjects

Patient #	Molecular Subgroup	Histology	WHO Grade	Extent of Tumor Resection Prior to RT	Gain of 1q at Diagnosis	Site of recurrence or Progressive Disease	Time to Treatment Failure, years	Time from Treatment Failure to Last Follow-Up, years	Survival Status
1	PF-EPN-A	Anaplastic	III	GTR	No	Local	0.14	4.84	Dead
2	PF-EPN-A	Anaplastic	III	GTR	Yes	Distant	5.42	3.13	Dead
3	PF-EPN-A	Anaplastic	III	GTR	No	Local	5.97	0.97	Dead
4	PF-EPN-A	Classic	II	STR	No	Distant	1.10	2.03	Dead
5	PF-EPN-A	Classic	II	GTR	No	Distant	7.26	1.35	Alive
6	PF-EPN-A	Anaplastic	III	GTR	No	Combined	3.66	0.07	Dead
7	PF-EPN-A	Classic	II	STR	No	Local	4.75	0.94	Alive
8	PF-EPN-A	Anaplastic	III	GTR	Yes	Local	4.15	2.89	Alive
9	ST-EPN-RELA	Anaplastic	III	NTR	Not applicable	Distant	2.38	4.67	Dead
10	PF-EPN-A	Classic	II	GTR	No	Distant	2.12	0.89	Alive
11	PF-EPN-A	Classic	II	GTR	No	Local	0.14	0.08	Alive
12	PF-EPN-A	Anaplastic	III	GTR	No	Local	0.18	3.87	Alive
13	PF-EPN-A	Anaplastic	III	STR	No	Distant	2.27	5.75	Alive
14	PF-EPN-A	Classic	II	GTR	No	Local	3.28	1.27	Dead
15	PF-EPN-A	Anaplastic	III	STR	Yes	Distant	1.85	0.33	Dead
16	PF-EPN-A	Anaplastic	III	GTR	Yes	Local	1.01	3.05	Alive
17	PF-EPN-A	Anaplastic	III	GTR	No	Distant	6.56	0.3	Alive
18	PF-EPN-A	Anaplastic	III	NTR	No	Distant	1.81	0.25	Alive

number of patients in the ST-EPN-RELA group, thus limiting our outcome analysis. Second, the median duration of follow-up in our study was relatively short. This is important because of the possibility of late recurrences and treatment failures in ependymoma well documented in the literature.[3,28,29] Therefore, longer follow-up is warranted to determine if these results would change. Third, retrospective study results, like those reported by Pajtler et al, have inherent limitations due to the heterogeneity of the cohort studied. Hence, a larger cohort of uniformly treated and prospectively followed subjects with ST-EPN-RELA will be required to evaluate long-term outcomes in this group of ependymoma. Finally, the role of adjuvant chemotherapy in not only facilitating repeat resections but also improving outcomes, as in our study, cannot be ruled out. A definitive assessment of this question is currently under way via randomized studies in larger cohorts of patients, such as the current Children's Oncology Group (COG) trial ACNS0831 (ClinicalTrials.gov identifier NCT01096368) and the SIOP-EP-II studies (ClinicalTrials.gov identifier NCT02265770). Our findings are consistent with the superior survival reported for the ST-EPN-YAP group. All 4 subjects in the ST-EPN-YAP group are alive without recurrence of their tumor; 2 were treated with chemotherapy alone and both are disease free at 3 and 5 years from diagnosis. It is abundantly clear, though, that PF and ST ependymomas are biologically distinct diseases with unique molecular alterations, and hence caution must be exercised when comparing outcomes across different molecular groups.[17]

Several studies have demonstrated tumor 1q+ to be a poor prognostic factor for the PF-EPN-A group.[10–12,23,30,31] The results from the COG trial ACNS0121 demonstrated the importance of 1q+ as a prognostic factor.[32] In patients treated with immediate postoperative radiation therapy, the 5-year event-free survival was 82.2% for 1q– versus 47.4% for 1q+. The differences in OS were also significant: the 5-year OS was 91.3% for 1q– versus 68.4% for 1q+. The impact of 1q+ was greatest in the PF-EPN-A group. Our results demonstrate that PFS but not OS was worse for PF-EPN-A patients with 1q+. This difference in survival between PFS and OS by 1q status likely reflects a lack of effective salvage therapies for patients with relapsed or recurrent ependymoma, irrespective of their molecular classification and risk factors. A similar discrepancy in outcomes by the presence of 1q+ was also noticed by Fukuoka and colleagues.[33] In our cohort, 3 patients in the PF-EPN-A group who did not have 1q+ at the time of diagnosis had tumors harboring this aberration at relapse. One possible explanation for the intriguing finding of the emergence of 1q+ at relapse is that small clones of cells with 1q+ were present from onset but at a level below the threshold for detection, and those clones subsequently expanded at relapse, in the face of treatment resistance.

Although the role of RT in treating ependymoma is unequivocal, the optimal dose to be administered and CTV margin remain under debate. To reduce the risk of radiation-specific and combined-modality toxicity, the SJYC07 trial was designed to limit the primary site dose to 54 Gy/CGE and the CTV margin to 5 mm. The rationale for

the protocol-specified dose and target volume parameters was the successful use of a 10 mm CTV margin and 54 Gy in children younger than 18 months after GTR in a previous trial.[3] The ability to reduce the targeted volume was considered an important research question and logical next step. The study, though, was not powered to determine if the dose of radiation could be safely reduced to 54 Gy without compromising survival. However, the possibility of limiting the radiation dose to 54 Gy/CGE in children with ependymoma should be considered in the design of future clinical trials, when treatment can be stratified by both clinical and molecular features. Indeed, the proportion of patients with specific molecular features may affect the results of series attempting to report differences in outcome based on RT parameters. In the COG trial ACNS0121, there was a significant association between 1q+ status and local and distant patterns of failure.[32]

Approximately half the patients in our study who experienced treatment failure had only a metastatic recurrence. Previous reports have suggested local failures as the predominant mode of disease recurrence in children with ependymoma.[2,4,7,8,18,19,28,29,34] However, other studies have suggested a higher incidence of metastatic or distant failures.[3,35] Indelicato and colleagues reported a higher percentage of distant failures in their study of 179 patients with nonmetastatic disease; they also found that GTR/NTR prior to RT was correlated with a higher percentage of local control.[35] In addition, a higher percentage of subjects in the study who underwent STR experienced local failure. A similar finding was reported by Merchant and colleagues.[3] It is very likely, given the historical data of best outcomes for pediatric ependymoma with a GTR, that aggressive attempts at a GTR contribute to the pattern of predominantly distant treatment failures. However, it is pertinent to understand the biology of ependymoma and determine if molecular factors contribute to a pattern of metastatic failures in some patients. A case in point is the study by Tsang et al, who treated relapsed ependymoma with repeat RT. They found a higher proportion of distant or combined (local and distant) treatment failure in subjects with a PF primary with 1q+.[36]

As our understanding of ependymoma biology evolves, the heterogeneity within the PF groups and the prognostic significance of 1q+ continue to unravel.[24,31] Given the small number of patients with 1q+ in our study, we were unable to determine the prognostic implication of this finding with respect to the site(s) of treatment failure. Our study also demonstrated the risk of late treatment failures in pediatric patients with ependymoma, which reinforces the need for prolonged surveillance of these patients, as suggested by other studies.[3,28,29]

Our study has its limitations. A high proportion (46%) of the total patients in our study did not complete all the planned treatment as per the trial. It is, however, pertinent to note that this discontinuation was primarily after the completion of 4 cycles of induction chemotherapy, and includes those that completed focal RT off trial and patients who came off study due to family requests, and treatment failures. The historical reports of limited benefit from chemotherapy for patients with ependymoma likely contributed to the refusal of maintenance chemotherapy by families. Additionally, since only 29 patients completed all the trial mandated therapy, we are unable to determine the role of oral maintenance chemotherapy in improving survival outcomes for our cohort.

Conclusions

In a uniformly treated, prospective cohort of children 3 years or younger with newly diagnosed ependymoma, we found no significant difference in outcome by molecular group or histologic grade. Tumors harboring 1q+ in the PF-EPN-A group had inferior PFS, validating the importance of molecular grouping in this disease. Chemotherapy can be used to enable a maximal safe surgical resection with a goal of GTR before RT and to delay RT in infants until they are at least 1 year of age. Extent of resection remains predictive of treatment failure, with patients who underwent STR before RT having inferior outcomes compared with those with NTR/GTR, which was demonstrable in the PF-EPN-A group as well. Close surveillance and follow-up of patients with ependymoma beyond the traditional 5 years from diagnosis is warranted due to the risk of late disease progression. Our findings are expected to help improve risk stratification on future trials by integrating the most important aspects of molecular grouping and clinical risk factors for pediatric ependymoma.

Supplementary Material

Supplementary data are available at *Neuro-Oncology* online.

Keywords

1q gain | chemotherapy | clinical target volume | ependymoma groups

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