Characteristics, treatments, and outcomes of adolescents and adults with neuroblastoma: a retrospective study in China

Weiji Xie*, Yu Zhang*, Jiaqian Xu*, Feifei Sun, Jia Zhu, Yi Que, Junting Huang, Zijun Zhen, Suying Lu, Juan Wang and Yizhuo Zhang (iD)

Abstract

Background: Neuroblastoma (NB) is rare in adolescents and adults, resulting in limited availability of data.

Objectives: We comprehensively investigated the characteristics, treatments, and outcomes of adolescent and adult patients with NB, aiming to provide a more in-depth insight into this disease.

Design: A retrospective, single-center study.

Methods: We retrieved and analyzed the medical data of patients with NB aged 10 years or older at diagnosis who were treated at Sun Yat-sen University Cancer Center between June 2005 and January 2024.

Results: Sixty-five patients (30 males and 35 females) were enrolled, with a median age of 20 years (interquartile range, 14–26 years), including 27 patients aged 10–18 years and 38 patients aged >18 years. Most patients were classified as M-stage disease ($n = 40$, 61.5%), high-risk ($n = 42$, 64.6%), and poorly differentiated NB ($n = 27$, 41.5%). Additionally, 3 (6.7%) patients had *MYCN* amplification, and 5 (25%) had *ALK* mutations. The genomic landscape revealed that mutations in the cell cycle and DNA repair pathways are related to chemotherapy sensitivity. After induction therapy, 34 (52.3%) patients achieved complete response (CR). The 5-year progression-free survival (PFS) and overall survival (OS) rates were $33.1\% \pm 6.9\%$ and $55.1\% \pm 7.6\%$, respectively. Patients who achieved CR after induction therapy had superior PFS ($p = 0.009$), with 5-year PFS rates of $44.0\% \pm 10.6\%$ compared to $18.5\% \pm 8.5\%$ in non-CR patients.

Conclusion: Adolescent and adult patients with NB exhibit distinct characteristics, less chemotherapy sensitivity, and poorer outcomes compared to pediatric patients. Achieving CR after induction therapy is associated with better outcomes. Further investigation for new therapies is required.

Keywords: adolescent, adult, neuroblastoma, outcome, treatment

Correspondence to:
Suying Lu
Juan Wang
Yizhuo Zhang
State Key Laboratory of Oncology in South China, Guangdong Provincial Clinical Research Center for Cancer, Sun Yat-sen University Cancer Center, Guangzhou 510060, P.R. China

Department of Pediatric Oncology, Sun Yat-sen University Cancer Center, 651 Dongfeng East Road, Yuexiu District, Guangzhou City, Guangdong 510060, P.R. China

lusy@sysucc.org.cn
wangjuan@sysucc.org.cn
zhangyzh@sysucc.org.cn

Weiji Xie
Jiaqian Xu
Feifei Sun
Jia Zhu
Yi Que
Junting Huang
Zijun Zhen
State Key Laboratory of Oncology in South China, Guangdong Provincial Clinical Research Center for Cancer, Sun Yat-sen University Cancer Center, Guangzhou, P.R. China

Department of Pediatric Oncology, Sun Yat-sen University Cancer Center, Guangzhou, P.R. China

Yu Zhang
State Key Laboratory of Oncology in South China, Guangdong Provincial Clinical Research Center for Cancer, Sun Yat-sen University Cancer Center, Guangzhou, P.R. China

Department of Pathology, Sun Yat-sen University Cancer Center, Guangzhou, P.R. China

*These authors contributed equally.

Introduction

Neuroblastoma (NB) is the most common extracranial solid tumor in early childhood, constituting approximately 10% of all malignant tumors in children.[1] Typically, the median age at diagnosis is 18 months, with more than 95% of tumors occurring before 10 years of age.[2] Age at diagnosis is a prognostic factor for NB.[3] Older age is a prognostic factor of poor survival, with outcomes gradually worsening with increasing age at diagnosis.[4] Newborn patients aged <18 months generally have better outcomes, with some experiencing

spontaneous regression of tumors, whereas adolescents and adults tend to have an unfavorable prognosis.[5]

Adolescents and adults rarely develop NB, resulting in limited clinical and biological data in the literature. A limited series of reports have revealed distinct characteristics and biological features of NB in adolescent and adult populations: more indolent behavior, a longer disease course, predominance of advanced stages, unusual pattern of metastatic localizations, lower urine catecholamine levels, higher incidence of unfavorable histology, rare *MYCN* amplification, less sensitivity or poor tolerance to pediatric chemotherapy regimens, and generally worse outcomes.[6–17] The genetic landscape of adolescent and adult NB patients remains poorly characterized. Limited results from targeted exome sequencing found a high frequency of loss-of-function mutations in *ATRX* and *ALK*, as well as less common mutations in genes such as *BRCA1*, *MUTYH*, *ERBB3*, *MAGI2*, *RUNX1*, *MLL*.[17,18] Currently, standard NB treatments are not specifically adapted to the unique pathophysiology of adolescent and adult patients. Given the poor outcomes and limited understanding of optimal treatment strategies for this rare population, comprehensive clinical and biological data are urgently needed to guide future research and improve therapeutic approaches.

Herein, we retrospectively retrieved and analyzed the medical data of patients with NB aged ⩾10 years at the time of diagnosis to provide a more in-depth insight into the clinical characteristics, treatment modalities, and prognostic data of this rare disease in adolescents and adults.

Materials and methods

Patients
We reviewed all medical records of patients with NB aged ⩾10 years treated at Sun Yat-sen University Cancer Center (SYSUCC) between June 2005 and January 2024.

This study was approved by the Institutional Review Board and Ethics Committee of the SYSUCC (B2024-504-01) and was conducted in accordance with the Code of Ethics of the World Medical Association (Declaration of Helsinki) for experiments involving humans and Good Clinical Practice. The requirement for written informed consent was waived owing to the study's retrospective nature. The reporting of this study conforms to the STROBE (Strengthening the Reporting of Observational Studies in Epidemiology) statement[19] (Supplemental File 1).

Diagnosis and risk stratification
The diagnosis of NB was confirmed by histopathology or the demonstration of bone marrow involvement with extrinsic cells, accompanied by elevated urinary catecholamine metabolites. Workup at the initial diagnosis and subsequent response assessment were as follows: urinary homovanillic acid, vanillylmandelic acid, bilateral bone marrow aspiration and core biopsy, computed tomography (CT), magnetic resonance imaging, I^{123} metaiodobenzylguanidine (MIBG) scans, and fluorodeoxyglucose-positron emission tomography scans (PET/CT, if non-MIBG avid). The *MYCN* status was detected by fluorescence in situ hybridization (FISH) or by next-generation sequencing (NGS). Status by FISH is defined as follows: not amplified (2 copies per cell), gain (>2–8 copies per cell or <4-fold increase), amplified (>8 copies per cell or ⩾4-fold increase, can be >30-fold increase). The staging system and risk stratification were based on the International Neuroblastoma Risk Group (INRG) Staging System[20] and the INRG classification system.[21]

Genomic profiling and circulating tumor DNA surveillance
We performed comprehensive genomic profiling using whole exome sequencing (WES) on paired samples from 12 patients. Genomic DNA was extracted from the formalin-fixed, paraffin-embedded tumor samples and matched white blood cells. After DNA extraction, 1 ng DNA was subjected to polymerase chain reaction to amplify Housekeeping gene fragments using a set of primers (synthesized by Invitrogen, Life Technologies, Carlsbad, CA, USA). The amplified products were then fragmented into approximately 250 base pairs using sonication. Sample library preparation was performed using VAHTSTM Universal DNA Library Prep Kit for Illumina® V3 (Vazyme Biotech, Nanjing, China). Target enrichment was done by NGS-based hybrid capture method using the pool of customized individually synthesized plus xGen Exome Research Panel. Hybridization capture using target gene panel and xGen® Lockdown® Reagents followed the manufacturer's protocol, in order to enrich for the targeted nearly 20,000 genes. Post-capture

libraries were pooled together, denatured, and diluted to 200–250 pM for sequencing on Illumina NovaSeq 6000 sequencing platform (Illumina, San Diego, CA, USA). Various genomic alterations, including single nucleotide variants (SNVs), insertions and deletions, copy number variations, and gene rearrangements and fusions, were subsequently analyzed.

Longitudinal circulating tumor DNA (ctDNA) surveillance was conducted in six patients at multiple time points, including baseline, every 2 months during treatment (if available), at the end of treatment, and every 6 months post-treatment. Briefly, peripheral blood samples were collected and processed for cell-free DNA extraction, followed by DNA library preparation and high-throughput sequencing. Molecular tracking and variant calling for SNVs were subsequently performed. Samples with three or more detected variants were classified as ctDNA-positive.

Treatment protocol and response assessment
Treatment followed pediatric protocols, with dosage adjustments based on patient tolerance. For the very low-risk- and low-risk groups, the primary treatment was surgery and chemotherapy using the CAV (cyclophosphamide, pirarubicin, vincristine)/EP (etoposide, cisplatin) regimen. Intermediate-risk patients underwent surgery, chemotherapy (CAV/EP regimen), and radiotherapy. For high-risk patients, the treatment regimen included surgery, chemotherapy using the CAV (cyclophosphamide, pirarubicin, vincristine)/VIP (etoposide, ifosfamide, cisplatin) regimen, ±autologous stem cell transplantation (ASCT), or radiotherapy, followed by maintenance therapy, which included anti-GD2 immunotherapy, cis-retinoic acid, and 1-year metronomic chemotherapy (cyclophosphamide, vinorelbine, etoposide, celecoxib).

The overall response was assessed using the International Neuroblastoma Response Criteria: complete response (CR), partial response (PR), mixed response (MR), stable disease (SD), and progressive disease (PD).[22]

Statistical analysis
All statistical analyses were performed with SPSS version 25 (IBM Corp., Armonk, NY, USA). Progression-free survival (PFS; time to disease relapse or progression) and overall survival (OS;

time to death from any cause or the last follow-up) were assessed from the time of diagnosis. Survival analysis was performed using the Kaplan–Meier method and compared using the log-rank test. A Cox regression model was used to assess potential factors that could predispose patients to survival. Pearson's Chi-square test was used to compare clinical and biological characteristics between the adolescent and adult groups. $p < 0.05$ indicated statistical significance.

Results

Characteristics of the patients and clinical features at diagnosis
A total of 65 patients (30 males and 35 females), aged 10–68 years old at the time of diagnosis, were analyzed. The median age at diagnosis was 20 years (interquartile range (IQR), 14–26 years), with 27 patients aged 10–18 years and 38 patients aged >18 years. The baseline characteristics and clinical features are summarized in Table 1, and the detailed clinical information for each patient is listed in Supplemental Table S1.

The primary tumor sites were the adrenal gland ($n = 24$, 36.9%), retroperitoneum ($n = 24$, 36.9%), and mediastinum ($n = 10$, 15.4%). At initial diagnosis, 40 (61.5%) patients exhibited distant tumor metastasis, with the most common sites of metastasis being the lymph nodes ($n = 25$, 62.5%), bone ($n = 20$, 50%), and bone marrow ($n = 15$, 37.5%). Among the 65 patients, 22 (33.8%) were initially diagnosed with NB based on the discovery of a mass during routine physical examination or self-awareness. The sites of the primary tumor and metastases correlated with the most common presenting symptoms at diagnosis, including abdominal pain ($n = 16$, 24.6%) and backache ($n = 8$, 12.3%). Twelve (18.5%) patients presented with L1-stage, 12 (18.5%) patients with L2-stage, and 40 (61.5%) patients with M-stage of the disease. Most patients were classified into the high-risk group ($n = 42$, 64.6%), whereas others were classified into the very low-risk ($n = 6$, 9.2%), low-risk ($n = 11$, 16.9%), and intermediate-risk ($n = 5$, 7.7%) groups.

Biological features and genetic landscape
Regarding histological category, 45 (69.2%) patients were diagnosed with NB (including 12 differentiating NB, 27 poorly differentiating NB, 3 undifferentiated NB, and 3 unclassified), and 20 (30.8%) patients were diagnosed with

Table 1. Summary of patient characteristics.

Characteristics	n (%)
Median age at diagnosis (IQR), years	20 (14–26)
Median follow-up (IQR), months	53.5 (24.3–86.9)
Age at diagnosis, years	
10–18	27 (41.5%)
>18	38 (58.5%)
Gender	
Female	35 (53.8%)
Male	30 (46.2%)
Year of initial diagnosis	
2019–2024	30 (46.2%)
2014–2018	23 (35.4%)
2013 and earlier	12 (18.5%)
INRGSS stage	
L1	12 (18.5%)
L2	12 (18.5%)
M	40 (61.5%)
NA	1 (1.5%)
INRG risk classification	
Very low-risk	6 (9.2%)
Low-risk	11 (16.9%)
Intermediate-risk	5 (7.7%)
High-risk	42 (64.6%)
NA	1 (1.5%)
Tumor site	
Adrenal gland	24 (36.9%)
Retroperitoneal	24 (36.9%)
Posterior mediastinum	10 (15.4%)
Pelvic cavity	2 (3.1%)
Anterior mediastinum	1 (1.5%)
Hepatogastric space	1 (1.5%)
Unknown	3 (4.6%)

(Continued)

Table 1. (Continued)

Characteristics	n (%)
Symptoms at diagnosis	
Abdominal pain	16 (24.6%)
Bone pain	5 (7.7%)
Discovering lumps by physical examination or self-awareness	22 (33.8%)
Pale complexion	1 (1.5%)
Chest pain	4 (6.2%)
Lymph node enlargement	3 (4.6%)
Backache	8 (12.3%)
Lower limb pain	2 (3.1%)
Cough	2 (3.1%)
Difficulty breathing	1 (1.5%)
Eye swelling	1 (1.5%)
Histologic classification	
NB	45 (69.2%)
NB-undifferentiated	3 (4.6%)
NB-poorly differentiated	27 (41.5%)
NB-differentiating	12 (18.5%)
NB-unclassified	3 (4.6%)
GNB	20 (30.8%)
GNB-intermixed	11 (16.9%)
GNB-nodular-undifferentiated	2 (3.1%)
GNB-nodular-differentiating	7 (10.8%)
MYCN status	
Amplification	3 (6.7%)
Gain	2 (4.4%)
Nonamplification	40 (61.5%)
ALK status	
Mutation	5 (25%)
No mutation	15 (75%)

(Continued)

Table 1. (Continued)

Characteristics	*n* (%)
Metastatic sites of stage M	
Distant lymph node	25 (62.5%)
Bone	20 (50%)
Bone marrow	15 (37.5%)
Liver	6 (15%)
Lung	2 (5%)
Pleura	2 (5%)
Meninges	1 (2.5%)

GNB, ganglionic neuroblastoma; INRG, International Neuroblastoma Risk Group; INRGSS, International Neuroblastoma Risk Group Staging System; IQR, interquartile range; NA, not available; NB, neuroblastoma.

ganglionic neuroblastoma (GNB, including 11 intermixed GNB, 7 nodular-differentiating GNB, and 2 nodular-undifferentiated GNB).

MYCN oncogene evaluation results were available for 45 patients, revealing 3 (6.7%) patients with *MYCN* gene amplification and 2 (4.4%) patients with *MYCN* gene gain. *ALK* oncogene testing was performed on 20 patients, and *ALK* mutations were identified in 5 (25%) patients. One patient was initially diagnosed with a wild-type *ALK* gene; however, an *ALK* mutation was detected at the time of disease recurrence. Additionally, 3 of the 12 patients (25%) had *ATRX* mutations.

In this study, 12 patients underwent WES (5 at initial diagnosis and 7 at disease relapse) to comprehensively characterize the mutational landscape of adolescent and adult patients with NB. The genomic landscape of somatic mutations is shown in Figure 1(a), and the summary of genetic alterations is listed in Supplemental Table S2. Patients were stratified into the response group (CR/PR) and the non-response group (SD/PD) based on their best response to chemotherapy. Our genomic analyses revealed that certain genetic alterations in DNA repair and cell cycle pathways occurred more frequently in the non-response group (SD/PD) than in the response group (CR/PR). Specifically, the mutation rates of *ATRX*, *ARID1B*, *MLH1*, *POLD1*, *TP53*, and *EED* were higher in the PD/SD group than those in the CR/PR group. Representative cases were selected to demonstrate the association between therapeutic response and specific pathway gene mutations. Both Patient 62 and Patient 9 were adults (aged >18 years) diagnosed with poorly differentiated NB, classified as high-risk with INRGSS M stage. Patient 62 exhibited only SD as the best response to chemotherapy, accompanied by mutations in *ATRX*, *ARID1B*, and *TP53* within DNA repair and cell cycle pathways. In contrast, Patient 9 achieved CR without detectable mutations in these pathways. This divergence in pathway-specific genomic alterations likely contributed to their differential chemosensitivity and clinical outcomes. Following induction therapy, Patient 9 attained CR and received metronomic oral chemotherapy as maintenance treatment. Despite disease recurrence at 18 months, salvage chemotherapy reinduced CR, and the patient has remained disease-free. Conversely, Patient 62 demonstrated poor sensitivity to induction therapy (evaluated as PD), experienced rapid disease progression within 1 month, failed to respond to the same salvage regimen, and succumbed to the disease 4 months post-diagnosis.

In addition, six patients underwent plasma ctDNA surveillance, three at initial diagnosis and three at disease progression, as depicted in Figure 1(b). The consistency between ctDNA surveillance and imaging response evaluations was 83.3% (5/6) by comparing the dynamics trend of ctDNA and the disease status around the same time. Among 13 CR events identified by imaging, 61.5% (8/13) exhibited corresponding ctDNA negativity. Patient 30, a high-risk relapse case, initially achieved imaging-confirmed CR but with concurrent ctDNA positivity. Despite two subsequent imaging-confirmed CR assessments, ctDNA remained continuously positive, and disease progression was ultimately detected at the third evaluation, indicating that ctDNA monitoring provided a ≥4-month lead time for progression prediction. Following treatment, ctDNA clearance paralleled the achievement of sustained CR. Patient 18, a newly diagnosed case, achieved postoperative CR with concordant ctDNA negativity throughout follow-up, demonstrating 100% consistency with imaging assessments. Similarly, Patient 28, a relapsed case, exhibited progressive ctDNA burden reduction following salvage therapy and surgery, culminating in dual CR confirmation of imaging and ctDNA negativity. These cases indicate that ctDNA surveillance demonstrates high concordance with imaging-based response evaluations and enables earlier prediction of disease progression, highlighting its
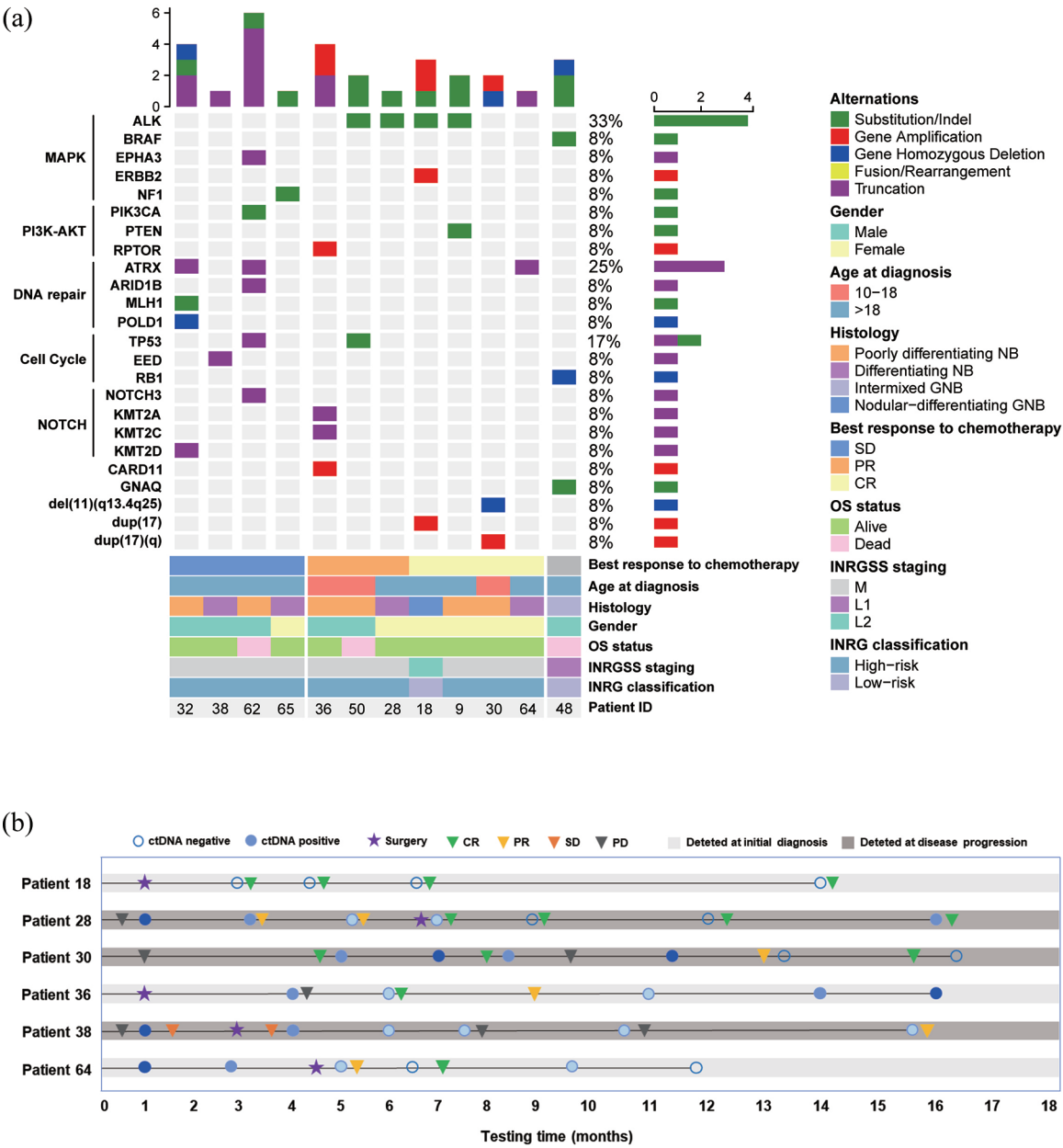
(a)

(b)

Figure 1. (a) The genomic landscape by WES from 12 patients. Clinical information is annotated in the bar plot at the bottom, with each column representing an individual case. Patients were stratified into the response group (CR/PR) and the non-response group (SD/PD) based on their best response to chemotherapy. Genomic analysis revealed a higher prevalence of genetic alterations in DNA repair and cell cycle pathways in the non-response group (SD/PD) compared to the response group (CR/PR). Specifically, mutations in *ATRX*, *ARID1B*, *MLH1*, *POLD1*, *TP53*, and *EED* were more frequently observed in the PD/SD group than in the CR/PR group. (b) Longitudinal monitoring by ctDNA and radiographic evaluation. Swimmer plot showing ctDNA status and clinical response evaluated by imaging. Solid blue dots indicate ctDNA positivity, with color intensity reflecting changes in ctDNA abundance, while hollow dots represent ctDNA negativity.
CR, complete response; ctDNA, circulating tumor DNA; PD, progressive disease; PR, partial response; SD, stable disease; WES, whole exome sequencing.

potential as a complementary biomarker for precision monitoring in adolescent and adult NB management.

Treatment and outcomes
All patients underwent chemotherapy ($n = 11$, 16.9%), surgery ($n = 12$, 18.5%), or combination

induction therapy (*n* = 42, 64.6%), with or without ASCT (*n* = 2, 3.1%), or radiation therapy (*n* = 23, 35.4%). At the end of the first induction therapy, 34 patients (53.2%) achieved overall CR. Other responses included 6 patients with PR, 1 patient with MR, 16 patients with SD, and 8 patients with PD.

Very low-risk and low-risk groups of NB

All 17 patients in the very low-risk and low-risk groups underwent surgical procedures, and complete resection was achieved in 13 (76.5%) patients. Additionally, 6 (35.3%) patients were administered chemotherapy. Following therapy, 16 patients achieved CR, and 1 patient had SD.

Tumor recurrence was observed in four patients, and various salvage treatments were explored, such as chemotherapy regimens for high-risk patients and a second surgery. At the data cutoff date, 1 patient had succumbed to the disease, 3 lived with the tumor, and the remaining 13 patients were disease-free.

Intermediate-risk group of NB

Among the five patients in the intermediate-risk group, four underwent surgery followed by chemotherapy, and one patient received only chemotherapy, with a median of 7 (IQR: 5–8) courses of chemotherapy. Additionally, radiotherapy was administered to two (45%) patients. At the end of treatment, three patients achieved CR, and two had SD.

Tumor recurrence occurred in one patient after 32.5 months, affecting the bones, lymph nodes, and pleura. The patient received subsequent treatments, including surgery, CAV/VIP chemotherapy regimens, VIT (vincristine, irinotecan, temozolomide) regimens, and radiotherapy. At the latest follow-up, one patient had died of disease progression, two were disease-free, and two continued to live with the tumor.

High-risk group of NB

All 42 high-risk patients received dose–intensity induction chemotherapy with a median of 7 (IQR: 5–8) cycles. Surgical intervention was performed in 32 (76.2%) patients, of whom 19 achieved complete resection, 10 underwent partial resection, and 3 lacked detailed surgical records. After

induction therapy, 14 (33.3%) patients achieved CR, 6 (14.3%) achieved PR, 1 (2.4%) showed MR, 13 (31%) had SD, and 8 (19%) had PD. In the consolidation therapy phase, 21 (50.0%) patients received radiation therapy: 13 for the primary lesion, 5 for both primary and metastatic lesions, 3 without radiation details, and 2 patients underwent ASCT. Subsequently, 18 (42.9%) patients received maintenance therapy, including two with anti-GD2 antibody (dinutuximab-β and naxitamab), 12 with metronomic chemotherapy, and four with cis-retinoic acid.

Nine patients had disease relapse, 22 had disease progression after first-line multimodal therapy, and the most common sites of recurrence or progression were the primary lesion (*n* = 11), bone (*n* = 10), and liver (*n* = 5). Based on the adopted treatment strategies, molecular testing results, and locations of tumor recurrence, various individualized salvage treatment regimens were attempted. These salvage strategies included IT (irinotecan, temozolomide)-based regimens, CT regimen (cyclophosphamide, topotecan), IC regimen (ifosfamide, carboplatin), and potentially beneficial drugs such as tyrosine kinase inhibitors (TKI: lorlatinib and anlotinib), apatinib, PD1 inhibitors, and chidamide. Second-line chemotherapy based on the IT regimen was administered to 17 patients, resulting in an objective response rate (ORR) of 41.2% (3 CR + 4 PR). Subsequent treatments included surgery in seven patients, radiotherapy in eight patients, anti-GD2 (naxitamab) immunotherapy in one patient, and metronomic chemotherapy in seven patients. Adverse effects in patients who received anti-GD2 immunotherapy were tolerable and manageable, including pain, paresthesia, elevated aminotransferase levels, and blurred vision. At the last follow-up, 18 patients remained alive, 5 were disease-free, and the others continued to live with the tumor. The 5-year PFS and OS rates of high-risk group patients were 16.6% ± 7.1% and 46.6% ± 9.6%, respectively.

Prognostic factors for survival

At a median follow-up time of 53.5 months (IQR: 24.3–86.9), the 3-year PFS and OS rates of the entire cohort were 47.2% ± 6.7% and 78% ± 5.7%, respectively; the 5-year PFS and OS rates were 33.1% ± 6.9% and 55.1% ± 7.6%, respectively (Figure 2).
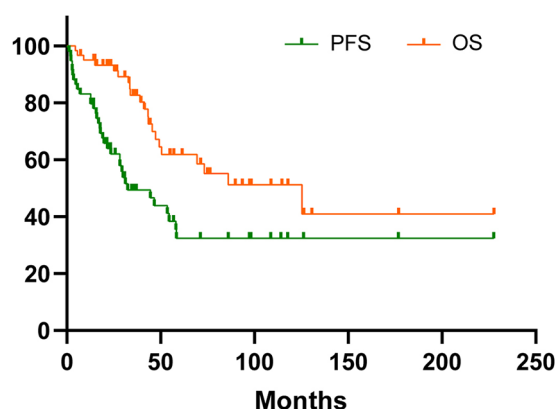
Figure 2. OS (orange line) and PFS (green line) for the entire cohort. The 3-year PFS and OS rates were 47.2% ± 6.7% and 78% ± 5.7%, respectively; the 5-year PFS and OS rates were 33.1% ± 6.9% and 55.1% ± 7.6%, respectively.
OS, overall survival; PFS, progression-free survival.

Among the 65 patients, Cox regression was used to assess the prognostic factors for PFS and OS with available follow-up data ($n = 60$; Table 2). In the univariate analysis, patients in the non-high-risk group ($p = 0.001$ for PFS, $p = 0.011$ for OS), locoregional (INRGSS L1 and L2 stage) NB ($p = 0.001$ for PFS, $p = 0.041$ for OS), and those who achieved CR after induction therapy ($p = 0.003$; PFS, $p = 0.004$ for OS) were associated with better outcomes. In addition, complete tumor resection ($p = 0.005$) and patients with histologic GNB ($p = 0.017$) showed superior PFS.

In multivariate analysis, CR after induction therapy (hazard ratio: 3.469, 95% confidence interval (CI): 1.359–8.854, $p = 0.009$) was independently associated with favorable PFS compared with non-CR (Figure 3). The 5-year PFS of these patients

Table 2. Univariate analysis for survival.

Variables	PFS		OS	
	HR (95% CI)	*p* Value	HR (95% CI)	*p* Value
Risk				
Non-high risk ($n = 22$)		0.001		0.011
High-risk ($n = 37$)	5.14 (1.98–13.38)		6.70 (1.55–28.88)	
INRGSS stage				
L1 ($n = 12$)		0.001		0.041
L2 ($n = 12$)	0.58 (0.10–3.49)		1.80 (0.16–19.23)	
M ($n = 35$)	4.86 (1.47–16.06)		7.03 (0.94–52.83)	
Histologic category				
GNB ($n = 19$)		0.017		
NB ($n = 41$)	3.17 (1.23–8.20)			
Response assessment after induction therapy				
CR ($n = 33$)		0.003		0.004
Non-CR ($n = 27$)	2.88 (1.44–5.73)		4.04 (1.56–10.49)	
Surgical procedures				
Complete resection ($n = 34$)		0.005		
Non-complete resection ($n = 14$)	4.42 (1.58–12.32)			

CI, confidence interval; CR, complete response; GNB, ganglionic neuroblastoma; HR, hazard ratio; INRGSS, International Neuroblastoma Risk Group Staging System; NB, neuroblastoma; OS, overall survival; PFS, progression-free survival.
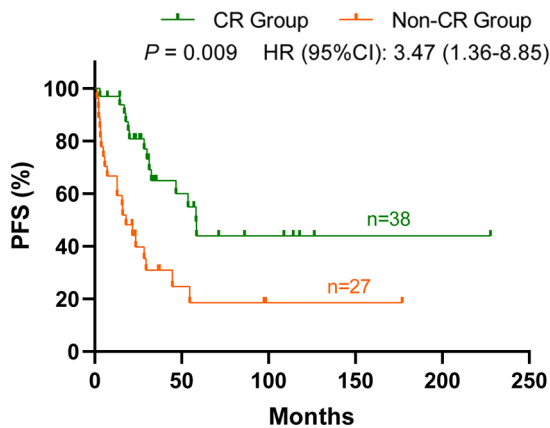
Figure 3. In multivariate analysis, achieving CR after induction therapy was associated with favorable PFS. The 5-year PFS of these patients with CR was 44.0% ± 10.6%, significantly superior to that of non-CR patients (18.5% ± 8.8%).
CR, complete response; PFS, progression-free survival.

with CR was 44.0% ± 10.6%, significantly superior to that of non-CR patients (18.5% ± 8.8%).

Comparison between adolescent and adult NB
The clinical and biological characteristics of the adolescent and adult patients with NB had no significant difference (Table 3). The survival data of the 3- and 5-year PFS rates were 41.3% ± 10.9% and 28.3% ± 10.7%, respectively, in adolescent patients; in adult patients, these rates were 55.5% ± 8.7% and 35.3% ± 9.8%, respectively ($p = 0.40$). The 3- and 5-year OS were 80.9% ± 8.6% and 54.5% ± 11.3% in the adolescent group, respectively; the 3- and 5-year OS were 84.0% ± 6.7% and 52.3% ± 10.7% in adult patients, respectively ($p = 0.208$). No significant difference was observed between the two groups.

Discussion
NB is rare in adolescents and adults, with unique clinical and biological characteristics. Standard treatment guidelines or chemotherapy protocols based on these populations are lacking, and recommendations are based on the experience from NB in younger patients. Enhancing our empirical understanding of this disease will provide guidance for future clinical practice. Therefore, we conducted a retrospective study to investigate the clinical characteristics, treatment modalities, and prognostic data of this rare disease, with the aim of providing more comprehensive references for clinical practice.

In our study, the observations on clinical characteristics are similar to those reported in previous adolescent and adult series.[6–8,17,23] The most common primary site of origin was the adrenal gland, but the most common metastatic site was the lymph nodes, which differed from the patterns in pediatric NB in which more bone marrow involvement was observed.[24] Previous studies have reported unusual patterns of metastatic localization in adolescents and adult patients with NB, such as in the lungs or central nervous system.[12,14] Similar to the findings of the pediatric cohort study,[1] patients in the high-risk NB group constituted the highest proportion. However, a higher proportion (61.5%) of patients presented with metastatic disease at diagnosis was observed, with only 40%–55% of pediatric patients presented with Stage 4 disease at diagnosis.[25–28] This discrepancy may be attributed to delayed diagnosis due to the rarity of adult-onset NB, as a longer interval between symptom onset and diagnosis has been reported.[7]

Although the induction regimens vary across different cooperative groups, the ORR for induction regimens typically ranges from 71% to 85% in pediatric patients with NB.[29,30] In contrast, our study showed that adolescent and adult patients with NB had a lower ORR of 61.5% to the first-line induction therapy. Similarly, regarding salvage chemotherapy regimens, a retrospective analysis of 46 pediatric patients with relapsed/refractory NB treated with the VIT regimen at our center demonstrated an ORR of 69.6%.[31] However, the adolescent and adult patients in our study responded poorly to salvage IT-based chemotherapy, with an ORR of only 41.2%. These findings suggest that adolescent and adult patients with NB may have lower chemotherapy sensitivity than pediatric patients, which may contribute to their poorer prognosis. Nevertheless, our multivariate analysis indicated that achieving CR after induction therapy was the only factor associated with better outcomes. Therefore, in clinical practice, an aggressive strategy and complete surgical resection should be adopted to achieve CR after first-line induction therapy to improve the prognosis of adolescent and adult patients with NB.[17,32]

Age at diagnosis is an independent factor highly correlated with NB prognosis.[33] However, in our

Table 3. Comparison of clinical, biological characteristics, and prognosis between adolescent and adult neuroblastomas.

Characteristics	Adolescent (*n* = 27)	Adult (*n* = 38)	*p* Value
Gender			0.074
Male	16 (59.3%)	14 (36.8%)	
Female	11 (40.7%)	24 (63.2%)	
Tumor site			0.234
Adrenal gland	9 (33.3%)	15 (39.5)	
Retroperitoneal	13 (48.1%)	11 (28.9%)	
Mediastinum	3 (11.1%)	8 (21.1%)	
Others	0	3 (7.9%)	
Unknown	2 (7.4%)	1 (2.6%)	
Tumor metastasis			0.621
With	7 (25.9%)	12 (31.6%)	
Without	20 (74.1%)	26 (68.4%)	
Distant metastasis model			0.131
Solitary distant lymph node metastasis	2 (11.1%)	6 (31.6%)	
Multiple metastasis	16 (88.9%)	13 (68.4%)	
INRGSS stage			0.162
L1	2 (7.4%)	10 (26.3%)	
L2	5 (18.5%)	7 (18.4%)	
M	19 (70.4)	21 (55.3%)	
NA	1 (3.7%)	0	
INRG risk classification			0.088
Very low-risk	0	6 (31.6%)	
Low-risk	3 (11.1%)	8 (21.1%)	
Intermediate-risk	3 (11.1%)	2 (5.3%)	
High-risk	20 (74.1%)	22 (57.9%)	
NA	1 (3.7%)	0	
MYCN status			0.073
Amplification	3 (15%)	0	
Gain	0	2 (5.3%)	
Nonamplification	17 (85%)	23 (92%)	

(Continued)

Table 3. (Continued)

Characteristics	Adolescent (*n* = 27)	Adult (*n* = 38)	*p* Value
ALK status			0.787
Mutation	2 (28.6%)	3 (23.1%)	
No mutation	5 (71.4%)	10 (76.9%)	
PFS rate			0.401
3-year	41.3% ± 10.9%	55.5% ± 8.7%	
5-year	28.3% ± 10.7%	35.3% ± 9.8%	
OS rate			0.208
3-year	80.9% ± 8.6%	84.0% ± 6.7%	
5-year	54.5% ± 11.3%	52.3% ± 10.7%	

INRG, International Neuroblastoma Risk Group; INRGSS, International Neuroblastoma Risk Group Staging System; OS, overall survival; PFS, progression-free survival; NA, not available.

subgroup analysis of adolescent and adult patients with NB, we did not observe a significant age-related impact on the outcomes. This aligns with findings from the SEER report[8] that age stratification did not show statistically significant prognostic differences. Nonetheless, when comparing the two major groups of adolescent/adult, and pediatric patients, adolescent and adult patients tend to exhibit poorer prognoses, as previously reported.[7,8,10,17] Specifically, in cases of *MYCN*-nonamplified in pediatric NB, the 3-year OS for the non-high-risk group typically exceeds 95%.[34–36] For high-risk pediatric patients, the 5-year OS rate is approximately 62.5%,[37] which is better than the survival rate observed in adults. Despite this, the survival outcomes in our study were more favorable than those of other large-scale studies on adult NB. In our cohort, the 3- and 5-year OS rates for patients over 20 years of age were 84.1% and 63.5%, respectively, which are higher than those of 125 participants based on SEER data, with 3- and 5-year OS rates of 45.9% and 36.3%.[8] Additionally, for adult patients with M-stage disease, the 3-, 5-, and 10-year OS rates were 77.9%, 49.5%, and 24.7%, respectively, which were superior to those reported by Conter et al.,[23] in which the 3-, 5-, and 10-year OS rates of 18 patients with M-stage disease were 68%, 33%, and 13%. In the near absence of ASCT and anti-GD2 antibody immunotherapy, our prognosis is comparable to that of a study conducted at the Memorial Sloan Kettering Cancer Center,

where 6 of 33 high-risk patients with NB underwent ASCT, and 7 of 33 patients received GD2 antibody treatment.[17] Compared to data from other upper-middle-income countries, our 5-year OS was significantly higher, at 55.1% versus 41.6%, 20.9%, and 14.2% reported by the other three centers.[12,38,39]

Several factors may have contributed to this discrepancy. Firstly, aggressive and personalized salvage treatment strategies were employed in patients with relapsed or refractory NB. Based on the patient's individual condition, molecular testing results, and family economic status, various treatment options were employed, including TKIs, VIT regimens, PD-1 monoclonal antibodies, anti-angiogenic drugs, and histone deacetylase inhibitors. Currently, the IT-based regimen has become the backbone chemotherapy for relapsed/refractory pediatric patients with NB, with ORR ranging from 26% to 69.6% when combined with other agents.[31,40–43] However, data on its efficacy in adult patients are limited. Our exploratory experience, which showed an ORR of 41.2%, suggests that the IT regimen may serve as an effective salvage therapy for adolescent and adult patients with NB. Moreover, for some high-risk patients, low-dose metronomic maintenance therapy was implemented. Previous studies have indicated that oral metronomic maintenance therapy can improve survival rates in high-risk pediatric patients with NB who have

not undergone ASCT or anti-GD2 antibody treatment.[44] Owing to the high cost of GD2 antibodies and the significant toxicity associated with ASCT, these treatments are less accessible in developing countries. Therefore, low-dose metronomic maintenance therapy is a viable alternative. Our limited data also suggest that metronomic maintenance therapy can benefit adult patients with NB. However, further prospective clinical trials are still needed.

NB relatively lacks recurrent somatic mutations, with only a few genes showing significant somatic mutation frequencies, including *ALK, PTPN11, ATRX, MYCN*, and *NRAS*.[45] Compared with pediatric patients with NB, adolescent and adult patients exhibit different patterns of genomic aberrations. *MYCN* amplification has been shown to be associated with advanced tumor stage and disease progression and is used as a biomarker for risk stratification.[46,47] *MYCN* amplification in NB is usually reported in 20%–30% of pediatric patients.[48] However, consistent with other studies,[8,10,17] only three patients in our cohort exhibited *MYCN* amplification. Franks et al.[10] inferred that the lack of *MYCN* amplification may contribute to the indolent course and slower progression of NB in adults. Conversely, a higher proportion of *ALK* gene mutations was observed in adult patients with NB, which is consistent with other studies.[17] Genetic alterations in *ALK* mutations and amplifications have also been observed in approximately 14% of high-risk NB cases and are regarded as independent predictors of poor survival.[49,50]

However, the genomic landscape of adult NB remains unclear. In our analysis of the genomic landscape of the 12 patients, those with poor responses to chemotherapy had a higher frequency of mutations in the cell cycle and DNA repair pathways. Chemotherapeutic medicines target proliferating cells, and alterations in cell cycle regulation and DNA damage repair can affect chemotherapy.[51] Studies on other cancer types have shown that the cell cycle and DNA repair pathways are associated with tumor sensitivity to chemotherapy.[52,53] Further investigation is needed to clarify the functional outcomes associated with these pathways in relation to chemotherapy response. Given the limited available guidelines and poor outcomes observed in adolescent and adult NB patients, comprehensive molecular testing and earlier implementation of targeted therapies may improve clinical outcomes and deepen our understanding of the genomic landscape of this rare malignancy.

ctDNA has been reported to correlate with disease burden, treatment response, and predict relapse.[54–56] In our study, the abundance of ctDNA was consistent with treatment response assessments based on imaging. Additionally, the noninvasive nature of ctDNA allows for multiple longitudinal tests. In our study, one patient was initially diagnosed with a wild-type *ALK* gene, but an *ALK* gene mutation was detected at recurrence. This reflects the heterogeneity and clonal evolution of tumors. Malignant clones of NB emerge early during embryonic development and rapidly evolve and diversify.[57] Capturing this dynamic and rapid clonal evolution is crucial for the personalized management and treatment of patients with NB. In summary, our limited data suggest that the dynamic monitoring of ctDNA could benefit the management of adult patients with NB, offering a more comprehensive option than single-molecule testing.

This study had some limitations. First, this retrospective study spanned 17 years, resulting in missing medical data. Second, GD2 antibody therapy was approved in China in 2021 and is costly, leading to limited data on anti-GD2 antibody therapy at our center. Finally, only 12 patients underwent genetic molecular testing, restricting a comprehensive molecular understanding of this rare disease.

Conclusion
Compared to pediatric patients, adolescent and adult patients with NB exhibit distinct biological characteristics and poorer prognoses, possibly due to reduced chemotherapy sensitivity. Mutations in the cell cycle and DNA repair pathways are associated with sensitivity to chemotherapy. ctDNA surveillance has potential benefits in the management of adolescent and adult patients with NB. Patients who achieve a CR after induction therapy may have a better prognosis. Metronomic maintenance therapy or various aggressive salvage strategies may improve survival in patients who do not receive anti-GD2 antibody therapy or ASCT. However, further exploration of treatment options is required.

Declarations

Author contributions
Weiji Xie: Conceptualization; Data curation; Formal analysis; Investigation; Methodology; Supervision; Writing – original draft; Writing – review & editing.

Yu Zhang: Conceptualization; Formal analysis; Methodology; Writing – review & editing.

Jiaqian Xu: Formal analysis; Writing – original draft; Writing – review & editing.

Feifei Sun: Writing – review & editing.

Jia Zhu: Data curation; Investigation.

Yi Que: Writing – review & editing.

Junting Huang: Writing – review & editing.

Zijun Zhen: Data curation; Investigation.

Suying Lu: Conceptualization; Data curation; Investigation; Supervision.

Juan Wang: Formal analysis; Writing – review & editing.

Yizhuo Zhang: Conceptualization; Supervision; Validation; Writing – review & editing.

ORCID iD
Yizhuo Zhang (iD) https://orcid.org/0000-0002-6128-3476

Supplemental material
Supplemental material for this article is available online.

References

1. Matthay KK, Maris JM, Schleiermacher G, et al. Neuroblastoma. *Nat Rev Dis Primers* 2016; 2: 16078.

2. London WB, Castleberry RP, Matthay KK, et al. Evidence for an age cutoff greater than 365 days for neuroblastoma risk group stratification in the Children's Oncology Group. *J Clin Oncol* 2005; 23: 6459–6465.

3. Shimada H, Umehara S, Monobe Y, et al. International neuroblastoma pathology classification for prognostic evaluation of patients with peripheral neuroblastic tumors: a report from the Children's Cancer Group. *Cancer* 2001; 92: 2451–2461.

4. Evans AE and D'Angio GJ. Age at diagnosis and prognosis in children with neuroblastoma. *J Clin Oncol* 2005; 23: 6443–6444.

5. Mossé YP, Deyell RJ, Berthold F, et al. Neuroblastoma in older children, adolescents and young adults: a report from the International Neuroblastoma Risk Group project. *Pediatr Blood Cancer* 2014; 61: 627–635.

6. Kushner BH, Kramer K, LaQuaglia MP, et al. Neuroblastoma in adolescents and adults: the Memorial Sloan-Kettering experience. *Med Pediatr Oncol* 2003; 41: 508–515.

7. Podda MG, Luksch R, Polastri D, et al. Neuroblastoma in patients over 12 years old:

a 20-year experience at the Istituto Nazionale Tumori of Milan. *Tumori* 2010; 96: 684–689.

8. Esiashvili N, Goodman M, Ward K, et al. Neuroblastoma in adults: incidence and survival analysis based on SEER data. *Pediatr Blood Cancer* 2007; 49: 41–46.

9. Rogowitz E, Babiker HM, Kanaan M, et al. Neuroblastoma of the elderly, an oncologist's nightmare: case presentation, literature review and SEER database analysis. *Exp Hematol Oncol* 2014; 3: 20.

10. Franks LM, Bollen A, Seeger RC, et al. Neuroblastoma in adults and adolescents: an indolent course with poor survival. *Cancer* 1997; 79: 2028–2035.

11. Mazzocco K, Defferrari R, Sementa AR, et al. Genetic abnormalities in adolescents and young adults with neuroblastoma: a report from the Italian Neuroblastoma group. *Pediatr Blood Cancer* 2015; 62: 1725–1732.

12. Tonya E, Jennifer G, Ané B, et al. Management outcomes of South African children diagnosed with neuroblastoma in adolescence. *Pediatr Hematol Oncol J* 2025; 10: 9–16.

13. Franks LM, Bollen A, Seeger RC, et al. Neuroblastoma in adults and adolescents. *Cancer* 1997; 79: 2028–2035.

14. Conte M, Parodi S, De Bernardi B, et al. Neuroblastoma in adolescents. *Cancer* 2006; 106: 1409–1417.

15. Gaspar N, Hartmann O, Munzer C, et al. Neuroblastoma in adolescents. *Cancer* 2003; 98: 349–355.

16. Castel V, Villamón E, Cañete A, et al. Neuroblastoma in adolescents: genetic and clinical characterisation. *Clin Transl Oncol* 2010; 12: 49–54.

17. Suzuki M, Kushner BH, Kramer K, et al. Treatment and outcome of adult-onset neuroblastoma. *Int J Cancer* 2018; 143: 1249–1258.

18. McCarthy LC, Chastain K, Flatt TG, et al. Neuroblastoma in adolescents and children older than 10 years: unusual clinicopathologic and biologic features. *J Pediatr Hematol Oncol* 2019; 41: 586–595.

19. von Elm E, Altman DG, Egger M, et al. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement: guidelines for reporting observational studies. *J Clin Epidemiol* 2008; 61: 344–349.

20. Monclair T, Brodeur GM, Ambros PF, et al. The International Neuroblastoma Risk Group (INRG) staging system: an INRG Task Force report. *J Clin Oncol* 2009; 27: 298–303.

21. Cohn SL, Pearson AD, London WB, et al. The International Neuroblastoma Risk Group (INRG) classification system: an INRG Task Force report. *J Clin Oncol* 2009; 27: 289–297.

22. Park JR, Bagatell R, Cohn SL, et al. Revisions to the international neuroblastoma response criteria: a consensus statement from the National Cancer Institute clinical trials planning meeting. *J Clin Oncol* 2017; 35: 2580–2587.

23. Conter HJ, Gopalakrishnan V, Ravi V, et al. Adult versus pediatric neuroblastoma: the M.D. Anderson Cancer Center experience. *Sarcoma* 2014; 2014: 375151.

24. DuBois SG, Kalika Y, Lukens JN, et al. Metastatic sites in stage IV and IVS neuroblastoma correlate with age, tumor biology, and survival. *J Pediatr Hematol Oncol* 1999; 21: 181–189.

25. Berthold F, Spix C, Kaatsch P, et al. Incidence, survival, and treatment of localized and metastatic neuroblastoma in Germany 1979–2015. *Paediatr Drugs* 2017; 19: 577–593.

26. Henderson TO, Bhatia S, Pinto N, et al. Racial and ethnic disparities in risk and survival in children with neuroblastoma: a Children's Oncology Group study. *J Clin Oncol* 2011; 29: 76–82.

27. Moroz V, Machin D, Faldum A, et al. Changes over three decades in outcome and the prognostic influence of age-at-diagnosis in young patients with neuroblastoma: a report from the International Neuroblastoma Risk Group Project. *Eur J Cancer* 2011; 47: 561–571.

28. Schroeder H, Wacher J, Larsson H, et al. Unchanged incidence and increased survival in children with neuroblastoma in Denmark 1981–2000: a population-based study. *Br J Cancer* 2009; 100: 853–857.

29. Garaventa A, Poetschger U, Valteau-Couanet D, et al. Randomized trial of two induction therapy regimens for high-risk neuroblastoma: HR-NBL1.5 International Society of Pediatric Oncology European Neuroblastoma Group Study. *J Clin Oncol* 2021; 39: 2552–2563.

30. Pinto N, Naranjo A, Hibbitts E, et al. Predictors of differential response to induction therapy in high-risk neuroblastoma: a report from the Children's Oncology Group (COG). *Eur J Cancer* 2019; 112: 66–79.

31. Zhu J, Wang J, Sun F, et al. Vincristine, irinotecan, and temozolomide in patients with

relapsed/refractory neuroblastoma. *Front Oncol* 2022; 12: 804310.

32. von Allmen D, Davidoff AM, London WB, et al. Impact of extent of resection on local control and survival in patients from the COG A3973 study with high-risk neuroblastoma. *J Clin Oncol* 2017; 35: 208–216.

33. Sokol E, Desai AV, Applebaum MA, et al. Age, diagnostic category, tumor grade, and mitosis-karyorrhexis index are independently prognostic in neuroblastoma: an INRG project. *J Clin Oncol* 2020; 38: 1906–1918.

34. Strother DR, London WB, Schmidt ML, et al. Outcome after surgery alone or with restricted use of chemotherapy for patients with low-risk neuroblastoma: results of Children's Oncology Group study P9641. *J Clin Oncol* 2012; 30: 1842–1848.

35. Baker DL, Schmidt ML, Cohn SL, et al. Outcome after reduced chemotherapy for intermediate-risk neuroblastoma. *N Engl J Med* 2010; 363: 1313–1323.

36. Bagatell R, Beck-Popovic M, London WB, et al. Significance of MYCN amplification in international neuroblastoma staging system stage 1 and 2 neuroblastoma: a report from the International Neuroblastoma Risk Group database. *J Clin Oncol* 2009; 27: 365–370.

37. Irwin MS, Naranjo A, Zhang FF, et al. Revised neuroblastoma risk classification system: a report from the Children's Oncology Group. *J Clin Oncol* 2021; 39: 3229–3241.

38. Casteleti P, Senerchia A and Caran E. Neuroblastoma in patients over 10 years old: a 21-year experience at a single institution. *Austin Pediatr* 2017; 4: 1052.

39. Jin QY, Du SB and Yuan XJ. Exploring the prognosis of neuroblastoma in adolescents and adults: a case series and literature review. *Neoplasma* 2022; 69: 464–473.

40. Mody R, Yu AL, Naranjo A, et al. Irinotecan, temozolomide, and dinutuximab with GM-CSF in children with refractory or relapsed neuroblastoma: a report from the Children's Oncology Group. *J Clin Oncol* 2020; 38: 2160–2169.

41. Kushner BH, Kramer K, Modak S, et al. Irinotecan plus temozolomide for relapsed or refractory neuroblastoma. *J Clin Oncol* 2006; 24: 5271–5276.

42. Lerman BJ, Li Y, Carlowicz C, et al. Progression-free survival and patterns of response in patients with relapsed high-risk neuroblastoma treated with irinotecan/temozolomide/dinutuximab/

granulocyte-macrophage colony-stimulating factor. *J Clin Oncol* 2023; 41: 508–516.

43. Moreno L, Weston R, Owens C, et al. Bevacizumab, irinotecan, or topotecan added to temozolomide for children with relapsed and refractory neuroblastoma: results of the ITCC-SIOPEN BEACON-neuroblastoma trial. *J Clin Oncol* 2024; 42: 1135–1145.

44. Sun X, Zhen Z, Guo Y, et al. Oral metronomic maintenance therapy can improve survival in high-risk neuroblastoma patients not treated with ASCT or anti-GD2 antibodies. *Cancers (Basel)* 2021; 13: 3494.

45. Pugh TJ, Morozova O, Attiyeh EF, et al. The genetic landscape of high-risk neuroblastoma. *Nat Genet* 2013; 45: 279–284.

46. Brodeur GM, Seeger RC, Schwab M, et al. Amplification of N-myc in untreated human neuroblastomas correlates with advanced disease stage. *Science* 1984; 224: 1121–1124.

47. Bordow SB, Norris MD, Haber PS, et al. Prognostic significance of *MYCN* oncogene expression in childhood neuroblastoma. *J Clin Oncol* 1998; 16: 3286–3294.

48. Seeger RC, Brodeur GM, Sather H, et al. Association of multiple copies of the N-myc oncogene with rapid progression of neuroblastomas. *N Engl J Med* 1985; 313: 1111–1116.

49. Bellini A, Pötschger U, Bernard V, et al. Frequency and prognostic impact of ALK amplifications and mutations in the European Neuroblastoma Study Group (SIOPEN) high-risk neuroblastoma trial (HR-NBL1). *J Clin Oncol* 2021; 39: 3377–3390.

50. Bresler SC, Weiser DA, Huwe PJ, et al. ALK mutations confer differential oncogenic activation and sensitivity to ALK inhibition therapy in neuroblastoma. *Cancer Cell* 2014; 26: 682–694.

51. Sun Y, Liu Y, Ma X, et al. The influence of cell cycle regulation on chemotherapy. *Int J Mol Sci* 2021; 22: 6923.

52. Iwamoto T, Bianchini G, Booser D, et al. Gene pathways associated with prognosis and chemotherapy sensitivity in molecular subtypes of breast cancer. *J Natl Cancer Inst* 2011; 103: 264–272.

53. Pitroda SP, Pashtan IM, Logan HL, et al. DNA repair pathway gene expression score correlates with repair proficiency and tumor sensitivity to chemotherapy. *Sci Transl Med* 2014; 6: 229ra42.

54. Newman AM, Bratman SV, To J, et al. An ultrasensitive method for quantitating circulating

tumor DNA with broad patient coverage. *Nat Med* 2014; 20: 548–554.

55. Dawson SJ, Tsui DW, Murtaza M, et al. Analysis of circulating tumor DNA to monitor metastatic breast cancer. *N Engl J Med* 2013; 368: 1199–1209.

56. Oxnard GR, Paweletz CP, Kuang Y, et al. Noninvasive detection of response and resistance in EGFR-mutant lung cancer using quantitative next-generation genotyping of cell-free plasma DNA. *Clin Cancer Res* 2014; 20: 1698–1705.

57. Gundem G, Levine MF, Roberts SS, et al. Clonal evolution during metastatic spread in high-risk neuroblastoma. *Nat Genet* 2023; 55: 1022–1033.