Pediatric RESEARCH

www.nature.com/pr

BASIC SCIENCE ARTICLE

Clinical features and prognosis of pediatric infradiaphragmatic craniopharyngioma relative to the tumor inflammatory response

Junxiang Peng[1], Lang Yang[1], Jun Pan[1], Chaohu Wang[1], Jing Nie[1], Yi Liu[1], Jun Fan[1], Jie Zhou[2] and Songtao Qi[1]

BACKGROUND: The relationship between clinical responses in pediatric infradiaphragmatic craniopharyngioma (Q-CP) and inflammatory response is still unclear. The objective of this study was to investigate the clinical significance of tumor inflammatory response in pediatric Q-CPs.

METHODS: The inflammatory response was evaluated by measuring the number of inflammatory cells in the tumor near adenohypophysis junction. The specimens were classified as mild, moderate, or severe based on the number of inflammatory cells. In addition, the levels of pro-inflammatory cytokines and chemokines in the specimens were measured using a cytokine antibody array. Clinical outcomes were analyzed and compared to the markers of inflammatory response.

RESULTS: IL-6 and IL-8 were highly expressed in pediatric Q-CPs, and the transcription level of IL-6 was the highest in the severe group. Most patients (87.3%) had hypopituitarism; the severe inflammation group had an increased incidence of hypopituitarism, which correlated with significantly lower probability of recurrence-free survival and worsened functional status.

CONCLUSIONS: Inflammatory response is common in craniopharyngiomas and is closely related to their biological behavior and the patients' clinical prognosis. Further studies of the relationship between craniopharyngiomas and the inflammatory response will enable the discovery of potential therapeutic targets, which will reduce morbidity and result in better outcomes for pediatric Q-CP patients.

Pediatric Research (2021) 89:1119–1125; https://doi.org/10.1038/s41390-020-1013-4

IMPACT:

- Pediatric infradiaphragmatic craniopharyngiomas are histologically benign brain tumors that often follow an aggressive clinical course.
- The inflammatory response in craniopharyngioma is common, which is closely related to the biological behavior and clinical prognosis.
- Several inflammatory and immune markers have been identified in CP; inflammation is an important role in the pathogenesis of hypopituitarism.
- The aim was to study the relationship between craniopharyngioma and inflammatory response and find potential therapeutic targets can reduce morbidity and result in better outcomes.

INTRODUCTION

Craniopharyngiomas (CPs) are benign tumors that are derived from the epithelial remains of Rathke's pouch.[1,2] Infradiaphragmatic CP originates from the infradiaphragmatic, and as its shape is like the letter "Q" in the magnetic resonance imaging (MRI), it was described as Q-type CP (Q-CP) for short in our previous paper.[3,4] Endocrine alterations associated with pediatric Q-CPs have been studied extensively, and inflammation is known to be a key factor in tumor progression.[4] However, none of the existing studies have examined the relationship between clinically significant endocrine changes and inflammatory responses. Using a large cohort of Q-CP patients, we aimed to evaluate the clinical significance of the tumor inflammatory response in pediatric Q-CPs.

METHODS

Patient characteristics

The methodology used in this study was approved by the Committee on Human Research at Nanfang Hospital. One hundred and two Q-CP patients were enrolled from January 2002 to December 2015, and all patients underwent primary surgery at Nanfang Hospital. Fifty-six male and 46 female patients were enrolled, and the average age of the patients at the time of primary surgery was 10.40 ± 4.26 years (range 6–18 years). Clinical and surgical data were recorded in detail for each patient. Tumor size was calculated as the greatest measurable dimension by MRI and classified as follows: small (≤2 cm), moderate (2–4 cm), large (4–6 cm), or massive (>6 cm). The presence of calcification was

[1]Department of Neurosurgery, Nanfang Hospital, Southern Medical University, Guangzhou 510515 Guangdong, China and [2]Department of Neurosurgery, Affiliated Hospital of Southwest Medical University, Luzhou 646000 Sichuan, China
Correspondence: Songtao Qi (sjwkqisongtao@126.com)
These authors contributed equally: Junxiang Peng, Lang Yang, Jun Pan, Chaohu Wang

Received: 20 March 2020 Revised: 19 May 2020 Accepted: 23 May 2020
Published online: 19 June 2020

determined by preoperative computed tomography and/or surgical inspection and classified as follows: no calcification, dotted or eggshell calcification, and massive calcification (≥10 mm in diameter).

Endocrinological assessment

Morning fasting venous blood was obtained from all the study participants 3 days before surgery to detect the levels of plasma thyrotropin (TSH), free thyroxine (FT4, FT3), thyroid function, cortisol (Cor), total testosterone, follicle-stimulating hormone (FSH), prolactin, luteinizing hormone (LH), insulin-like growth factor-1 (IGF-1), and growth hormone (GH). Informed consent was obtained from the patients' parents or guardians, and insulin provocative tests were performed before surgery.

Hypopituitarism was classified as partial or complete insufficiency of hormone secretion from the anterior pituitary.[5] Hypothalamus pituitary adrenal axis deficiency (HPAD) was diagnosed when the peak Cor value was <18 ng/ml after provocative testing (insulin tolerance test (ITT)) or the baseline plasma Cor was <3 ng/ml.[6] A low serum level of IGF-1 and a GH peak < 5.6 μg/L in the ITT was used to diagnose growth hormone deficiency (GHD).[7] The development of secondary sex characteristics was assessed in all the adolescent patients (females > 12 years, males > 14 years). Patients with delayed or absent puberty and a low serum testosterone level associated with inappropriately low or normal LH and FSH levels were diagnosed with hypothalamic pituitary gonadal axis deficiency (HPGD).[5,6] Delayed or absent development of puberty was defined as no breast development by 13 years of age in females and no testicular enlargement by 14 years of age in males. Hypothalamic pituitary thyroid axis deficiency (HPTD) was diagnosed in patients with both low or inadequate levels of TSH and low levels of free T4 (FT4).[6,8] A diagnosis of diabetes insipidus (DI) was established by measuring urine volume (>3 L/24 h) and urine osmolality (<300 mOsm/kg).[5] Body mass index (BMI) is expressed as standard deviation score as described previously.[9]

Immunohistochemical (IHC) staining

Briefly, all slides were dewaxed, dehydrated, and rehydrated. Primary antibodies against interleukin (IL)-6 (ab6672, abcam, USA), IL-8 (ab7747, abcam, USA), and monocyte chemoattractant protein-1 (MCP-1; ab25124, abcam, USA) were added to the sections, incubated overnight at 4 °C, and detected using secondary antibody kits (PV-9000, ZSCB-BIO, China).

Assessment of the inflammatory response

Inflammatory cytokines were measured using a cytokine antibody array, and the results were verified by reverse transcriptase polymerase chain reaction (RT-PCR). The specimens were immediately fixed in 10% formalin for 24 h and then embedded in paraffin using previously described methods.[10–12] The samples were then sectioned and stained with hematoxylin and eosin (ZSGB-Bio, Beijing, China). Two neuropathologists from the Southern Hospital Department of Pathology assessed the samples in a single blinded manner. To measure the number of inflammatory cells in the adenohypophysis junction, we randomly selected five fields of vision (high-power-field magnification, ×400) and calculated the average number of inflammatory cells in each field. Based on these calculations, the specimens were divided into three groups: mild group (<15 cells/field), moderate group (15–50 cells/field), and severe group (>50 cells/field) (Fig. 1).

Protein preparation and cytokine array

Protein extracts from CP patient specimens were analyzed by semiquantitative western spot blot analysis using the Human Inflammation 40 Factor Antibody Chip Kit III (AAH INF-G3) following the manufacturer's instructions. The chips were incubated for 30 min at room temperature, after which the samples were added to the chips and incubated overnight at 4 °C. After washing, diluted biotin-labeled antibody was added. After washing again, fluor streptavidin was added and incubated in the dark at room temperature. An Axon GenePix laser scanner was used to measure the signals, and Cy3 or green channel application chip analysis software was used to extract the data. The AAH-INF-G3 data analysis software was used for data analysis. The normalized gray value of each point on the chip film was compared with that of the control group, and then a statistical chart was created.

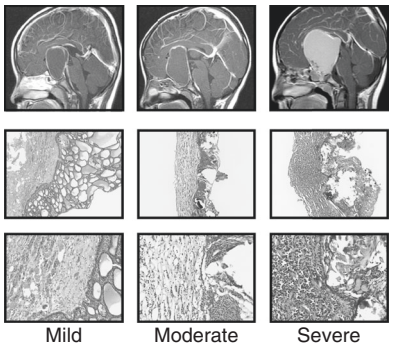

Fig. 1 Preoperative and pathological findings in pediatric infradiaphragmatic craniopharyngiomas. Preoperative MRI findings in infradiaphragmatic craniopharyngioma patients (first line). Second line (HE × 100) and third line (HE × 400): inflammation was measured in craniopharyngioma tissues using immunohistochemistry. Mild group < 15 cells/field; moderate group 15–50 cells/field; severe group > 50 cells/field.

Table 1. Gene-specific primer sequences for RT-qPCR.

Gene	Primer	Sequence (5′→3′)
IL-1A	Forward	TGGTAGTAGCAACCAACGGGA
	Reverse	ACTTTGATTGAGGGCGTCATTC
IL-1B	Forward	ATGATGGCTTATTACAGTGGCAA
	Reverse	GTCGGAGATTCGTAGCTGGA
IL-6	Forward	ACTCACCTCTTCAGAACGAATTG
	Reverse	CCATCTTTGGAAGGTTCAGGTTG
IL-8	Forward	TTTTGCCAAGGAGTGCTAAAGA
	Reverse	AACCCTCTGCACCCAGTTTTC
MCP-1	Forward	CAGCCAGATGCAATCAATGCC
	Reverse	TGGAATCCTGAACCCACTTCT
GAPDH	Forward	ACAACTTTGGTATCGTGGAAGG
	Reverse	GCCATCACGCCACAGTTTC

RT-quantitative PCR (RT-qPCR) analysis

TRIzol total RNA reagent (TaKaRa, Japan) was used to extract total RNA from tumor tissue. cDNA was obtained using the PrimeScript RT Master Mix Reagent Kit (TaKaRa, Japan), and RT-qPCR was performed using a Light Cycler 480 Real-time PCR System (Roche). Glyceraldehyde 3-phosphate dehydrogenase was used as an endogenous control. The $2^{-\Delta\Delta Ct}$ method was used to determine the relative expression levels. The gene-specific primers used for amplification are listed in Table 1. To aid in comparison, the change in expression in the mild group is set to 1.

Follow-up

Clinical follow-up information was obtained by reviewing the records of each patient's regular follow-up office visit. The median follow-up period for our patient cohort was 197.18 ± 56.65 months (range 3–216 months). When necessary, patients with hypopituitarism received hormone replacement therapy. Recurrence and progression of the tumor were defined as clinical and/or imaging

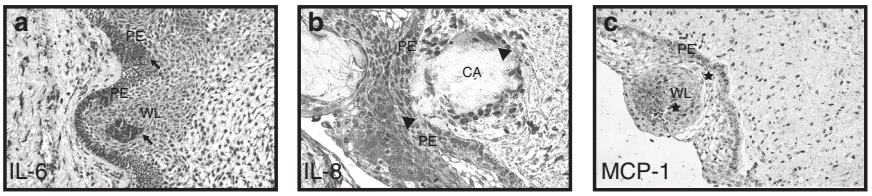

Fig. 2 Inflammation in pediatric infradiaphragmatic craniopharyngiomas. a IL-6 (arrows) expression in whorl-like cells and palisade epithelial cells; **b** IL-8 (triangle) expression in tumor palisade epithelial cells and around calcification; **c** MCP-1 (star) was expressed around whorl-like cells and palisade epithelial cells. PE palisade epithelial cells, CA calcification, WL whorl-like cells.

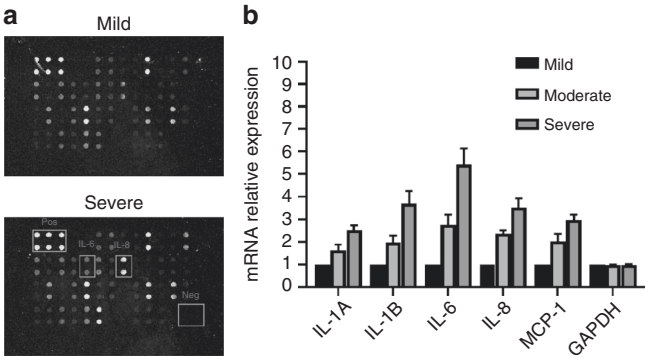

Fig. 3 Inflammatory cytokine expression at the protein and transcriptional levels in pediatric infradiaphragmatic craniopharyngiomas. a IL-6 and IL-8 were the most strongly expressed in the severe group compared to the mild group. **b** The transcription levels of IL-1A, IL-1B, IL-6, IL-8, and MCP-1 in the severe group were significantly higher than those in the moderate and mild groups.

progression upon follow-up MRI. The overall survival (OS) and progression-free survival (PFS) rates were calculated and statistically analyzed.

Statistical analysis

Statistical analysis was performed using SPSS 19.0 (SPSS Inc., Chicago, IL, USA). Continuous data are presented as mean ± standard deviation. Nonparametric variables between the three groups were compared using Wilcoxon rank-sum test (Mann–Whitney *U* test). Pearson correlation analysis was used to measure correlations between the variables. Survival curves were generated using the Kaplan–Meier method, and log-rank test was used to measure the differences between the curves. $P < 0.05$ was considered statistically significant.

RESULTS

Levels of inflammation in pediatric Q-CPs

Inflammation was present in 77.6% of the Q-CP patients in our cohort. The mild group contained 46 (45.1%) patients, the moderate group contained 31 (30.4%) patients, and the severe group had 25 (24.5%) patients. We used IHC to detect the localization and expression of IL-6, IL-8, MCP-1, and other inflammatory cytokines in the CPs. We found that the majority of the inflammatory cytokines were expressed in the palisade epithelial cells (Fig. 2). Next, we grouped the patients by counting inflammatory cells in the tumor (Fig. 1). The rationalization of the grouping was verified by cytokine chip and PCR. Figure 3a shows a representative array of tumor tissues in the mild and severe groups. Among the cytokines, IL-6 and IL-8 were most strongly expressed in the severe group. The transcription levels of IL-1A, IL-1B, IL-6, IL-8, and MCP-1 in the different inflammatory groups were detected by fluorescence qPCR, and we found that the transcription level of IL-6 was most highly upregulated in the severe group (Fig. 3b).

Table 2. Clinical and endocrine characteristics of infradiaphragmatic craniopharyngioma according to inflammatory response.

Variables	Inflammatory density score			*P*
	Mild ($n = 46$)	Moderate ($n = 31$)	Severe ($n = 25$)	
Age, years	10.28 ± 4.33	11.71 ± 4.53	9.00 ± 3.34	0.057
Sex (male %)	26 (56.5%)	17 (54.8%)	13 (52.0%)	0.935
Pathological type (ACP)	43 (93.5%)	30 (96.8%)	25 (100%)	0.390
Calcification				
None calcification	13 (28.3%)	3 (9.7%)	3 (12.0%)	0.000
Dotted or eggshell calcification	31 (67.4%)	21 (67.7%)	6 (24.0%)	
Massive calcification	2 (4.3%)	7 (22.6%)	16 (64.0%)	
Tumor size (cm)				
<2	4 (8.7%)	2 (6.5%)	2 (8.0%)	0.647
2–4	28 (66.9%)	19 (61.3%)	12 (48.0%)	
4–6	11 (23.9%)	8 (25.8%)	6 (24.0%)	
>6	3 (6.5%)	2 (6.5%)	5 (20.0%)	
HPAD preoperative	16 (34.8%)	18 (58.1%)	20 (80.0%)	0.001
HPTD preoperative	11 (23.9%)	17 (54.8%)	18 (72.0%)	0.000
HPGD preoperative	10 (21.7%)	13 (41.9%)	15 (60.0%)	0.005
GHD preoperative	28 (60.9%)	24 (77.4%)	23 (82.0%)	0.015
DI preoperative	1 (2.2%)	3 (9.7%)	4 (16.0%)	0.106
GH peak (μg/ml)	4.94 ± 1.89	4.08 ± 1.97	1.85 ± 1.68	0.000
Cor peak (μg/dl)	18.52 ± 5.23	14.25 ± 5.39	8.44 ± 5.61	0.000
IGF-1 preoperative (ng/ml)	64.94 ± 31.90	61.84 ± 32.80	36.12 ± 12.78	0.000

HPAD hypothalamus pituitary adrenal axis deficiency, *HPTD* hypothalamic pituitary thyroid axis deficiency, *HPGD* hypothalamic pituitary gonadal axis deficiency, *GHD* growth hormone deficiency, *DI* diabetes insipidus.

Clinical characteristics and the inflammatory response

Infradiaphragmatic tumors occurred predominantly in pediatric patients. Ninety-eight patient tumors were classified as adamantinomatous craniopharingiomas (ACPs), while 4 patients had typical squamous tumors. The majority of the Q-CPs had cysts. Calcification was observed in 80% of the patients. However, hydrocephalus rarely occurred. In all, 40% of patients had visual impairment, and a few had symptoms associated with elevated intracranial pressure and nonspecific headache. All 102 patients in this cohort underwent surgical resection with the aim of total tumor removal. The pterional approach was used in 10 cases, the anterior interhemispheric approach was used in 42 cases, and the transsphenoidal approach was used in 50 cases. Associations between clinical features and the inflammatory response are presented in Table 2. We found that there was a statistically

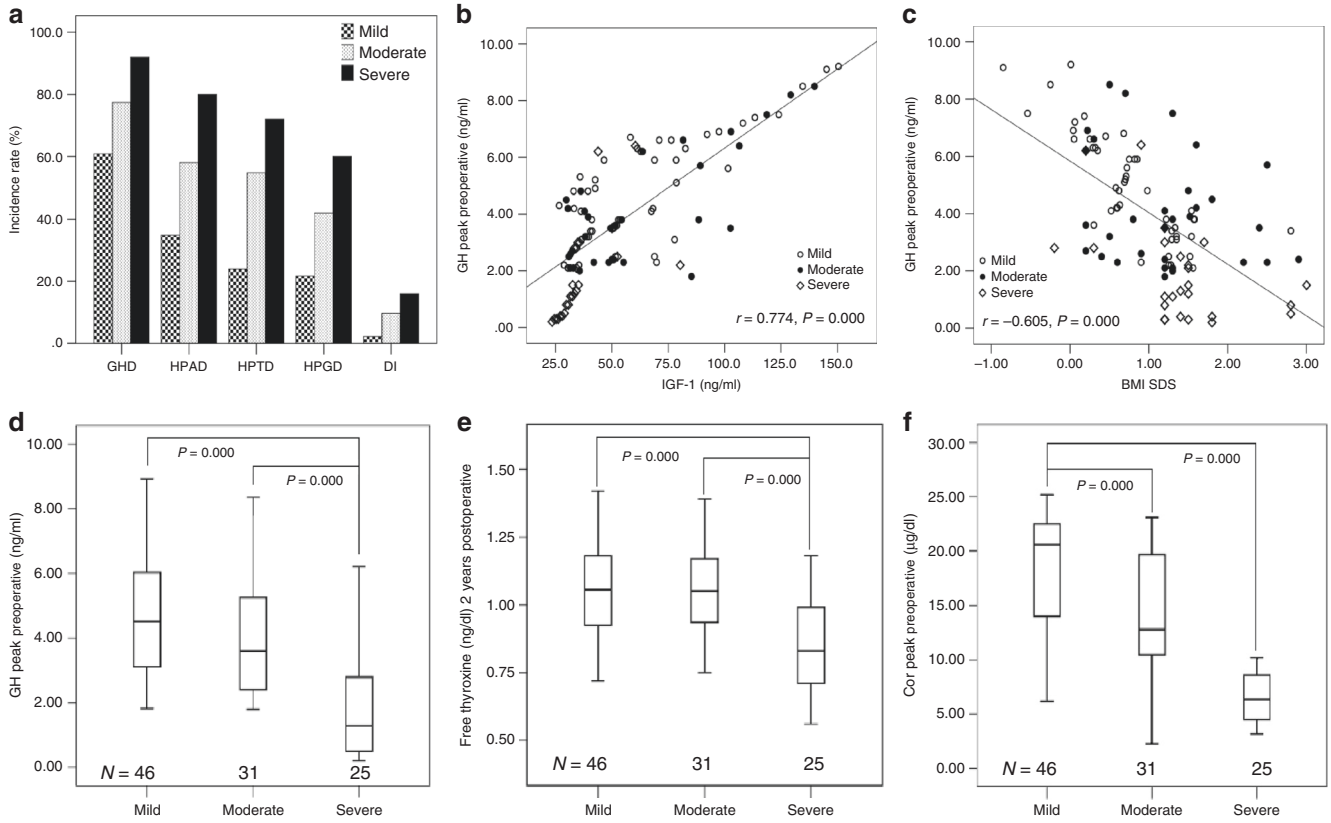

Fig. 4 Relationship between pituitary dysfunction and tumor inflammatory response in pediatric infradiaphragmatic craniopharyngiomas. **a** Frequency characteristics of pituitary dysfunction in pediatric infradiaphragmatic craniopharyngiomas according to the tumor inflammatory response. **b**, **c** Peak GH was highly correlated with serum IGF-I and BMI. GH peak (**d**), FT4 (**e**), and Cor peak (**f**) pre-surgery in patients with infradiaphragmatic craniopharyngioma according to the tumor inflammatory response. The horizontal line in the middle of the box represents the median. The top and bottom edges of the box mark the 25th and 75th percentiles, respectively. Whiskers indicate the range of values that fall within 1.5 box lengths.

significant difference between the inflammation density score and the degree of calcification ($P = 0.000$) and hypopituitarism ($P < 0.05$).

Hypopituitarism pattern and the inflammatory response

Eighty-nine patients (87.3%) in the cohort had hypopituitarism. Hypopituitarism was especially prevalent in juvenile patients, and 15.7% (16/102) of children had panhypopituitarism when admitted to the hospital. The incidence of hypopituitarism is shown in Fig. 4a. Hypopituitarism was most common in the severe group, and GHD was the most common deficiency (severe group 92.0% vs. moderate group 77.4% vs. mild group 60.9% ($P < 0.0001$)). GHD was the only deficiency in 13.3% of the patients, occurred with 1 other deficiency in 40% of patients, and occurred with 2–3 other deficiencies in 46.7% of patients. HPTD was the single deficiency in 1.7% of patients, occurred with 1 other deficiency in 32.6% of patients, and occurred with 2–3 other deficiencies in 65.2% of patients. In all, 4.6% of patients had HPAD as a single deficiency, 20.3% had HPAD with 1 other deficiency, and 75.1% had HPAD with 2–3 other deficiencies. HPGD occurred as a single deficiency in 2.6% of patients, occurred with 1 other deficiency in 26.3% of patients, and occurred with 2–3 other deficiencies in 71.1% of patients. Low IGF-1 occurred in 7.5% of patients as an isolated deficiency, occurred with 1 or 2 other deficiencies in 37.3% of patients each, and occurred with 3 other deficiencies in 55.2% of patients. Multivariable logistic regression analysis showed that the risk of hypopituitarism was significantly increased in subjects with severe inflammation ($P < 0.05$). We next used one-way analysis of variance to find the relationship between hypopituitarism and different inflammatory responses, tumor, size, texture, and calcification. The results showed that the extent of the endocrine disorder correlates with the inflammatory response ($P < 0.05$). Further application of the least significant difference method showed that the endocrine function disorders in the severe group were more serious than those of the other groups.

In this study, a total of 102 patients underwent ITTs. The peak GH response to ITT was <5.6 μg/ml in 75 subjects (73.5%) and >5.6 μg/ml (range 5.6–9.2 μg/ml) in 27 (26.5%) patients. The mean peak GH and Cor responses to ITT in the severe group were 1.85 ± 1.68 ng/ml and 8.44 ± 5.61 μg/dl, respectively. The mean peak GH and Cor responses were 4.94 ± 1.89 ng/ml and 18.52 ± 5.23 μg/dl, respectively, in the mild group. The GH peak and Cor peak were different, with the lowest peak in the severe group and the highest in the mild group ($P = 0.000$; Fig. 4d, f). For the GH peak response to ITT, there was a significant inverse relationship between BMI and peak GH ($r = -0.605$, $P = 0.000$). There was also a significant relationship between IGF-1 levels and peak GH ($r = 0.774$, $P = 0.000$; Fig. 4b, c). These results suggest that severe inflammation negatively affects the endocrine system and increases the incidence of endocrine disorders ($P < 0.05$).

Prognosis and inflammatory response

The OS rate of the cohort was 96.1% at 2 years, 93.1% at 5 years, and 89.2% at 10 years, and the PFS rate was 93.1% at 2 years, 89.1% at 5 years, and 85.1% at 10 years. Detailed survival curves are shown in Fig. 5. The criteria detailed by Fahlbusch[13] were used to assess the functional outcomes of the patients. Normal function

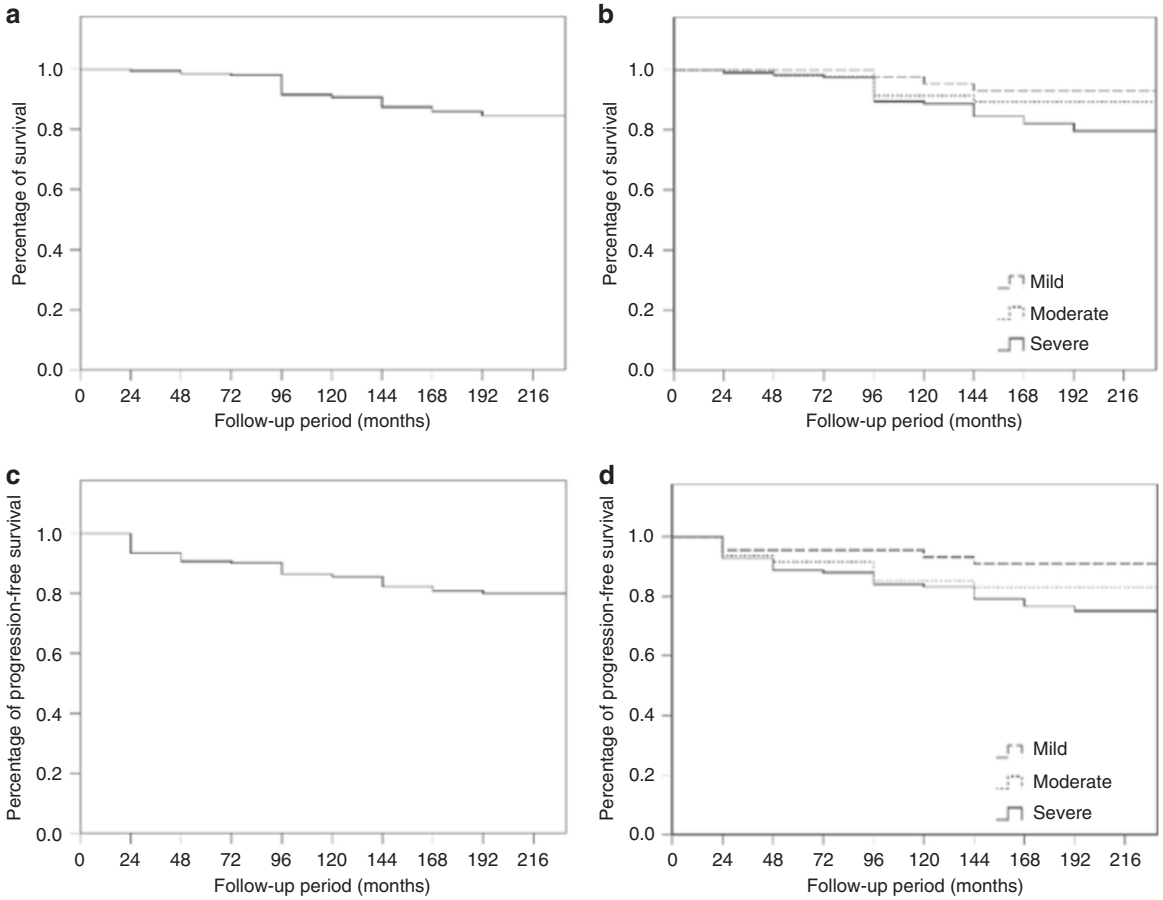

Fig. 5 Overall and progression-free survival in pediatric patients with infradiaphragmatic craniopharyngioma. a The overall survival rate was 96.1% at 2 years, 93.1% at 5 years, and 89.2% at 10 years. **b** The long-term survival rate was higher in the mild inflammation group than the other groups. **c** The progress-free survival rate was 93.1% at 2 years, 89.1% at 5 years, and 85.1% at 10 years. **d** There was significant difference in progress-free survival between groups; Patients in the severe group had a significantly lower probability of recurrence-free survival than those with other types of tumors.

was completely restored in 26 postoperative patients, and 2 patients got married and had children. Seventy patients (82.4%) will obtain a good quality of life while undergoing hormone replacement therapy. The long-term survival rate was higher in the mild inflammation group than the other groups. The outcome was excellent (good recovery and only mild disability) in 76% of severe group patients, 92% of mild group patients, and 82% of moderate group patients. The worsened functional status in the severe group was mostly attributable to panhypopituitarism and visual deficits. Patients in the severe group had significantly lower recurrence-free survival and significantly worsened functional status compared to the mild group ($P = 0.001$).

DISCUSSION

Although CPs are benign, they are aggressive tumors. They occur along the craniopharyngeal duct and are most often located in the sellar or suprasellar regions.[1–4] Because the cranial base is complex in structure and close to several critical structures, hypopituitarism is common in pediatric Q-CPs. Recent studies have shown that inflammation correlates with CP, particularly in ACPs.[14] Inflammatory cytokines, such as IL-1A, IL-1B, IL-6, IL-8, and MCP-1, have been shown to induce progression in CP cells. Several efforts have been made to study ACPs at the genomic, transcriptomic, and proteomic levels in order to identify potential therapeutic targets.[15–18]

Origination and pathogenesis of pediatric Q-CPs

Q-CPs arise from Rathke's pouch cells and are mainly located under the diaphragma. This type of tumor is of major significance as it involves pituitary gland, which causes hypopituitarism. It is well known that ACP pathogenesis is mainly driven by mutations in the gene CTNNB1 that encodes β-catenin, the major regulator of the WNT pathway.[19] In addition, tumor cells promote the production of inflammatory cytokines by immune cells, which may cause tumor proliferation, invasion, and angiogenesis.[20] It has been shown that the tumor microenvironment, which is largely regulated by inflammatory cells, is a key player in promoting diffusion, survival, and migration of tumors.[21,22]

Clinical features and prognosis of pediatric Q-CPs

Most Q-CPs in this study were ACPs and the majority were cystic, with more than half of the tumors being almost entirely cystic. Hydrocephalus was rare, and hypopituitarism is more serious than supersellar tumors. The following clinical manifestations were seen: (1) impairment of the optical path structure; (2) impairment of endocrine function, including hypopituitarism, DI, and electrolyte imbalances; and (3) high intracranial pressure triggered by the tumor and hydrocephalus (non-communicative). Long-term postoperative treatment focused on hormone replacement, and severe obesity was rare.

In this cohort, most pediatric Q-CP patients (89.7%) presented with pituitary dysfunction, which manifested as HPTD, GHD, HPAD,

HPGD, and/or DI. However, the etiology of hypopituitarism related to the CP is still uncertain.[23,24] We found that even patients with tumors < 2 cm in size had some degree of hypopituitarism, suggesting that tumor size is not a major contributor to hypopituitarism pathogenesis. Inflammation in CP was associated with hypopituitarism and calcification, and the expression of many inflammatory cytokines was higher in the severe group than in the mild group. It has been postulated that inflammation has an important role in the pathogenesis of hypopituitarism. A possible mechanism may be as follows: First, inflammatory cytokines play a crucial role in tumor progression, including each of the distinct stages—initiation, promotion, progression, and metastasis. The results of this study showed that many inflammatory cytokines were more highly expressed in the severe group than in the mild group. In particular, MCP-1, IL-1, and IL-1R were found to be highly expressed in the severe group. These inflammatory cytokines can promote cell adhesion, tumor cell invasion, and migration.[25] In addition, activation of autophagy in tumor cells has been found to promote hypopituitarism.[26] Inflammatory cytokines can also induce the chemotaxis of microglia, promote the formation of the glial proliferation zone, and then induce the migration and infiltration of microglia. Microglia play a regulatory role by maintaining the stemness of the tumor. However, tumor stem cells can alter the phenotype of the microglia, promoting the development of hypopituitarism.[27–30]

Implications of the inflammatory response in Q-CP treatment

Inflammatory response is common in Q-CP and is closely related to the biological behavior and clinical prognosis of the disease. There is still controversy surrounding the best treatment for CP, especially for children. Current treatment paradigms, including surgical resection and radiotherapy, have increased morbidity in patients. Therefore, there is a great need for safer and more effective treatments for this disease. In recent years, great efforts have been made to fully detail the genome, transcriptome, and proteome of these tumors to identify potential therapeutic targets. The classical Wnt pathway plays an important role in the inflammatory response in ACP, but there are still no effective drugs that target this pathway. Several lines of evidence have shown that the activation of inflammation and the immune response plays a critical role in ACP progress. The use of inflammatory cytokine inhibitors and immunotherapy may aid in controlling tumor progression, reducing surgical difficulty, and improving long-term endocrine dysfunction.

CONCLUSION

Q-CPs are histologically benign brain tumors that often follow an aggressive clinical course. Given the relatively young age of patients with Q-CP and the need to improve prognosis, understanding the etiology of hypopituitarism has become increasingly important. Therefore, it is of great significance to elucidate the molecular mechanism by which the inflammatory response impacts CP pathogenesis. Analysis of these mechanisms has the potential to identify immunological therapeutic targets, which may help to reduce the difficulty of CP surgery and improve prognosis.

ACKNOWLEDGEMENTS

The authors thank Xu Hai-yan, RN and Qiu Xiao-yu for the collection of data and care of patients enrolled in this study. This research received grant from four fund projects in the public, including Science and Technology Program of Guangdong (No. 2017A020215048), the Natural Science Foundation of Guangdong Province (No. 2019A1515012140), the National Natural Science Foundation of China (No. 81972352), and Clinical Research Startup Program of Southern Medical University by High-level University Construction Funding of Guangdong Provincial Department of Education (LC2016YM001).

AUTHOR CONTRIBUTIONS

J.P., L.Y., and J.P. performed primary data analysis and wrote the manuscript. L.Y., Y.L., C.W., J.F., J.N., and J.Z. performed H&E, RT-qPCR analysis, protein preparation, and cytokine array. S.Q. and J.P. designed and supervised the study. All authors edited and approved the final manuscript.

ADDITIONAL INFORMATION

Competing interests: We declare that we have no financial and personal relationships with other people or organizations that can inappropriately influence our work, the patient consent was not required, and there is no professional or other personal interest of any nature or kind in any product, service and/or company that could be construed as influencing the position presented in, or the review of, the manuscript.

Publisher's note Springer Nature remains neutral with regard to jurisdictional claims in published maps and institutional affiliations.

REFERENCES

1. Li, D. et al. Risk score for the prediction of severe obesity in pediatric craniopharyngiomas: relative to tumor origin. *Pediatr. Res.* **83**, 645–654 (2018).
2. Sterkenburg, A. S. et al. Nuchal skinfold thickness: a novel parameter for assessment of body composition in childhood craniopharyngioma. *J. Clin. Endocrinol. Metab.* **101**, 4922–4930 (2016).
3. Pan, J. et al. Growth patterns of craniopharyngiomas: clinical analysis of 226 patients. *J. Neurosurg. Pediatr.* **17**, 418–433 (2016).
4. Apps, J. R. et al. Molecular pathology of adamantinomatous craniopharyngioma: review and opportunities for practice. *Neurosurg. Focus* **41**, E4 (2016).
5. Grimberg, A. et al. Guidelines for growth hormone and insulin-like growth factor-I treatment in children and adolescents: growth hormone deficiency, idiopathic short stature, and primary insulin-like growth factor-I deficiency. *Horm. Res. Paediatr.* **86**, 361–397 (2016).
6. Oto, Y. et al. Delayed peak response of cortisol to insulin tolerance test in patients with Prader-Willi syndrome. *Am. J. Med. Genet. A* **176**, 1369–1374 (2018).
7. Malina, R. M. & Beunen, G. P. Assessment of skeletal maturity and prediction of adult height (TW3 method). *Am. J. Hum. Biol.* **14**, 788–789 (2002).
8. Koulouri, O. et al. Diagnosis and treatment of hypothyroidism in TSH deficiency compared to primary thyroid disease: pituitary patients are at risk of under-replacement with levothyroxine. *Clin. Endocrinol (Oxf).* **74**, 744–749 (2011).
9. Zong, X. N. & Li, H. Construction of a new growth references for China based on urban Chinese children: comparison with the WHO growth standards. *PloS one* **8**, e59569 (2013).
10. Zhou, J., Zhang, C., Pan, J., Chen, L. & Qi, S.-T. Interleukin-6 induces an epithelial-mesenchymal transition phenotype in human adamantinomatous craniopharyngioma cells and promotes tumor cell migration. *Mol. Med. Rep.* **15**, 4123–4131 (2017).
11. Yi, L. et al. Pathological relationship between adamantinomatous craniopharyngioma and adjacent structures based on QST classification. *J. Neuropathol. Exp. Neurol.* **77**, 1017–1023 (2018).
12. Chen, M., Zheng, S. H., Liu, Y., Shi, J. & Qi, S. T. Periostin activates pathways involved in epithelial-mesenchymal transition in adamantinomatous craniopharyngioma. *J. Neurol. Sci.* **360**, 49–54 (2016).
13. Fahlbusch, R., Honegger, J., Paulus, W., Huk, W. & Buchfelder, M. Surgical treatment of craniopharyngiomas: experience with 168 patients. *J. Neurosurg.* **90**, 237–250 (1999).
14. Martinez-Barbera, J. P. Molecular and cellular pathogenesis of adamantinomatous craniopharyngioma. *Neuropathol. Appl. Neurobiol.* **41**, 721–732 (2015).
15. Whelan, R., Prince, E., Gilani, A. & Hankinson, T. The inflammatory milieu of adamantinomatous craniopharyngioma and its implications for treatment. *J. Clin. Med.* **9**, 519 (2020).
16. Apps, J. R. et al. Tumour compartment transcriptomics demonstrates the activation of inflammatory and odontogenic programmes in human adamantinomatous craniopharyngioma and identifies the MAPK/ERK pathway as a novel therapeutic target. *Acta Neuropathol.* **135**, 757–777 (2018).
17. Donson, A. M. et al. Molecular analyses reveal inflammatory mediators in the solid component and cyst fluid of human adamantinomatous craniopharyngioma. *J. Neuropathol. Exp. Neurol.* **76**, 779–788 (2017).
18. Coy, S. et al. Multiplexed immunofluorescence reveals potential PD-1/PD-L1 pathway vulnerabilities in craniopharyngioma. *Neuro Oncol.* **20**, 1101–1112 (2018).
19. Xekouki, P. et al. Non-secreting pituitary tumours characterised by enhanced expression of YAP/TAZ. *Endocr. Relat. Cancer* **26**, 215–225 (2019).

20. Dayyani, F., Gallick, G. E., Logothetis, C. J. & Corn, P. G. Novel therapies for metastatic castrate-resistant prostate cancer. *J. Natl Cancer Inst.* **103**, 1665–1675 (2011).
21. Wang, T. et al. Overexpression of the human ZNF300 gene enhances growth and metastasis of cancer cells through activating NF-kB pathway. *J. Cell. Mol. Med.* **16**, 1134–1145 (2012).
22. Pettorini, B. L. et al. The role of inflammation in the genesis of the cystic component of craniopharyngiomas. *Childs Nerv. Syst.* **26**, 1779–1784 (2010).
23. Daubenbuchel, A. M. & Muller, H. L. Neuroendocrine disorders in pediatric craniopharyngioma patients. *J. Clin. Med.* **4**, 389–413 (2015).
24. Muller, H. L., Merchant, T. E., Warmuth-Metz, M., Martinez-Barbera, J. P. & Puget, S. Craniopharyngioma. *Nat. Rev. Dis. Prim.* **5**, 75 (2019).
25. Huang, F., Wang, B. R. & Wang, Y. G. Role of autophagy in tumorigenesis, metastasis, targeted therapy and drug resistance of hepatocellular carcinoma. *World J. Gastroenterol.* **24**, 4643–4651 (2018).
26. Wang, N. & Feng, Y. Elaborating the role of natural products-induced autophagy in cancer treatment: achievements and artifacts in the state of the art. *Biomed. Res. Int.* **2015**, 934207 (2015).
27. Goschzik, T. et al. Genomic alterations of adamantinomatous and papillary craniopharyngioma. *J. Neuropathol. Exp. Neurol.* **76**, 126–134 (2017).
28. Martinez-Barbera, J. P. & Andoniadou, C. L. Concise review: paracrine role of stem cells in pituitary tumors: a focus on adamantinomatous craniopharyngioma. *Stem Cells* **34**, 268–276 (2016).
29. Krishnamurthy, N. & Kurzrock, R. Targeting the Wnt/beta-catenin pathway in cancer: Update on effectors and inhibitors. *Cancer Treat. Rev.* **62**, 50–60 (2018).
30. Martelli, C. et al. Investigating the protein signature of adamantinomatous craniopharyngioma pediatric brain tumor tissue: towards the comprehension of its aggressive behavior. *Dis. Markers* **2019**, 3609789 (2019).