Received: 13 August 2018 | Revised: 17 May 2019 | Accepted: 29 May 2019

DOI: 10.1002/pbc.27900

Pediatric Blood & Cancer | SIOP International Society of Paediatric Oncology | aspho The American Society of Pediatric Hematology/Oncology | WILEY

RESEARCH ARTICLE

Phase I dose-escalation study of volasertib in pediatric patients with acute leukemia or advanced solid tumors

François Doz[1] | Franco Locatelli[2] | André Baruchel[3] | Nicolas Blin[4] | Barbara De Moerloose[5] | Didier Frappaz[6] | Michael Dworzak[7] | Matthias Fischer[8] | Jan Stary[9] | Rene Fuertig[10] | Kathrin Riemann[11] | Tillmann Taube[12] | Dirk Reinhardt[13]

[1]Oncology Center SIREDO (Care Innovation and Research for Children, Adolescents and Young Adults with Cancer), Institute Curie and University Paris Descartes, Paris, France

[2]Department of Paediatric Haematology and Oncology, IRCCS (Istituto di Recovero e Cura a Carattere Scientifico), Bambino Gesù Children's Hospital, Sapienza University of Rome, Rome, Italy

[3]Department of Paediatric Haemato-immunology, Hôpital Robert Debré (APHP), University Paris Diderot, Paris, France

[4]Paediatric Haematology and Oncology, Hôpital Mère-Enfant, CHU de Nantes, Nantes, France

[5]Pediatric Hematology-Oncology and Stem Cell Transplantation, Ghent University Hospital, Ghent, Belgium

[6]Paediatric Oncology Department, Léon Bérard Centre, Lyon, France

[7]St. Anna Children's Hospital, Department of Paediatrics, Medical University of Vienna, Vienna, Austria

[8]Department of Experimental Paediatric Oncology, University Children's Hospital Cologne, Centre of Molecular Medicine, Medical Faculty, University of Cologne, Cologne, Germany

[9]Department of Paediatric Haematology and Oncology, University Hospital Motol, Prague, Czech Republic

[10]Translational Medicine and Clinical Pharmacology, Boehringer Ingelheim Pharma GmbH & Co. KG, Biberach, Germany

[11]Clinical Operations, Boehringer Ingelheim Pharma GmbH & Co. KG, Ingelheim, Germany

[12]Medical Oncology, Boehringer Ingelheim International GmbH, Biberach, Germany

[13]Department of Paediatrics, University Hospital Essen, Essen, Germany

Correspondence
Dirk Reinhardt, Department of Paediatrics, University Hospital Essen, Hufelandstrasse 55, D-45147 Essen, Germany.
Email: dirk.reinhardt@uk-essen.de

Funding information
This study was sponsored by Boehringer Ingelheim Pharma GmbH & Co. KG, Germany.

Abstract

Background: Volasertib induces mitotic arrest and apoptosis by targeting Polo-like kinases. In this phase I dose-escalation study, the maximum tolerated dose (MTD), pharmacokinetics (PK), and preliminary efficacy of volasertib were determined in pediatric patients.

Methods: Patients aged 2 to <18 years with relapsed/refractory acute leukemia/advanced solid tumors (ST) without available effective treatments were enrolled—cohort C1 (aged 2 to <12 years); cohort C2 (aged 12 to <18 years). The patients received volasertib intravenously (starting dose: 200 mg/m^2 body surface area on day 1, every 14 days). The primary endpoint was the pediatric MTD for further development.

Results: Twenty-two patients received treatment (C1: leukemia, n = 4; ST, n = 8; C2: leukemia, n = 3; ST, n = 7). No dose-limiting toxicities (DLTs) occurred up to 300 mg/m^2 volasertib in C1;

Abbreviations: AEs, adverse events; ALL, acute lymphoblastic leukemia; AML, acute myeloid leukemia; AUC0-∞,norm, dose-normalized area under the curve from time 0 extrapolated to infinity; Cmax,norm, dose-normalized maximum plasma concentration; CNS, central nervous system; CTCAE, Common Terminology Criteria for Adverse Events; DLT, dose-limiting toxicity; DMC, data monitoring committee; EFS, event-free survival; IC_{50}, the half maximal inhibitory concentration; MTD, maximum tolerated dose; OR, overall response; OS, overall survival; PD, progressive disease; PFS, progression-free survival; PK, pharmacokinetics; PLK1, Polo-like kinase 1; QT, uncorrected QT interval; QTcF, corrected QT interval by Fridericia's correction of formula; SD, stable disease; ST, solid tumors.

two patients in C2, at 250 mg/m^2 volasertib, had DLTs in cycle 1, one of which led to death; therefore, the MTD of volasertib in C2 was 200 mg/m^2. The most common grade 3/4 adverse events (all patients) were febrile neutropenia, thrombocytopenia, and neutropenia (41% each). Stable disease (SD) was the best objective response (leukemia, n = 5; ST, n = 2); the duration of SD was short in all patients, except in one with an ST. PK profiles were generally comparable across dose groups and were consistent with those in adults.

Conclusion: The pediatric MTD/dose for further development was identified. There were no unexpected safety or PK findings; limited antitumor/antileukemic activity was demonstrated.

KEYWORDS

leukemia, pediatric cancers, pharmacokinetics, phase I, PLK inhibitor, solid tumors, volasertib

1 | INTRODUCTION

Approximately 1 in 285 children and adolescents will be diagnosed with cancer before they are 20 years old.[1] Over the last 50 years, treatment advances have increased the overall 5-year survival rate for childhood cancers to approximately 80%.[1,2] However, cancer is still the leading disease-related cause of death in those aged ≤19 years.[3] Mortality rates in pediatric patients with cancer are partially driven by the negative health impact of the intensive chemotherapy regimens necessary for effectively treating some cancers. These regimens can lead to acute morbidity and mortality, or cause significant long-term side effects that prevent therapy intensification and negatively affect treatment outcomes; survivors' quality of life may also be impaired.[2,4] As such, agents with innovative mechanisms of action and molecular targets, which provide improved clinical efficacy and favorable safety profiles, are urgently needed.

One such target is Polo-like kinase 1 (PLK1), a serine/threonine kinase that is active in several key stages of mitosis,[5] and—when dysregulated—may play a role in promoting carcinogenesis and malignant transformation.[6] High PLK1 expression has been observed in several human cancers and has been correlated with a poor prognosis.[7] Inhibition of PLK1 can cause cell-cycle arrest and apoptosis, preferentially in cancer cells over normal diploid or nondividing cells.[6,8–14] These data provide a strong rationale for targeting PLK1 with novel anticancer therapies.[15]

Volasertib is a small-molecule, ATP-competitive kinase inhibitor that potently and specifically inhibits PLK1 and two closely related kinases, PLK2 and PLK3 (with half maximal inhibitory concentration [IC_{50}] values of 0.87, 5, and 56 nmol/L, respectively).[16] Volasertib has an antimitotic mechanism of action, inhibiting cell proliferation via perturbation of spindle assembly, which leads to prometaphase cell-cycle arrest and subsequent apoptosis.[16] Preclinically, volasertib has shown antiproliferative effects in various cancer cell lines, alongside in vivo activity in multiple xenograft models.[16,17] Phase I and II clinical trials have determined the maximum tolerated dose (MTD) of volasertib, and evaluated its pharmacokinetics (PK), safety, and efficacy in adults with acute myeloid leukemia (AML) or solid tumors (ST). The MTD of volasertib monotherapy in adults with relapsed/refractory AML was 450 mg every 2 weeks. The most common adverse events (AEs) were cytopenia and related complications, with febrile neutropenia and infections being among the most common grade ≥3 AEs.[18] In adults with ST, the MTD was 400 mg every 3 weeks (Q3W); based on additional safety results, 300 mg Q3W was chosen as the recommended dose for further phase II development. Most AEs were hematologic and included anemia, neutropenia, and thrombocytopenia.[19] In a phase I dose-escalation trial, volasertib monotherapy demonstrated favorable PK and preliminary antitumor activity in adult patients with advanced ST.[15] However, the lack of convincing evidence of efficacy with monotherapy in unselected adult populations with ST in phase II trials led to discontinuation of volasertib development in this indication.[20–22] In a phase II trial in adult patients with previously untreated AML, volasertib combined with low-dose cytarabine (LDAC) achieved a higher response rate and improved event-free survival (EFS) compared to that achieved with LDAC monotherapy.[23] There have been promising preclinical data in childhood tumor models, which indicate volasertib monotherapy may have clinical benefit in pediatric ST.[8,24–27] This phase I study was conducted to identify the MTD or the relevant pediatric dose for further development of volasertib, alongside its safety, PK/pharmacodynamics, and efficacy in children with either relapsed/refractory acute leukemia or advanced ST, for whom no effective treatment was available.

2 | METHODS

2.1 | Patients

This multinational, multisite, open-label, noncontrolled, phase I dose-escalation trial enrolled patients aged 2 to <18 years with either relapsed/refractory acute leukemia after at least two previous intensive treatment regimens, or advanced ST, including lymphomas, for whom there was no known effective treatment at the time of trial entry. Patients were included if they had a Lansky score >60 (2 to <12 years age group) or Karnofsky score >60 (12 to <18 years age group), and a life expectancy of more than 6 weeks as judged by the investigator. Parents or legal guardians of the enrolled patients provided written informed consent; this was obtained from the patient whenever feasible.

Key exclusion criteria included Down syndrome, or the presence of symptomatic central nervous system (CNS) tumors, left ventricular shortening fraction by echocardiography <30%, and clinically relevant QT interval (QTcF; corrected QT interval by Fridericia's correction formula) prolongation. The study was conducted in accordance with the principles of the Declaration of Helsinki, the International Council for Harmonisation Guideline for Good Clinical Practice, and local legislation. The trial, approved by the ethical committee/institutional review board of the participating institutions, was registered at ClinicalTrials.gov (NCT01971476).

2.2 | Study design and endpoints

The primary endpoint was the pediatric MTD of volasertib, based on dose-limiting toxicities (DLTs) occurring in the first treatment cycle (cycle 1). If the MTD was not reached due to early signals of activity or tolerability issues, then the recommended pediatric dose of volasertib appropriate for further development was defined, based on the recommendations of an appointed external independent data monitoring committee (DMC). The MTD was defined as the highest dose at which no more than one out of six patients experienced DLTs during cycle 1.

Dose escalations were performed separately in two age groups of at least six evaluable patients (aged 2 to <12 years and 12 to <18 years) and proceeded via a standard "3 + 3" design. The DMC was responsible for the overall safety of the treated patients and for providing recommendations on dose escalation and de-escalation. Volasertib was administered as a single dose (intravenous infusion over ~1 h) on day 1 of each 14-day cycle. The starting dose was 200 mg/m^2 body surface area on day 1, and the dose was either escalated or de-escalated in steps of up to 50 mg/m^2 at each administration. Treatment cycles were repeated, until progressive disease (PD), drug-related AEs without recovery to predose grades (graded according to the National Cancer Institute's Common Terminology Criteria for Adverse Events [CTCAE] version 3.0), or other reasons for discontinuation. Secondary endpoints included the overall safety of volasertib, tolerability and PK outcomes, best overall response (OR), antileukemia activity, EFS/progression-free survival (PFS), overall survival (OS), and pharmacodynamics.

Patients could receive supportive care, including blood products, anti-infectious agents and analgesics, and growth factors such as granulocyte colony stimulating factor, according to local guidelines.

2.3 | Safety analysis

DLTs were defined as any drug-related CTCAE grade ≥3 AEs. Exceptions were reduced blood cell count (any grade) without associated clinical complications qualifying for DLT; febrile neutropenia (grade 3); infection (grade 3) with neutrophil count <1000 mm^3; elevated uric acid (grade 3); and nausea, vomiting, and/or diarrhea managed by adequate therapy (ie, recovery to grade ≤2). The first DLT analysis focused on MTD determination during the first treatment course only; a second analysis of DLTs during all courses provided support for determination of the MTD. The MTD was determined using data from all patients who received treatment during the dose-escalation/de-escalation phase. Following recovery from a DLT, therapy could be continued at a reduced dose. Analyses of overall safety and tolerability focused on the incidence and severity of AEs, the number of patients with clinically relevant laboratory value changes, and the number of patients with cardiac activity changes (prolonged QTcF interval).

2.4 | Efficacy analysis

Efficacy in patients with acute leukemia was assessed via bone marrow evaluation at the end of cycle 2, or earlier if PD was suspected; further bone marrow samples could be taken at the investigator's discretion. The best ORs were determined as described previously.[23,28] Assessment of efficacy in patients with ST was planned after four treatment cycles, and at the time of progression using tumor measurements and evaluation according to the Response Evaluation Criteria in Solid Tumors version 1.1.[29]

2.5 | PK analysis

Plasma concentrations of volasertib were determined by a validated high-performance liquid chromatography-tandem mass spectrometry assay. For quantification of plasma concentrations, 2 mL blood was taken as follows: immediately before volasertib administration; immediately after the end of the infusion; and at five additional time points in cycle 1: 1:20-1:40 h, 2:00-4:00 h, 24:00 h (± 4), 96:00 h (± 48), and 216:00 h (± 48) after the start of infusion. The following PK parameters were calculated by noncompartmental analysis and evaluated as secondary endpoints: dose-normalized area under the curve from time 0 extrapolated to infinity ($AUC_{0\text{-}\infty,norm}$); dose-normalized maximum plasma concentration ($C_{max,norm}$); plasma half-life ($t_{1/2}$); plasma clearance rate (CL); steady-state volume of distribution (V_{ss}); and mean residence time.

2.6 | Pharmacodynamic analysis

Pharmacodynamic endpoints included changes in neutrophil and platelet cell counts, QTcF prolongation, and reduction of leukemia blast cell count in peripheral blood (patients with leukemia only) after volasertib administration.

2.7 | Statistical analysis

All analyses were descriptive and exploratory in nature. Patients receiving at least one dose of the study drug (the treated set) were included in the primary endpoint analysis. Patients who had not completed at least one cycle for reasons other than DLT were replaced in order to determine the MTD, but were included in the treated set for all other analyses.

TABLE 1 Baseline patient characteristics

	Age 2 to <12 years				Age 12 to <18 years		
Volasertib dose (mg/m^2)	200	250	300	Total	200	250	Total
Number of patients, n (%)	3 (100.0)	3 (100.0)	6 (100.0)	12 (100.0)	6 (100.0)	4 (100.0)	10 (100.0)
Median age, years (range)	8.0 (3-10)	7.0 (3-9)	7.0 (2-11)	7.5 (2-11)	14.0 (12-17)	16.0 (14-17)	15.0 (12-17)
Male, n (%)	2 (66.7)	1 (33.3)	3 (50.0)	6 (50.0)	4 (66.7)	4 (100.0)	8 (80.0)
Median performance status[a] (range)	90.0 (70-90)	90.0 (80-100)	95.0 (70-100)	90.0 (70-100)	90.0 (80-100)	90.0 (70-100)	90.0 (70-100)
Cancer type, n (%)							
Acute leukemia[b]	2 (66.7)	2 (66.7)	0	4 (33.3)	2 (33.3)	1 (25.0)	3 (30.0)
ALL	2 (66.7)	1 (33.3)	0	3 (25.0)	1 (16.7)	1 (25.0)	2 (20.0)
AML	0	1 (33.3)	0	1 (8.3)	1 (16.7)	0	1 (10.0)
Solid tumor	1 (33.3)	1 (33.3)	6 (100.0)	8 (66.7)	4 (66.7)	3 (75.0)	7 (70.0)
Renal	0	0	1 (16.7)	1 (8.3)	2 (33.3)	0	2 (20.0)
Liver	0	0	1 (16.7)	1 (8.3)	0	0	0
Neuroblastoma	0	0	0	0	1 (16.7)	0	1 (10.0)
Bone[c]	0	0	2 (33.3)	2 (16.7)	0	2 (50.0)	2 (20.0)
Soft tissue	1 (33.3)	1 (33.3)	1 (16.7)	3 (25.0)	1 (16.7)	0	1 (10.0)
Other[d]	0	0	1 (16.7)	1 (8.3)	0	1 (25.0)	1 (10.0)
Previous treatment, n (%)							
Systemic anti-solid-tumor chemotherapies	1 (33.3)	1 (33.3)	6 (100.0)	8 (66.7)	4 (66.7)	3 (75.0)	7 (70.0)
Radiotherapy	1 (33.3)	1 (33.3)	5 (83.3)	7 (58.3)	5 (83.3)	2 (50.0)	7 (70.0)
Surgeries	1 (33.3)	0	6 (100.0)	7 (58.3)	4 (66.7)	3 (75.0)	7 (70.0)
Systemic antileukemia chemotherapies	2 (66.7)	2 (66.7)	0	4 (33.3)	2 (33.3)	1 (25.0)	3 (30.0)
Stem cell transplantations	2 (66.7)	2 (66.7)	0	4 (33.3)	4 (66.7)	2 (50.0)	6 (60.0)

Abbreviations: ALL, acute lymphoblastic leukemia; AML, acute myeloid leukemia.
[a]Lansky score (age 2 to <12 years) or Karnofsky score (age 12 to <18 years).
[b]All patients with leukemia failed chemotherapy and allogeneic stem cell transplantation prior to enrolment into this trial.
[c]Ewing sarcoma (n = 1, age 12 to <18 years); osteosarcomas (n = 2, age 2 to <12 years; n = 1, age 12 to <18 years).
[d]Desmoplastic small round cell tumors (n = 2).

3 | RESULTS

3.1 | Patients and treatment

Between October 2013 and March 2016, 22 patients were enrolled and received at least one cycle of volasertib; these included 12 patients in the 2 to <12 years age group (acute leukemia, n = 4; ST, n = 8) and 10 patients in the 12 to <18 years age group (acute leukemia, n = 3; ST, n = 7). Patient baseline characteristics are shown in Table 1. The median age was 7.5 years (range: 2-11) in the 2 to <12 years age group and 15.0 years (range: 12-17) in the 12 to <18 years age group; all patients had previously received systemic chemotherapy. Overall, seven patients had acute leukemia (acute lymphoblastic leukemia [ALL], n = 5; AML, n = 2) and 15 patients had ST. At data cut-off (March 31, 2016), all 22 patients had discontinued treatment, mostly due to PD (2 to <12 years, 91.7%; 12 to <18 years, 80.0%), and all seven patients with leukemia and 13/15 patients with ST had died.

In the 2 to <12 years age group, three patients received volasertib at a dose of 200 mg/m^2, three at 250 mg/m^2, and six at 300 mg/m^2 (Table 1). In the 12 to <18 years age group, six patients were treated with a volasertib dose of 200 mg/m^2 and four received 250 mg/m^2. The median treatment duration in the 2 to <12 years age group was 22.0 days (range: 15-71), and was longer in patients with leukemia than in patients with ST (37.5 days [range: 29-71] vs 15.0 days [range: 15-36]). In the 12 to <18 years age group, the median treatment duration was 15.0 days (range: 15-484), and did not differ by cancer type.

3.2 | DLTs and MTD

In patients aged 2 to <12 years, no DLTs were observed in any treatment cycle at volasertib doses of 200, 250, or 300 mg/m^2. Therefore, the DMC recommended that sufficient safety data had been collected for volasertib 300 mg/m^2 with no efficacy demonstrated that justified exposing additional patients to volasertib monotherapy; consequently, there were no dose escalations beyond 300 mg/m^2.

In patients aged 12 to <18 years, two patients treated at 250 mg/m^2 experienced six DLTs in cycle 1. These were intracranial hemorrhage (grade 5) in a patient with ALL (progressive leukemia had occurred in the CNS) and a platelet count of 24 × 10^9/L; and anemia (grade 4),

TABLE 2 Adverse events[a] occurring in >15% of patients in either age group by system organ class

System organ class Preferred term	Age 2 to <12 years n = 12			Age 12 to <18 years n = 10		
	All grades	G3	G4	All grades	G3	G4
Blood disorders	9 (75)	0	8 (67)	8 (80)	3 (30)	5 (50)
Anemia	6 (50)	3 (25)	1 (8)	4 (40)	2 (20)	1 (10)
Febrile neutropenia	5 (42)	5 (42)	0	4 (40)	3 (30)	1 (10)
Thrombocytopenia	4 (33)	0	4 (33)	5 (50)	1 (10)	4 (40)
Neutropenia	5 (42)	0	5 (42)	4 (40)	0	4 (40)
Leukopenia	2 (17)	0	2 (17)	2 (20)	0	2 (20)
Gastrointestinal disorders	6 (50)	0	0	7 (70)	3 (30)	0
Vomiting	3 (25)	0	0	6 (60)	1 (10)	0
Nausea	2 (17)	0	0	5 (50)	1 (10)	0
Abdominal pain	3 (25)	0	0	3 (30)	0	0
Diarrhea	2 (17)	0	0	1 (10)	0	0
Constipation	0	0	0	2 (20)	0	0
General disorders/ administration-site conditions	6 (50)	2 (17)	0	7 (70)	2 (20)	0
Pyrexia	2 (17)	0	0	3 (30)	1 (10)	0
Fatigue	2 (17)	0	0	3 (30)	0	0
Chest pain	0	0	0	2 (20)	0	0
Asthenia	1 (8)	1 (8)	0	2 (20)	1 (10)	0
Musculoskeletal and connective tissue disorders	0	0	0	5 (50)	1 (10)	0
Myalgia	0	0	0	2 (20)	0	0
Arthralgia	0	0	0	2 (20)	0	0
Nervous system disorders[b]	3 (25)	0	0	5 (50)	0	1 (10)
Headache	1 (8)	0	0	4 (40)	1 (10)	0
Investigations	5 (42)	0	2 (17)	5 (50)	3 (30)	2 (20)
WBC count decreased	2 (17)	1 (8)	1 (8)	2 (20)	0	2 (20)
Platelet count decreased	2 (17)	0	1 (8)	2 (20)	0	1 (10)
ALT/AST increased	2 (17)	0	0	2 (20)	0	0
Blood creatinine increased	2 (17)	0	0	1 (10)	1 (10)	0
Lymphocyte count decreased	2 (17)	1 (8)	0	2 (20)	0	0
Neutrophil count decreased	1 (8)	0	1 (8)	2 (20)	0	2 (20)
Blood phosphorus decreased	0	0	0	2 (20)	1 (10)	0
Electrocardiogram QT prolonged	1 (8)	0	0	2 (20)	1 (10)	0
Metabolism and nutrition disorders	4 (33)	2 (17)	0	4 (40)	1 (10)	0
Hyponatremia	2 (17)	0	0	2 (20)	1 (10)	0
Decreased appetite	3 (25)	0	0	0	0	0
Hyperglycemia	1 (8)	0	0	2 (20)	0	0
Hypercalcemia	1 (8)	0	0	2 (20)	0	0
Neoplasms benign, malignant, and unspecified	3 (25)	1 (8)	0	1 (10)[c]	0	0
Malignant neoplasm progression	2 (17)[c]	0	0	1 (10)	0	0
Respiratory, thoracic, and mediastinal disorders	4 (33)	0	1 (8)	3 (30)	0	1 (10)
Epistaxis	0	0	0	2 (20)	0	0

(Continues)

TABLE 2 (Continued)

System organ class Preferred term	Age 2 to <12 years n = 12			Age 12 to <18 years n = 10		
	All grades	G3	G4	All grades	G3	G4
Skin and subcutaneous tissue disorders	5 (42)	0	0	3 (30)	0	0
Pruritus	1 (8)	0	0	2 (20)	0	0

Abbreviations: AEs, adverse events; ALT, alanine aminotransferase; AST, aspartate aminotransferase; CTCAE, Common Terminology Criteria for Adverse Events; G, grade; QT, uncorrected QT interval; WBC, white blood cell.

Note. The worst CTCAE grade AEs are listed independently for system organ class and preferred term; all observed AEs are listed for the preferred term, while only the AE of the highest grade for each patient is listed for the system organ class.

[a]AEs by treatment, worst CTCAE grade, system organ class, and preferred team are listed.

[b]In the 2 to <12 year age group, this includes cranial nerve disorder, headache, and dysesthesia (each n = 1). In the 12 to <18 year age group, this includes intracranial hemorrhage, paresthesia, third nerve disorder, paraplegia, and peripheral sensory neuropathy (each n = 1) and headache (n = 4).

[c]All events were grade 5.

febrile neutropenia (grade 4), thrombocytopenia (grade 4), leukopenia (grade 4), gastrointestinal hemorrhage (grade 3), and asthenia (grade 3) in a patient with osteosarcoma. Therefore, in patients aged 12 to <18 years, the MTD of volasertib was 200 mg/m^2. Additional DLTs were observed in later treatment cycles—QTcF prolongation occurred in one patient with leukemia treated at 200 mg/m^2 (grade 3, cycle 6), and in one patient with an ST treated at 250 mg/m^2 (grade 2, cycle 22).

3.3 | Other safety outcomes

All patients experienced at least one AE. AEs occurring in more than 15% of patients overall are summarized in Table 2. In the overall study population, the most frequent grade 3/4 AEs were febrile neutropenia, thrombocytopenia, neutropenia (41% each), and anemia (32%). The most common drug-related AEs of any grade were anemia (41%), febrile neutropenia (36%), neutropenia (36%), and thrombocytopenia (36%); these are consistent with volasertib's known myelosuppressive effects. Seventeen (77%) patients experienced at least one serious AE; these included 7/12 (58%) patients aged 2 to <12 years and 10/10 (100%) patients aged 12 to <18 years.

Analysis of QTcF changes in relation to plasma concentrations indicated potential concentration-dependent QTcF prolongation in pediatric patients; QTcF prolongation with volasertib has been found to be due to inhibition of the hERG potassium channel (IC_{50} 2.4 μmol/L). The mean increase from baseline QTcF to the end of volasertib infusion ranged from 36.7 to 40.4 ms in patients aged 2 to <12 years, and from 21.6 to 41.8 ms in patients aged 12 to <18 years. After the end of the infusion, the mean QTcF prolongation decreased and reached values lower than or equaling baseline values 1 day after infusion in patients aged 12 to <18 years, and in patients aged 2 to <12 years treated at 200 mg/m^2. For patients aged 2 to <12 years who received volasertib ≥250 mg/m^2, mean QTcF prolongation persisted beyond 24 hours following the end of infusion.

3.4 | Efficacy outcomes

Efficacy results by age group and tumor type are summarized in Table 3. Across all the patients treated, the best OR was stable disease (SD). SD was achieved in five patients with leukemia (2 to <12 years, n = 4/4 [100%]; 12 to <18 years, n = 1/3 [33%]), all of whom had reduced leukemia blast counts in peripheral blood compared with that at baseline disease. In addition, two patients with ST (neuroblastoma and osteosarcoma) achieved SD, both aged 12 to <18 years (n = 2/7 [29%]). For the patient with neuroblastoma (age 15 years at study entry), 8.5 years had elapsed between the first diagnosis and study entry, during which time the patient received three lines of polychemotherapy. For the patient with osteosarcoma (age 17 years at study entry), the time between the first diagnosis and study entry was 6.8 years; the patient had received five lines of polychemotherapy. For both patients, mitotic spindle inhibitors (vincristin, vindesin, and vinblastin) were among the previous treatments.

For patients with leukemia with a best OR of SD, EFS and OS were short in duration (maximum EFS ≤3.2 months, maximum OS ≤7.3 months). Of the two patients with ST who had a best response of SD, one patient (male, osteosarcoma) had received 28 cycles of volasertib treatment with a PFS of 15.7 months and was alive at data cut-off (censored OS of 15.9 months). This patient initially received 250 mg/m^2; this was subsequently reduced to 200 mg/m^2 due to grade 2 QTcF prolongation following cycle 22 (recovered within 2 days). The second patient (male, neuroblastoma) received six cycles of volasertib treatment with a PFS of 5.2 months and was alive at data cut-off (censored OS of 10.3 months). The patient had been treated with 200 mg/m^2 volasertib.

3.5 | Volasertib PK parameters

PK data were available for all treated patients. Volasertib geometric mean plasma concentration-time curves for cycle 1 are displayed by age group (Figure 1). The profiles were of similar shape for all dose groups, and volasertib C_{max} was generally observed around the end of the ~1-h intravenous administration. The $t_{1/2}$ of volasertib was long, and low but quantifiable plasma concentrations could be measured 10-14 days after the first infusion. Moderate to high interindividual variability was evident. PK parameters for volasertib are summarized in Table 4. Dose-normalized exposure parameters (AUC, C_{max}) tended to be higher in the age group of 2 to <12 years compared with the 12 to

TABLE 3 Best OR, EFS/PFS, and OS with volasertib in patients with leukemia and ST

	Age 2 to <12 years	Age 12 to <18 years
Patients with leukemia, n	4	3
Patient with any response evaluations post baseline, n (%)	4 (100)	3 (100)
Best OR, n (%)		
CR	0	0
CRi	0	0
PR	0	0
SD	4 (100)[a]	1 (33)[a]
PD	0	2 (67)
Median EFS, months (range)	2.1 (1.0-3.2)	0.9 (0.2-3.1)
Median OS, months (range)	2.7 (1.7-4.0)	7.3 (0.5-8.7)
Patients with an ST, n	8	7
Patients with any response evaluations post baseline, n (%)	7 (88)	6 (86)
Best OR, n (%)		
CR	0	0
PR	0	0
SD	0	2 (29)[b,c]
PD	7 (88)	4 (57)
Missing	1 (13)	1 (14)
Median PFS, months (range)	0.7 (0.4-1.0)	1.7 (0.7-15.7)
Median OS, months (range)	1.9 (0.7-11.9)	3.2 (0.9-15.9)

Abbreviations: CR, complete remission; CRi, complete remission with incomplete blood count recovery; EFS, event-free survival; OR, overall response; OS, overall survival; PD, progressive disease; PFS, progression-free survival; PR, partial response; SD, stable disease; ST, solid tumors.

[a]Including transient reduction of blasts.

[b]For up to 28 cycles in a patient with osteosarcoma.

[c]Including transient reduction of tumor marker neuron-specific enolase in a patient with neuroblastoma.

<18 years age group. CL and V_{ss} were higher in the 12 to <18-year-old group than in the 2 to <12-year-old group. No difference in mean residence time was apparent between age groups; however, $t_{1/2}$ was longer in the younger than in the older age group. There was no apparent deviation from dose proportionality, as dose-normalized $AUC_{0-\infty}$ was comparable between doses in the two age groups.

4 | DISCUSSION

In this open-label, noncontrolled, phase I dose-escalation study in pediatric patients with acute leukemia or ST without suitable alternative treatment options, the MTD of volasertib was 200 mg/m^2 in patients aged 12 to <18 years. In patients aged 2 to <12 years, the pediatric dose of volasertib for use in further development was 300 mg/m^2 (MTD not reached). Volasertib's safety profile was similar in both groups, and AEs were as expected given volasertib's mechanism of action and the safety outcomes reported in adult studies. The most frequently observed grade ≥3 AEs were either hematological (thrombocytopenia, neutropenia, and anemia) or resulted from myelosuppression (febrile neutropenia).

In patients with heavily pretreated relapsed/refractory malignancies, we observed preliminary evidence of limited antitumor and antileukemic activity, with SD reported as the best response in five patients with leukemia and two patients with ST. No specific reasons could be identified as to why the two patients with ST achieved disease stabilization and consequently received extended volasertib treatment. For both, the time between initial diagnosis and enrolment was prolonged, which may indicate that their particular tumors were only slowly proliferating. The observation that volasertib had limited activity in the remaining patients is consistent with findings in adults showing only limited antileukemic activity with volasertib monotherapy at doses less than or equal to 350 mg.[23,30] The populations of both age groups were heterogeneous; together with the small sample size, this meant the analysis of EFS/PFS and OS was not possible.

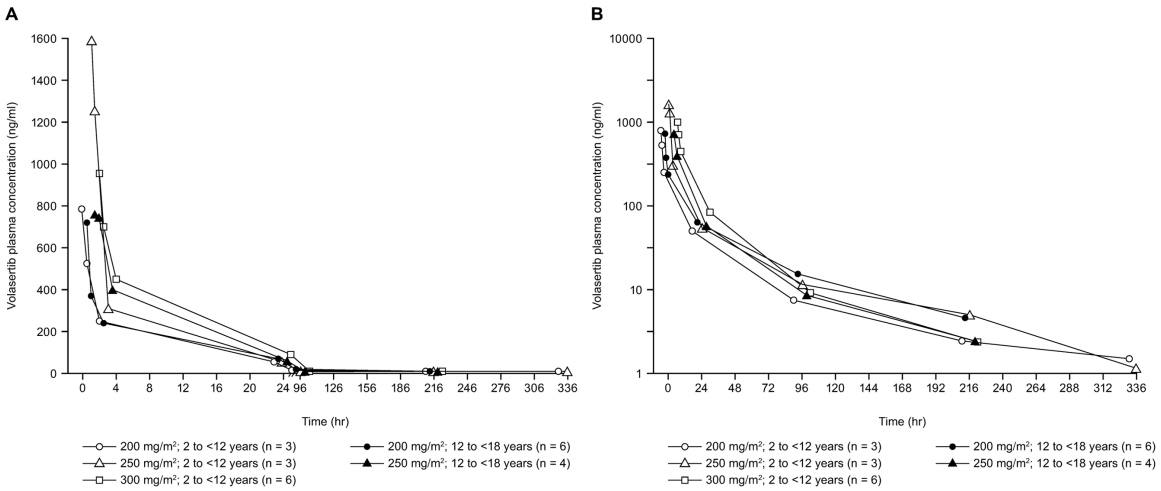

FIGURE 1 Geometric mean drug plasma concentration-time profiles of volasertib after intravenous infusion of 200, 250, and 300 mg/m^2 of volasertib in cycle 1, by age group—(A) linear scale, (B) semi-log scale

TABLE 4 Summary of PK parameters of volasertib after the first infusion

Parameter[a]	Dose group, mg/m^2	Pooled		Age 2 to <12 years		Age 12 to <18 years	
		n	gMean (gCV, %)	n	gMean (gCV, %)	n	gMean (gCV, %)
$AUC_{0-\infty,norm}$, ng × h/mL/mg	200	9	32.4 (31.6)	3	41.7 (40.1)	6	28.6 (19.0)
	250	7	31.6 (70.3)	3	51.2 (69.8)	4	22.1 (37.9)
	300	6	36.4 (28.2)	6	36.4 (28.2)	0	–
$C_{max,norm}$, ng/mL/mg	200	9	3.19 (62.2)	3	5.34 (22.0)	6	2.46 (54.9)
	250	7	4.47 (149)	3	9.71 (128)	4	2.50 (94.6)
	300	6	3.60 (35.1)	6	3.60 (35.1)	0	–
$t_{1/2}$, h	200	9	85.6 (29.0)	3	102 (28.6)	6	78.6 (27.3)
	250	7	77.6 (53.8)	3	130 (14.1)	4	52.7 (17.7)
	300	6	54.8 (27.2)	6	54.8 (27.2)	0	–
CL, mL/min	200	9	514 (31.6)	3	400 (40.1)	6	583 (19.0)
	250	7	527 (70.3)	3	326 (69.8)	4	755 (37.9)
	300	6	458 (28.2)	6	458 (28.2)	0	–
V_{ss}, L	200	9	1820 (50.5)	3	1270 (4.0)	6	2180 (52.9)
	250	7	1710 (116)	3	1170 (103)	4	2270 (128)
	300	6	855 (54.6)	6	855 (54.6)	0	–
MRT, h	200	9	59.0 (47.7)	3	53.1 (35.7)	6	62.3 (55.7)
	250	7	54.1 (70.8)	3	59.9 (44.8)	4	50.1 (98.0)
	300	6	31.1 (29.4)	6	31.1 (29.4)	0	–

Abbreviations: $AUC_{0-\infty,norm}$, dose-normalized area under the curve from time 0 extrapolated to infinity; $C_{max,norm}$, dose-normalized maximum plasma concentration; CL, plasma clearance rate; gCV, geometric coefficient of variation; gMean, geometric mean; MRT, mean residence time; PK, pharmacokinetics; $t_{1/2}$, plasma half-life; V_{ss}, volume of distribution at steady state.

[a]The trough concentration of volasertib (predose concentration in plasma immediately before administration of the second dose [$C_{pre,2}$]) was not evaluated, as most patients had only received one cycle of volasertib treatment.

Albeit based on a limited patient population, PK data were comparable with data from clinical studies in adults, in which volasertib demonstrated dose proportionality, moderate plasma clearance, a high volume of distribution, and a long $t_{1/2}$.[19,30] Given the limited number of subjects and the moderate-to-high PK variability, it is not possible to draw robust conclusions about age-related differences in these patients. However, it appears that the younger age group had higher systemic exposure and higher peak plasma concentrations of volasertib than the older age group did, whereas CL and V_{ss} were higher in the 12 to <18-year-old group than in the 2 to <12-year-old group. There was no difference in mean residence time between age groups, but $t_{1/2}$ appeared to be longer in the younger group than in the older group. Given the PK properties of volasertib, $t_{1/2}$ derived from noncompartmental analysis may not directly reflect elimination, but instead may be related to redistribution from deep compartments. Volasertib distributes to lysosomes, and is a cationic amphiphilic compound with a tendency to accumulate in acidic organelles[31]; this may explain the large observed V_{ss}. A more detailed assessment and development of PK models is required to characterize the potential age dependence of clearance or volume of distribution in children. The fact that dose-normalized data were similar between age groups indicates that exposure was proportional to dose within the range administered. No direct relationship between plasma exposure and toxicity could be identified. The small sample size and the heterogeneity of the patients enrolled may explain why complications related to myelosuppression were observed in the older age group only. However, the incidence of cytopenia resulting from the myelosuppressive effect of volasertib was similar between age groups.

Of note, this study did not enroll patients <2 years of age, so the results may not apply to this group. Neutrophil and platelet counts decreased after administration of volasertib compared with pretreatment values, although no meaningful interpretation of the correlation with PK parameters was possible due to the small sample size. Correlation analyses suggested a positive relationship between volasertib plasma concentrations and changes in QTcF in pediatric patients, in agreement with previous observations in adult patients.[19,22,32]

Clinical development of volasertib was discontinued in 2017 following a strategic decision made by the sponsor. Nevertheless, given that high expression of PLK1 has been correlated with poor prognosis,[7] and its inhibition causes cell-cycle arrest and apoptosis in tumor cells,[6,8–14] PLK1 remains an attractive target for anticancer drugs. The results from this trial provide further insight into the efficacy and pharmacodynamics of volasertib in different pediatric tumor types, which may help to identify patients likely to respond to alternative PLK1 inhibitors. Our findings confirm that PLK1 inhibition can mediate antitumor effects, albeit modest, in pediatric patients. Further investigation in studies on other PLK inhibitors may be beneficial to

investigate combination treatments based on preclinical evidence of synergy,[33–35] and to identify potential biomarkers predictive of clinical activity.

In conclusion, the MTD of volasertib when given every 14 days was 200 mg/m^2 in patients aged 12 to <18 years, while for patients aged 2 to <12 years, the pediatric dose for further development was 300 mg/m^2. The pediatric doses described here are in the same range or higher than the doses of single-agent volasertib recommended for adults. Safety, PK, and pharmacodynamic findings were as expected, and volasertib showed limited antitumor/antileukemic activity in this heavily pretreated pediatric population. Although the clinical development of volasertib has been terminated, these results may be relevant to the development of other PLK inhibitors for pediatric patients with cancer.

ACKNOWLEDGMENTS

We thank all participating patients and their families, and the nursing and medical staff, especially Dr Maria Pia Cefalo, Dr Giuseppe Maria Milano, Dr Benoit Brethon, Dr Marie-Dominique Tabone, Dr Bram De Wilde, Dr Nadège Corradini, Cécile Giraud, Dr Isabelle Aerts, Dr Irene Jimenez, and Anne Maclou.

Medical writing assistance, supported financially by Boehringer Ingelheim, was provided by Nabeela Farooq of GeoMed, an Ashfield company, part of UDG Healthcare plc, during the preparation of this manuscript. The authors were fully responsible for all content and editorial decisions, were involved at all stages of manuscript development, and have approved the final version.

CONFLICT OF INTEREST

Rene Fuertig, Kathrin Riemann, and Tillmann Taube are employees of Boehringer Ingelheim. Matthias Fischer received honoraria from Novartis. The remaining authors have nothing to declare.

DATA SHARING

To ensure independent interpretation of clinical study results, Boehringer Ingelheim grants all external authors access to all relevant material, including participant-level clinical study data, and relevant material as needed by them to fulfill their role and obligations as authors under ICMJE criteria.

Furthermore, clinical study documents (eg, study report, study protocol, statistical analysis plan) and participant clinical study data are available to be shared after publication of the primary manuscript in a peer-reviewed journal and if regulatory activities are complete and other criteria met per the BI Policy on Transparency and Publication of Clinical Study Data (https://trials.boehringer-ingelheim.com/transparency_policy.html).

Prior to providing access, documents will be examined and, if necessary, redacted and the data will be de-identified, to protect the personal data of study participants and personnel, and to respect the boundaries of the informed consent of the study participants.

Clinical study reports and related clinical documents can be requested via the following link: https://trials.boehringer-ingelheim.com/trial_results/clinical_submission_documents.html. All such requests will be governed by a document sharing agreement.

Bona fide, qualified scientific and medical researchers may request access to de-identified, analyzable participant clinical study data with corresponding documentation describing the structure and content of the datasets. Upon approval, and governed by a data sharing agreement, data are shared in a secured data-access system for a limited period of 1 year, which may be extended upon request.

Researchers should use https://clinicalstudydatarequest.com to request access to study data.

ORCID

Didier Frappaz https://orcid.org/0000-0002-3684-9909
Dirk Reinhardt https://orcid.org/0000-0001-6300-3434

REFERENCES

1. Ward E, DeSantis C, Robbins A, Kohler B, Jemal A. Childhood and adolescent cancer statistics, 2014. *CA Cancer J Clin*. 2014;64(2):83-103.
2. Kaspers GJ, Creutzig U. Pediatric acute myeloid leukemia: international progress and future directions. *Leukemia*. 2005;19(12):2025-2029.
3. National Cancer Institute. Cancer in children and adolescents fact sheet. 2017. https://www.cancer.gov/types/childhood-cancers/child-adolescent-cancers-fact-sheet#r1 [Accessed December 18, 2018].
4. Hunger SP, Lu X, Devidas M, et al. Improved survival for children and adolescents with acute lymphoblastic leukemia between 1990 and 2005: a report from the Children's Oncology Group. *J Clin Oncol*. 2012;30(14):1663-1669.
5. Takaki T, Trenz K, Costanzo V, Petronczki M. Polo-like kinase 1 reaches beyond mitosis-cytokinesis, DNA damage response, and development. *Curr Opin Cell Biol*. 2008;20(6):650-660.
6. Smith MR, Wilson ML, Hamanaka R, et al. Malignant transformation of mammalian cells initiated by constitutive expression of the Polo-like kinase. *Biochem Biophys Res Commun*. 1997;234(2):397-405.
7. Liu Z, Sun Q, Wang X. PLK1, a potential target for cancer therapy. *Transl Oncol*. 2017;10(1):22-32.
8. Ackermann S, Goeser F, Schulte JH, et al. Polo-like kinase 1 is a therapeutic target in high-risk neuroblastoma. *Clin Cancer Res*. 2011;17(4):731-741.
9. Guan R, Tapang P, Leverson JD, Albert D, Giranda VL, Luo Y. Small interfering RNA-mediated Polo-like kinase 1 depletion preferentially reduces the survival of p53-defective, oncogenic transformed cells and inhibits tumor growth in animals. *Cancer Res*. 2005;65(7):2698-2704.
10. Jackson JR, Patrick DR, Dar MM, Huang PS. Targeted anti-mitotic therapies: can we improve on tubulin agents? *Nat Rev Cancer*. 2007;7(2):107-117.
11. Liu X, Lei M, Erikson RL. Normal cells, but not cancer cells, survive severe Plk1 depletion. *Mol Cell Biol*. 2006;26(6):2093-2108.
12. Renner AG, Dos Santos C, Recher C, et al. Polo-like kinase 1 is overexpressed in acute myeloid leukemia and its inhibition preferentially targets the proliferation of leukemic cells. *Blood*. 2009;114(3):659-662.
13. Steegmaier M, Hoffmann M, Baum A, et al. BI 2536, a potent and selective inhibitor of Polo-like kinase 1, inhibits tumor growth in vivo. *Curr Biol*. 2007;17(4):316-322.
14. Xie S, Xie B, Lee MY, Dai W. Regulation of cell cycle checkpoints by Polo-like kinases. *Oncogene*. 2005;24(2):277-286.

15. Schöffski P. Polo-like kinase (PLK) inhibitors in preclinical and early clinical development in oncology. *Oncologist*. 2009;14(6):559-570.
16. Rudolph D, Steegmaier M, Hoffmann M, et al. BI 6727, a Polo-like kinase inhibitor with improved pharmacokinetic profile and broad antitumor activity. *Clin Cancer Res*. 2009;15(9):3094-3102.
17. Gjertsen BT, Schöffski P. Discovery and development of the Polo-like kinase inhibitor volasertib in cancer therapy. *Leukemia*. 2015;29(1):11-19.
18. Ottmann OG, Muller-Tidow C, Kramer A, et al. Phase I dose-escalation trial investigating volasertib as monotherapy or in combination with cytarabine in patients with relapsed/refractory acute myeloid leukaemia. *Br J Haematol*. 2019;184:1018-1021.
19. Schöffski P, Awada A, Dumez H, et al. A phase I, dose-escalation study of the novel Polo-like kinase inhibitor volasertib (BI 6727) in patients with advanced solid tumours. *Eur J Cancer*. 2012;48(2):179-186.
20. Pujade-Lauraine E, Selle F, Weber B, et al. Volasertib versus chemotherapy in platinum-resistant or -refractory ovarian cancer: a randomized phase II Groupe des Investigateurs Nationaux pour l'Etude des Cancers de l'Ovaire study. *J Clin Oncol*. 2016;34(7):706-713.
21. Ellis PM, Leighl NB, Hirsh V, et al. A randomized, open-label phase II trial of volasertib as monotherapy and in combination with standard-dose pemetrexed compared with pemetrexed monotherapy in second-line treatment for non-small-cell lung cancer. *Clin Lung Cancer*. 2015;16(6):457-465.
22. Stadler WM, Vaughn DJ, Sonpavde G, et al. An open-label, single-arm, phase 2 trial of the Polo-like kinase inhibitor volasertib (BI 6727) in patients with locally advanced or metastatic urothelial cancer. *Cancer*. 2014;120(7):976-982.
23. Döhner H, Lübbert M, Fiedler W, et al. Randomized, phase 2 trial comparing low-dose cytarabine with or without volasertib in AML patients not suitable for intensive induction therapy. *Blood*. 2014;124(9):1426-1433.
24. Gorlick R, Kolb EA, Keir ST, et al. Initial testing (stage 1) of the Polo-like kinase inhibitor volasertib (BI 6727), by the Pediatric Preclinical Testing Program. *Pediatr Blood Cancer*. 2014;61(1):158-164.
25. Harris PS, Venkataraman S, Alimova I, et al. Polo-like kinase 1 (PLK1) inhibition suppresses cell growth and enhances radiation sensitivity in medulloblastoma cells. *BMC Cancer*. 2012;12:80.
26. Lanvers-Kaminsky C, Abbou S, udigeos-Dubus E, et al. Anti-tumor activity of the PLK inhibitor volasertib (BI 6727) and the aurora kinase inhibitor BI 811283 in pediatric malignancies. *Eur J Cancer*. 2012;48(Suppl 6):78.
27. Smith M, Keir S, Maris J, et al. Pediatric Preclinical Testing Program (PPTP) evaluation of volasertib (BI 6727), a Polo-like kinase (PLK) inhibitor. *Cancer Res*. 2012;72(8):LB-317.
28. Creutzig U, van den Heuvel-Eibrink MM, Gibson B, et al. Diagnosis and management of acute myeloid leukemia in children and adolescents: recommendations from an international expert panel. *Blood*. 2012;120(16):3187-3205.
29. Eisenhauer EA, Therasse P, Bogaerts J, et al. New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1). *Eur J Cancer*. 2009;45(2):228-247.
30. Bug G, Müller-Tidow C, Schlenk RF, et al. Phase I/II study of volasertib (BI 6727), an intravenous Polo-like kinase (PLK) inhibitor, in patients with acute myeloid leukemia (AML): updated results of the dose finding phase I part for volasertib in combination with low-dose cytarabine (LD-Ara-C) and as monotherapy in relapsed/refractory AML. *Blood*. 2011;118(21):1549.
31. Solans BP, Fleury A, Freiwald M, Fritsch H, Haug K, Troconiz IF. Population pharmacokinetics of volasertib administered in patients with acute myeloid leukaemia as a single agent or in combination with cytarabine. *Clin Pharmacokinet*. 2018;57(3):379-392.
32. Kobayashi Y, Yamauchi T, Kiyoi H, et al. Phase I trial of volasertib, a Polo-like kinase inhibitor, in Japanese patients with acute myeloid leukemia. *Cancer Sci*. 2015;106(11):1590-1595.
33. Abbou S, Lanvers-Kaminsky C, Daudigeos-Dubus E, et al. Polo-like kinase inhibitor volasertib exhibits antitumor activity and synergy with vincristine in pediatric malignancies. *Anticancer Res*. 2016;36(2):599-609.
34. Weiss LM, Hugle M, Romero S, Fulda S. Synergistic induction of apoptosis by a Polo-like kinase 1 inhibitor and microtubule-interfering drugs in Ewing sarcoma cells. *Int J Cancer*. 2016;138(2):497-506.
35. Hugle M, Belz K, Fulda S. Identification of synthetic lethality of PLK1 inhibition and microtubule-destabilizing drugs. *Cell Death Differ*. 2015;22(12):1946-1956.

How to cite this article: Doz F, Locatelli F, Baruchel A, et al. Phase I dose-escalation study of volasertib in pediatric patients with acute leukemia or advanced solid tumors. *Pediatr Blood Cancer*. 2019;66:e27900. https://doi.org/10.1002/pbc.27900