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Original Article

Effectiveness and Safety of Dabrafenib in the Treatment of 20 Chinese Children with *BRAF*^{V600E}-Mutated Langerhans Cell Histiocytosis

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Purpose We sought to investigate the effectiveness and safety of dabrafenib in children with *BRAF*^{V600E}-mutated Langerhans cell histiocytosis (LCH).

Materials and Methods A retrospective analysis was performed on 20 children with *BRAF*^{V600E}-mutated LCH who were treated with dabrafenib.

Results The median age at which the patients started taking dabrafenib was 2.3 years old (range, 0.6 to 6.5 years). The ratio of boys to girls was 2.3:1. The median follow-up time was 30.8 months (range, 18.9 to 43.6 months). There were 14 patients (70%) in the risk organ (RO)$^+$ group and six patients (30%) in the RO$^-$ group. All patients were initially treated with traditional chemotherapy and then shifted to targeted therapy due to poor control of LCH or intolerance to chemotherapy. The overall objective response rate and the overall disease control rate were 65% and 75%, respectively. During treatment, circulating levels of cell-free *BRAF*^{V600E} (cf*BRAF*^{V600E}) became negative in 60% of the patients within a median period of 3.0 months (range, 1.0 to 9.0 months). Grade 2 or 3 adverse effects occurred in five patients.

Conclusion Some children with *BRAF*^{V600E}-mutated LCH may benefit from monotherapy with dabrafenib, especially high-risk patients with concomitant hemophagocytic lymphohistiocytosis and intolerance to chemotherapy. The safety of dabrafenib is notable. A prospective study with a larger sample size is required to determine the optimal dosage and treatment duration.

Key words Langerhans cell histiocytosis, *BRAF*^{V600E}, Dabrafenib, Children

Introduction

Langerhans cell histiocytosis (LCH) is a rare disease characterized by the proliferation of specific dendritic cells arising from myeloid progenitor cells and is currently considered a clonal disease [1]. LCH presents with a spectrum of clinical manifestations [2]. Low-risk children are affected in only a single organ or system, with a certain degree of self-limitation, and first-line chemotherapy is effective. High-risk patients are often under 3 years old and frequently have multiple organs or systems involved, especially the liver, spleen, or hematologic system. Thus, the risk of treatment-related death or disease progression is higher in the high-risk group than in the low-risk group [3,4].

BRAF kinase, which is encoded by the *BRAF* gene, plays a significant role in the RAS–mitogen-activated protein kinase (MAPK) signaling pathway. It was reported that the oncogenic *BRAF*^{V600E} mutation was identified in 35 of 61 LCH specimens (57%) and in up to 87.8% of children with multisystem- and risk-organ-involved LCH [4,5]. This mutation can drive

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constitutive ERK activation and lead to impaired migration and apoptosis in pathological LCH cells, which gives rise to LCH [1,6].

Dabrafenib is a reversible, ATP-competitive, selective BRAF kinase inhibitor [7] with a low incidence of adverse effects [8,9]. The role of dabrafenib in the treatment of *BRAF*^{V600E}-mutated LCH has been tested in a clinical trial (NCT01677741). The most common adverse events of dabrafenib are skin-related toxic effects (including maculopapular rash, skin pain and severe events including squamous cell carcinoma and keratocanthoma), fever, fatigue, arthralgia, etc. [10].

This study was designed to analyze the effectiveness and safety of dabrafenib in the treatment of 20 children with *BRAF*^{V600E}-mutated LCH who were cared for at our center.

Materials and Methods

1. Patients

From November 1, 2016, to November 30, 2018, 20 children (≤ 18 years old) with LCH who fulfilled all of the following criteria were enrolled in this study: (1) diagnosed with LCH according to clinical features, positive staining of CD1a and/or Langerin (CD207) of biopsy tissue; (2) *BRAF*^{V600E} could be detected in the peripheral blood or affected tissue at the onset of the disease; (3) chemotherapy could not be tolerated due to serious condition or severe chemotherapy-related adverse effects, or the disease continued to progress after chemotherapy, or the pituitary lesion was not improved after chemotherapy; and (4) no other BRAF kinase inhibitors had been used previously. In addition, Eastern Cooperative Oncology Group performance status 0-2 was a key inclusion criterion. All patients were followed until June 1, 2020.

2. Patient stratification

According to the involvement of risk organs, patients were divided into a risk organ-involved group (RO^+ or high-risk group) or a risk organ-noninvolved group (RO^- or low-risk group). According to the cf*BRAF*^{V600E} level at the end of targeted therapy, patients were divided into a negative mutation group or a positive mutation group.

3. Therapeutic regimen

All patients were treated with chemotherapy according to the same protocol modified from LCH-III (without high-dose methotrexate) and HS-LCH salvage treatment after diagnosis, including first-line (vindesine+prednisone) and second-line treatment (cytarabine+vindesine+dexamethasone+/–cladribine). The details of the treatment elements are shown in S1 Table.

Targeted therapy with dabrafenib was initiated after the discontinuation of first-line or second-line treatment due to disease progression or intolerance to chemotherapy. For some patients not complicated by hemophagocytic lymphohistiocytosis (HLH), the disease was not severe enough, so dabrafenib was usually chosen after evaluation, which was performed one week after the discontinuation of chemotherapy. Patients with HLH were always in critical condition. Thus, if there was no improvement observed after chemotherapy, the patients underwent targeted therapy as soon as possible.

The dosage of dabrafenib was 2 mg/kg according to dose escalation and dose limiting toxicity evaluation in a phase I/II study on dabrafenib in children with refractory/resistant *BRAF*^{V600E} solid tumors [11], and for the treatment of pediatric *BRAF*^{V600E} mutated high-grade gliomas [12], dabrafenib was administered orally once every 12 hours. The general duration of the course of dabrafenib treatment was 6 months to 1 year, adjusted according to disease assessment and patient tolerance to the drug. The condition and adverse effects were evaluated after 1 month, 3 months, and every 3 months thereafter. After targeted therapy was completed, maintenance chemotherapy was given (6-mercaptopurine+vindesine+prednisone). This regimen has been registered as a clinical trial (ChiCTR2000032844).

4. Determination of *BRAF*^{V600E} in tissue and plasma

The *BRAF*^{V600E} mutation was initially detected in tissue biopsies from 14 patients. Genomic DNA was extracted from 10×5 μm unstained sections of formalin-fixed paraffin-embedded (FFPE) tissue using the QIAamp DNA FFPE Tissue Kit (Qiagen, Hilden, Germany). The presence of the *BRAF*^{V600E} mutation was then determined using a droplet digital polymerase chain reaction (ddPCR) assay with a QX200 Droplet Digital PCR system (Bio-Rad, Hercules, CA). FFPE tissues could not be obtained from the other six patients because no remaining tissue sample could be used for *BRAF*^{V600E} detection.

Plasma cell-free DNA from all 20 patients was isolated using the QIAamp Circulating Nucleic Acid Kit (Qiagen). Detection of cf*BRAF*^{V600E} was performed by ddPCR assay with a lower limit of detection of 0.04% and a lower limit of quantitation of 0.1%. The patient with cf*BRAF*^{V600E} level lower than 0.04% was regarded as negative accordingly. The laboratory has been certificated by Beijing Municipal Health and Family Planning Commission as Clinical Gene Amplification Test Laboratory, which is equivalent to Clinical Laboratory Improvement Amendments (CLIA)–approved laboratory.

5. Evaluation of the disease state and treatment response

The disease state and treatment response of the patients were evaluated according to the Histiocyte Society Evalu-

Table 1. Details of each patient's disease characteristics

Case No.	Sex	Age at disease onset (yr)	Age started taking dabrafenib (yr)	Affected organs	Complicated with HLH	Disease state before dabrafenib treatment	Disease state level at the end of dabrafenib treatment	cf*BRAF*^{V600E} level before dabrafenib treatment	cf*BRAF*^{V600E} level at the end of dabrafenib treatment	Duration of dabrafenib treatment (mo)
1	M	1.7	2.9	Skin, bones, lung, liver, ear, lymph nodes	N	AD-Worse	AD-Better	+ (5.00%)	-	3.9
2	M	0.1	2.1	Skin, bones, lung, liver, spleen, ear lymph nodes, thymus	N	AD-Stable	AD-Better	+ (5.00%)	+ (1.73%)	17.7
3	F	1.4	2.3	Skin, bones, lung, liver, spleen, hematopoietic system, lymph nodes	Y	AD-Stable	AD-Better	-	-	3.7
4	M	1.2	3.1	Liver, spleen, ear, pituitary, thyroid	N	AD-Worse	AD-Better	+ (6.31%)	-	6.1
5	F	0.3	0.8	Skin, bones, lung, liver, spleen, hematopoietic system, ear, lymph nodes	Y	AD-Worse	AD-Better	+ (2.80%)	+ (0.51%)	19.2
6	M	1.0	2.1	Skin, bones, liver, spleen, ear	N	AD-Worse	AD-Better	+ (0.20%)	-	6.4
7	M	3.2	4.3	Bones, liver, pituitary, thyroid, biliary tract	N	AD-Worse	AD-Mixed	+ (0.75%)	-	7.0
8	M	1.0	2.4	Bones, pituitary, oral cavity	N	AD-Stable	AD-Better	-	-	9.8
9	M	5.1	6.5	Bones, pituitary	N	AD-Stable	AD-Stable	-	-	12.1
10	M	1.1	2.7	Skin, bones, lung, liver, ear	N	AD-Worse	AD-Better	-	-	13.0
11	F	0.1	0.8	Skin, bones, lung, liver, spleen, hematopoietic system, lymph nodes, thymus	N	AD-Worse	AD-Better	+ (24.05%)	-	18.4
12	M	2.0	2.6	Bones, liver, spleen, ear	N	AD-Worse	AD-Better	+ (0.51%)	-	11.7
13	M	2.5	3.5	Skin, bones, ear, pituitary	N	AD-Worse	AD-Stable	+ (0.27%)	-	11.1
14	F	1.0	3.3	Skin, bones, lung, ear, pituitary	N	AD-Worse	AD-Worse	-	-	4.4
15	M	0.8	0.9	Skin, bones, ear pituitary	N	AD-Worse	AD-Better	+ (3.28%)	+ (0.24%)	14.0
16	M	0.2	0.6	Skin, liver, spleen, hematopoietic system	Y	AD-Worse	AD-Better	+ (3.29%)	+ (1.13%)	16.5
17	F	0.2	0.8	Skin, bones, lung, liver, spleen, hematopoietic system, lymph nodes	Y	AD-Worse	AD-Better	+ (22.52%)	+ (0.13%)	13.8
18	M	0.9	1.1	Skin, lung, liver, hematopoietic system, ear, biliary tract, thyroid	Y	AD-Stable	AD-Worse	+ (1.80%)	+ (0.89%)	10.0
19	M	0.3	1.2	Skin, bones, lung, liver	N	AD-Mixed	AD-Worse	+ (0.18%)	-	18.3
20	F	1.6	2.6	Bones, thyroid	N	AD-Worse	AD-Worse	+ (1.03%)	-	3.1

AD, active disease; F, female; HLH, hemophagocytic lymphohistiocytosis; M, male; N, no; Y, yes.

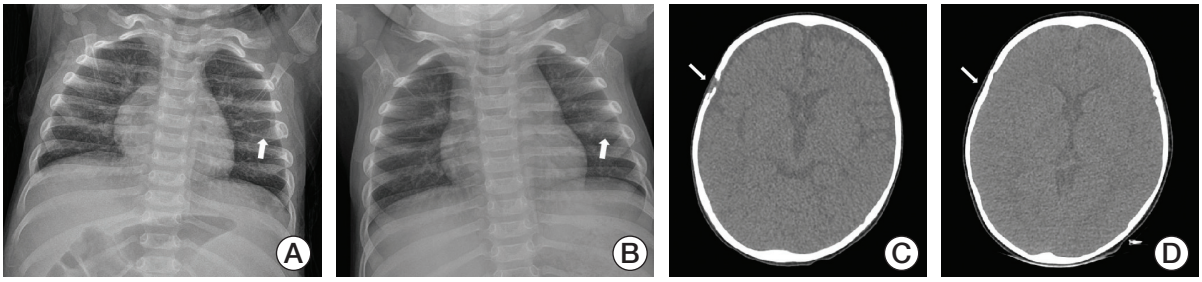

Fig. 1. Improvement of bone lesions (arrows) after dabrafenib treatment. (A, C) Bone lesions in the rib and skull before dabrafenib treatment. (B, D) Improvement of bone lesions after dabrafenib treatment.

Table 2. Details of the evaluation results at each observation point

Observation points	AD-Better	AD-Stable	AD-Mixed	AD-Worse	Drug withdrawal
Month 1	15	2	3	0	0
Month 3	13	2	2	3	0
Month 6	13	2	1	1	3
Month 9	7	1	2	2	8
Month 12	6	1	1	1	11

AD, active disease.

ation and Treatment Guidelines published in 2009 [13,14]. Briefly, the disease states included nonactive disease (NAD) and active disease (AD). The treatment response was categorized as complete resolution (NAD), regression (AD-Better), mixed (AD-Mixed), stable (AD-Stable), and progression (AD-Worse).

For assessment of disease states, imaging modalities including radiography, ultrasound, computed tomography, and magnetic resonance imaging were used. Positron emission tomography computed tomography was used only in cases of inconclusive results from other imaging modalities.

The objective response rate (ORR) was defined as the percentage of patients with AD-Better among all patients, and the disease control rate (DCR) was defined as the percentage of patients with AD-Better and AD-Stable among all patients. Adverse effects were followed from the beginning of dabrafenib treatment through the last contact with the patients and were graded according to the National Cancer Institute Common Terminology Criteria for Adverse Events, ver. 5.0 [15].

6. Statistical analysis

Statistical analysis was performed by using IBM SPSS ver. 24.0 software (IBM Corp., Armonk, NY). The count data were expressed by the number of cases or percentages, and Fisher exact test was used for the categorical variables. The measurement data with a normal distribution are expressed as the mean±standard deviation, while the measurement data with a nonnormal distribution are expressed as the median (minimum-maximum). For continuous variables, data were analyzed using the t test and Mann-Whitney U test, depending on the data distribution. Survival curves were estimated using the Kaplan-Meier method, and the log-rank test was used to estimate the differences among the groups. A p-value of < 0.05 was considered statistically significant.

Results

1. General information of the patients

The median ages of disease onset and initiation of dabrafenib were 1 year (range, 0.1 to 5.1 years) and 2.3 years (range, 0.6 to 6.5 years), respectively. The ratio of boys to girls was 2.3:1 (Table 1). The median follow-up time was 30.8 months (range, 18.9 to 43.6 months).

There were 14 patients (70%) in the RO^+ group and six patients (30%) in the RO^- group. Four patients with disease states of AD-Stable or AD-Worse were treated with dabrafenib because of intolerance to chemotherapy. The remaining 16 patients had disease states of AD-Mixed (1 patient), AD-Stable (3 patients), or AD-Worse (12 patients) due to no improvement in disease or persistent progression (S2A-D, S3A-C, S4A-D, S5 and S6 Figs.). Of these 16 patients, seven patients experienced uncontrolled liver and spleen involve-

Table 3. Details of five patients complicated by HLH[a)]

	Case 3	Case 5	Case 16	Case 17	Case 18
Before dabrafenib					
Temperature (°C)	Normal	Normal	38.6	Normal	39
CRP (mg/L)	24	57	57	62	45
Hemoglobin (g/L)	105	72	74	87	80
Platelet ($\times10^9$/L)	68	271	28	165	172
Neutrophil ($\times10^9$/L)	1.37	5.38	0.84	1.84	0.61
Splenomegaly (below the left costal margin) (cm)	4.0	6.5	5.7	3.0	1.3
Triglyceride (mmol/L)	2.04	4.04	6.57	6.88	5.18
Fibrinogen (g/L)	2.90	0.87	1	1.45	1.16
Hemophagocytosis	+	+	+	+	–
NK-cell activity (%)	13.62	17.01	NA	14.49	11.46
Ferritin (μg/L)	NA	NA	124.2	135.2	98.8
sCD25 (U/mL)	NA	NA	20,725	22,803	14,242
Three days after starting dabrafenib					
Temperature (°C)	Normal	Normal	Normal	Normal	Normal
CRP (mg/L)	< 5	< 5	< 5	8	< 5
Hemoglobin (g/L)	127	112	96	92	91
Platelet ($\times10^9$/L)	180	302	100	347	409
Neutrophil ($\times10^9$/L)	2.40	1.87	0.38	2.24	1.80
Splenomegaly (below the left costal margin)	NA	NA	NA	NA	NA
Triglyceride (mmol/L)	1.33	NA	4.42	3.85	NA
Fibrinogen (g/L)	NA	0.87	1.01	NA	NA
Hemophagocytosis	NA	NA	NA	NA	NA
NK-cell activity (%)	NA	NA	NA	NA	NA
Ferritin (μg/L)	NA	548.9	NA	132.5	NA
sCD25 (U/ml)	NA	NA	NA	NA	NA
One month after starting Dabrafenib					
Temperature (°C)	Normal	Normal	Normal	Normal	Normal
CRP (mg/L)	NA	NA	NA	NA	NA
Hemoglobin (g/L)	127	131	118	118	115
Platelet (10^9/L)	180	283	370	530	567
Neutrophil (10^9/L)	2.40	5.37	3.49	3.12	5.44
Splenomegaly (below the left costal margin) (cm)	3.8	2.3	No splenomegaly	No splenomegaly	No splenomegaly
Triglyceride (mmol/L)	0.7	0.9	1	0.8	2.73
Fibrinogen (g/L)	11.9	NA	NA	3.2	2.36
Hemophagocytosis	NA	NA	NA	NA	NA
NK-cell activity (%)	14.83	NA	NA	NA	NA
Ferritin (μg/L)	NA	521.2	NA	NA	NA
sCD25 (U/mL)	NA	NA	NA	NA	NA

CRP, C-reactive protein; HLH, hemophagocytic lymphohistiocytosis; NA, not available; NK-cell, natural killer cell; sCD25, soluble CD25.
[a)]The diagnosis of HLH can be established if one of either 1 or 2 below is fulfilled: (1) a molecular diagnosis consistent with HLH, (2) diagnostic criteria for HLH fulfilled (five out of the eight criteria below): (a) fever, (b) splenomegaly, (c) cytopenias (affecting ≥ 2 of 3 lineages in the peripheral blood): hemoglobin < 90 g/L (in infants < 4 weeks: hemoglobin < 100 g/L), platelets < 100×10^9/L, neutrophils < 1.0×10^9/L, (d) ferritin ≥ 500 μg/L, (e) hypertriglyceridemia and/or hypofibrinogenemia: fasting triglycerides ≥ 3.0 mmol/L, fibrinogen ≤ 1.5 g/L, (f) hemophagocytosis in bone marrow or spleen or lymph nodes, (g) soluble CD25 (i.e., soluble IL-2 receptor) ≥ 2,400 U/mL, (h) NK-cell activity < 15.11% (according to our laboratory reference).

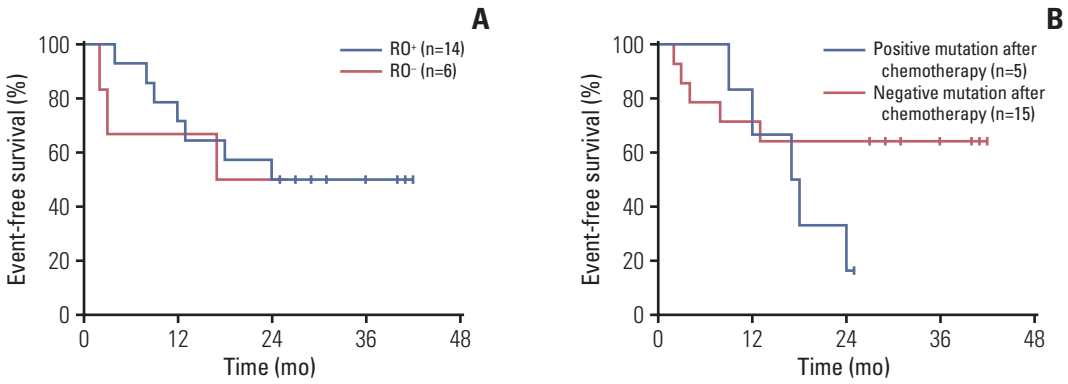

Fig. 2. Survival curves. (A) Survival analysis of the risk organ $(RO)^+$ group and RO^- group. (B) Survival analysis of the positive mutation group or negative mutation group.

ment (hepatomegaly and/or splenomegaly with elevated liver enzymes or serum bile acids), five patients with involvement in bones developed newly affected bone lesions (3 patients) or dilated lesions (2 patients) during chemotherapy (S7A-D, S8A, S8B Figs.), and seven patients had pituitary involvement (S9 Fig.). Some patients chose dabrafenib for more than one reason. In addition, the conditions of five patients in the RO^+ group were complicated by HLH according to the diagnostic criteria of HLH-2004 guidelines [16].

All 20 children were initially treated with first-line chemotherapy for 0.4-13.1 months, with a median of 2.0 months. Subsequently, 12 patients received second-line chemotherapy for 1-14 courses (median of 4 courses). All patients then received dabrafenib treatment. The median duration of dabrafenib treatment was 11.4 months (range, 3.1 to 19.2 months). All patients have since completed dabrafenib monotherapy for 0.6-37.9 months. Ten patients suffered relapse or disease progression.

2. Effectiveness of dabrafenib treatment

Thirteen of the 20 patients were in the AD-Better state at the end of dabrafenib treatment (Fig. 1), with two patients in the AD-Stable state, four patients in the AD-Worse state and one patient in the AD-Mixed state. The overall ORR was 65%, and the overall DCR was 75%. The evaluation results at each observation point can be found in Table 2.

There were 14 patients in the RO^+ group. However, no significant difference in treatment response to dabrafenib treatment was observed between RO^+ and RO^- patients (78.6% vs. 33.3%, p=0.122). Notably, four of the five patients complicated by HLH experienced disease improvement, with rapid increases in temperature, hemogram and C-reactive protein (CRP) observed (Table 3). In two of these patients, the thermal spike decreased after one day of treatment, and body temperature returned to normal within 48 hours. The hemogram and CRP were restored to normal after 3 days of treatment. However, a comparable response to dabrafenib was observed between patients with and without HLH (80% vs. 60%, p=0.613), which might be due to the small sample size.

Among the four patients intolerant to chemotherapy, three patients (75%) experienced improvement, while one patient was in stable condition.

In the seven patients with poor control of the disease in the liver and spleen, five patients (71.4%) experienced improvement in all lesions except hepatic cirrhosis after dabrafenib treatment. The other two patients had only liver enlargement, elevated liver enzymes and no cirrhosis-related manifestations before the usage of dabrafenib. After dabrafenib treatment, both hepatosplenomegaly and liver damage were alleviated.

In four of five patients (80%) treated with dabrafenib due to newly affected bone lesions or dilated lesions in the bones involved during chemotherapy, the imaging findings indicated improvement in the new or dilated lesions after dabrafenib treatment.

Seven patients were treated with dabrafenib because of pituitary involvement (two patients also had progressive lesions, and one patient had an uncontrolled liver lesion), and three of them had diabetes insipidus (DI). No progression was observed by imaging. Only one of the three patients with DI had improvement of symptoms after dabrafenib treatment.

All patients survived, and 10 patients suffered from relapse or progression after dabrafenib treatment. No statistically significant difference in the event-free survival rate was observed between the RO^+ and RO^- groups (χ^2=0.062, p=0.804) (Fig. 2A) or between the positive mutation (at the end of targeted therapy) group and the negative mutation group (χ^2=1.849, p=0.174) (Fig. 2B).

Table 4. Different grades of treatment-related adverse events

Adverse events	Grade	No. of cases[a]	Adverse events occurred after starting targeted treatment (wk)
Maculopapule	1	3	1-16
	2	4	0-2
	3	1	1
Diarrhea	1	2	4
Skin pain	2	1	1
Eye swelling and conjunctival petechia	2	1	4
Blurred vision and photophobia	1	1	4
Vomiting	1	1	5
Ostealgia	1	1	12
Fever	1	2	0-1

[a]Some patients had more than one adverse event.

3. Relationship between dabrafenib treatment and cf*BRAF*^{V600E}

cf*BRAF*^{V600E} has become a promising indicator of treatment response and prognosis in LCH [17,18], so we next investigated the relationship between dabrafenib treatment and cf*BRAF*^{V600E} in our cohort.

Conversion to negative detection of circulating levels of cf*BRAF*^{V600E} after chemotherapy was observed in five patients. Among the other 15 patients with positive cf*BRAF*^{V600E} mutations before targeted therapy, decreased cf*BRAF*^{V600E} levels were observed at the end of targeted therapy (mean, 5.133% vs. 0.309%; t=2.429, p=0.029) (Table 1). Conversion to negative detection was achieved in nine of 15 patients (60%) after treatment with dabrafenib. The median time from dabrafenib treatment to negative detection was 3.0 months (range, 1.0 to 9.0 months). However, the association of persistent positive cf*BRAF*^{V600E} with relapse and/or treatment failure after dabrafenib treatment (83.3% vs. 35.7%, p=0.141) was not observed. This needs to be clarified in future studies with large sample sizes and long-term observations.

In addition, we did not find an association between the duration of dabrafenib treatment and positive or negative cf*BRAF*^{V600E} before targeted therapy (Z=–1.178, p=0.239).

4. Adverse effects and treatments

The most common adverse events of dabrafenib reported in the literature are skin-related toxic effects (including maculopapular rash, skin pain, and severe events, including squamous cell carcinoma and keratocanthoma), fever, fatigue, arthralgia, etc. [11].

In our cohort, 17 adverse events were observed in nine patients. Maculopapular rash was the most common (8 events, 47.1%), including grade 2/3 events in five patients and grade 1 events in three patients. Grade 2 skin pain and eye swelling with conjunctival petechia were observed in two patients, with grade 2/3 maculopapules. Six grade one adverse events were observed in eight patients, including maculopapular rash, diarrhea, blurred vision and photophobia, vomiting, ostealgia, and fever. No other adverse effects were observed in this study (Table 4).

Maculopapules faded in two of five patients after treatment with anti-allergic drugs. The anti-allergic drugs were not effective for the remaining three patients, but the rashes subsided after dabrafenib withdrawal. Skin pain in one patient was relieved by reducing the dabrafenib dosage. Eye swelling and conjunctival petechia in another patient were improved by intensive care of local lesions. Moreover, severe adverse effects of skin, such as squamous cell carcinoma and keratoacanthoma, which are common in dabrafenib treatment of melanoma patients, were not observed, suggesting the safety of dabrafenib in treating children with LCH.

Discussion

A high frequency of the *BRAF*^{V600E} mutation has been found in patients with multisystem and risk-organ-involved LCH [4,5]. Patients with the *BRAF*^{V600E} mutation are less sensitive to the standard first-line treatment, vinblastine combined with corticosteroids, and the incidence of disease recurrence and permanent sequelae are markedly more common [1,3,5]. BRAF kinase inhibitors such as dabrafenib have shown excellent effectiveness in the treatment of *BRAF*^{V600E}-positive malignancies, including melanoma, colon cancer, papillary thyroid cancers, and non-small-cell lung cancer [19-22]. Thus, it is useful to also clarify the role of dabrafenib and other BRAF kinase inhibitors in the treatment of patients with *BRAF*^{V600E}-mutated LCH.

According to the results, children with risk organs involved seem to experience benefits from targeted drugs. However, to confirm this observation, it is necessary to expand the sample size and carry out prospective research with concurrent controls. It is worth noting that patients with HLH generally have risk organ involvement and cannot tolerate strong chemotherapy. Considering the rapid recovery from symptoms and hemogram after targeted treatment, the ineffectiveness of first-line chemotherapy for most patients with concurrent HLH, and the toxic side effects of second-line chemotherapy, it may be better to use targeted drugs as the first-line treatment or for a short period followed by chemotherapy.

The results revealed satisfactory effectiveness of dabra-

fenib in patients who had failed to tolerate chemotherapy. In addition, improvements in liver function were obtained after the administration of dabrafenib in patients without hepatic cirrhosis, whereas cirrhosis that occurred before targeted therapy did not improve after dabrafenib treatment. Our results also showed that dabrafenib relieved DI symptoms in one patient who had developed DI for a half-year; however, no improvement was observed in two other patients who had DI for 1.5 and 2 years. Allen et al. [1] reported that three out of four patients with LCH who were treated with targeted drugs (dabrafenib or vemurafenib) for neurodegeneration (ND), a sequela of LCH due to active pathological process driven by common *BRAF*^{V600E}-positive myeloid precursors, improved in terms of clinical manifestation and imaging findings in relatively early-onset LCH-ND [23]. Thus, these results suggest that dabrafenib could have certain effects on DI (DI, like LCH-ND, is also a sequela of LCH and may be caused by persistent active pathological process as well) at the early stages of this disease, but further study is needed for confirmation.

It has been shown that half of all patients experience relapse or progression, which may be attributed to reactivation of the MAPK pathway upstream of MEK, perhaps resulting from other gene mutations in the pathway [10,24]. Due to their parents' refusal to determine gene mutations at relapse, we cannot identify relapse-causing mutations. Relapse may also be related to low plasma concentration resulting from individual differences in pharmacokinetics or too short a duration of dabrafenib treatment. Thus, this finding highlights the significance of the role of other mutations and plasma concentrations of dabrafenib in relapse. Furthermore, the optimal therapy strategy of dabrafenib, including the time window for treatment and effective combinations with chemotherapy or other targeted drugs in children with LCH, should be explored [7,25,26].

A few patients had treatment-related adverse effects of grade 2 or above. Relief of these adverse effects was achieved after symptomatic treatment, reduction of dosage, or withdrawal. Furthermore, no severe adverse effects (squamous cell carcinoma and keratoacanthoma) were observed. Thus, dabrafenib appears to be safe to some extent for children with LCH.

Due to the limited number of patients and short-term follow-up in this study, we continue to recommend the use of dabrafenib only in patients intolerant of chemotherapy or with disease progression after chemotherapy. The main reason for dabrafenib as a second-line therapy is its unclear long-term safety in the treatment of children with LCH at present. To explore the long-term efficacy and safety of dabrafenib, a prospective study with a larger sample size is required to fully clarify the optimal dosage and duration of dabrafenib treatment.

The major limitations of this study were its retrospective nature and small sample size. A prospective study with a larger sample size and long-term observation is required.

Dabrafenib may have a beneficial effect for some children with *BRAF*^{V600E}-mutated LCH, especially high-risk patients with concomitant HLH and intolerance to chemotherapy. The adverse effects were controllable. However, further research is needed to determine the optimal dosage and duration, long-term efficacy and safety of dabrafenib.

Electronic Supplementary Material

Supplementary materials are available at Cancer Research and Treatment website (https://www.e-crt.org).

Ethical Statement

Written informed consent was obtained from the parents or guardians of the children who served as subjects of the study. This study was conducted in accordance with the Declaration of Helsinki and approved by the Institutional Review Board (IRB) of Beijing Children's Hospital, Capital Medical University (number: 2019-k-109).

Author Contributions

Conceived and designed the analysis: Zhang R.
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Contributed data or analysis tools: Cui L, Zhang Q, Zhang LP.
Performed the analysis: Zhang R, Wang D.
Wrote the paper: Yang Y, Zhang R, Li ZG.
Revised the paper: Zhao XX, Zhao YZ, Li N, Wang TY.

Conflicts of Interest

Conflict of interest relevant to this article was not reported.

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References

1. Allen CE, Merad M, McClain KL. Langerhans-cell histiocytosis. N Engl J Med. 2018;379:856-68.
2. Tran G, Huynh TN, Paller AS. Langerhans cell histiocytosis: a neoplastic disorder driven by Ras-ERK pathway mutations. J Am Acad Dermatol. 2018;78:579-90.
3. Heritier S, Jehanne M, Leverger G, Emile JF, Alvarez JC, Haroche J, et al. Vemurafenib use in an infant for high-risk Langerhans cell histiocytosis. JAMA Oncol. 2015;1:836-8.
4. Heritier S, Emile JF, Barkaoui MA, Thomas C, Fraitag S, Boudjemaa S, et al. BRAF mutation correlates with high-risk Langerhans cell histiocytosis and increased resistance to first-line therapy. J Clin Oncol. 2016;34:3023-30.
5. Badalian-Very G, Vergilio JA, Degar BA, MacConaill LE, Brandner B, Calicchio ML, et al. Recurrent BRAF mutations in Langerhans cell histiocytosis. Blood. 2010;116:1919-23.
6. Abla O, Weitzman S. Treatment of Langerhans cell histiocytosis: role of BRAF/MAPK inhibition. Hematology Am Soc Hematol Educ Program. 2015;2015:565-70.
7. Cebollero A, Puertolas T, Pajares I, Calera L, Anton A. Comparative safety of BRAF and MEK inhibitors (vemurafenib, dabrafenib and trametinib) in first-line therapy for BRAF-mutated metastatic melanoma. Mol Clin Oncol. 2016;5:458-62.
8. Chapman PB, Hauschild A, Robert C, Haanen JB, Ascierto P, Larkin J, et al. Improved survival with vemurafenib in melanoma with BRAF V600E mutation. N Engl J Med. 2011;364:2507-16.
9. Hauschild A, Grob JJ, Demidov LV, Jouary T, Gutzmer R, Millward M, et al. Dabrafenib in BRAF-mutated metastatic melanoma: a multicentre, open-label, phase 3 randomised controlled trial. Lancet. 2012;380:358-65.
10. Hertzman Johansson C, Egyhazi Brage S. BRAF inhibitors in cancer therapy. Pharmacol Ther. 2014;142:176-82.
11. Kieran MW, Geoerger B, Dunkel IJ, Broniscer A, Hargrave D, Hingorani P, et al. A phase I and pharmacokinetic study of oral dabrafenib in children and adolescent patients with recurrent or refractory BRAF V600 mutation-positive solid tumors. Clin Cancer Res. 2019;25:7294-302.
12. Toll SA, Tran HN, Cotter J, Judkins AR, Tamrazi B, Biegel JA, et al. Sustained response of three pediatric BRAF(V600E) mutated high-grade gliomas to combined BRAF and MEK inhibitor therapy. Oncotarget. 2019;10:551-7.
13. Wang D, Cui L, Li ZG, Zhang L, Lian HY, Ma HH, et al. Clinical research of pulmonary Langerhans cell histiocytosis in children. Chin Med J (Engl). 2018;131:1793-8.
14. LCH evaluation and treatment guidelines [Internet]. Pitman, NJ: Histiocyte Society; 2009 [cited 2020 Sep 14]. Available from: https://histiocytesociety.org/document.doc?id=290.
15. Common Terminology Criteria for Adverse Events (CTCAE) [Internet]. Bethesda, MD: National Cancer Institute; 2017 [cited 2020 Sep 14]. Available from: https://ctep.cancer.gov/.
16. Henter JI, Horne A, Arico M, Egeler RM, Filipovich AH, Imashuku S, et al. HLH-2004: Diagnostic and therapeutic guidelines for hemophagocytic lymphohistiocytosis. Pediatr Blood Cancer. 2007;48:124-31.
17. Ozer E, Sevinc A, Ince D, Yuzuguldu R, Olgun N. BRAF V600E mutation: a significant biomarker for prediction of disease relapse in pediatric Langerhans cell histiocytosis. Pediatr Dev Pathol. 2019;22:449-55.
18. Heritier S, Helias-Rodzewicz Z, Lapillonne H, Terrones N, Garrigou S, Normand C, et al. Circulating cell-free BRAF(V600E) as a biomarker in children with Langerhans cell histiocytosis. Br J Haematol. 2017;178:457-67.
19. Schadendorf D, Hauschild A, Santinami M, Atkinson V, Mandala M, Chiarion-Sileni V, et al. Patient-reported outcomes in patients with resected, high-risk melanoma with BRAF(V600E) or BRAF(V600K) mutations treated with adjuvant dabrafenib plus trametinib (COMBI-AD): a randomised, placebo-controlled, phase 3 trial. Lancet Oncol. 2019;20:701-10.
20. Awada G, Seremet T, Fostier K, Everaert H, Neyns B. Long-term disease control of Langerhans cell histiocytosis using combined BRAF and MEK inhibition. Blood Adv. 2018;2:2156-8.
21. Mourah S, Lorillon G, Meignin V, Vercellino L, de Margerie-Mellon C, Pages C, et al. Dramatic transient improvement of metastatic BRAF(V600E)-mutated Langerhans cell sarcoma under treatment with dabrafenib. Blood. 2015;126:2649-52.
22. Hyman DM, Puzanov I, Subbiah V, Faris JE, Chau I, Blay JY, et al. Vemurafenib in multiple nonmelanoma cancers with BRAF V600 mutations. N Engl J Med. 2015;373:726-36.
23. McClain KL, Picarsic J, Chakraborty R, Zinn D, Lin H, Abhyankar H, et al. CNS Langerhans cell histiocytosis: common hematopoietic origin for LCH-associated neurodegeneration and mass lesions. Cancer. 2018;124:2607-20.
24. Robert C, Karaszewska B, Schachter J, Rutkowski P, Mackiewicz A, Stroiakovski D, et al. Improved overall survival in melanoma with combined dabrafenib and trametinib. N Engl J Med. 2015;372:30-9.
25. Yamazaki N, Tsutsumida A, Takahashi A, Namikawa K, Yoshikawa S, Fujiwara Y, et al. Phase 1/2 study assessing the safety and efficacy of dabrafenib and trametinib combination therapy in Japanese patients with BRAF V600 mutation-positive advanced cutaneous melanoma. J Dermatol. 2018;45:397-407.
26. Davies MA, Saiag P, Robert C, Grob JJ, Flaherty KT, Arance A, et al. Dabrafenib plus trametinib in patients with BRAF(V600)-mutant melanoma brain metastases (COMBI-MB): a multicentre, multicohort, open-label, phase 2 trial. Lancet Oncol. 2017;18:863-73.