Published OnlineFirst February 18, 2019; DOI: 10.1158/1078-0432.CCR-18-2675

Clinical Trials: Targeted Therapy

Clinical Cancer Research

A Phase II Study of Alisertib in Children with Recurrent/Refractory Solid Tumors or Leukemia: Children's Oncology Group Phase I and Pilot Consortium (ADVL0921)

Yael P. Mossé[1,2], Elizabeth Fox[1,2], David T. Teachey[1,2], Joel M. Reid[3], Stephanie L. Safgren[3], Hernan Carol[4], Richard B. Lock[4], Peter J. Houghton[5], Malcolm A. Smith[6], David Hall[7], Donald A. Barkauskas[7], Mark Krailo[8], Stephan D. Voss[9], Stacey L. Berg[10], Susan M. Blaney[10], and Brenda J. Weigel[9]

Abstract

Purpose: Aurora A kinase (AAK) plays an integral role in mitotic entry, DNA damage checkpoint recovery, and centrosome and spindle maturation. Alisertib (MLN8237) is a potent and selective AAK inhibitor. In pediatric preclinical models, antitumor activity was observed in neuroblastoma, acute lymphoblastic leukemia, and sarcoma xenografts. We conducted a phase 2 trial of alisertib in pediatric patients with refractory or recurrent solid tumors or acute leukemias (NCT01154816).

Patients and Methods: Alisertib (80 mg/m^2/dose) was administered orally, daily for 7 days every 21 days. Pharmacogenomic (PG) evaluation for polymorphisms in the AURK gene and drug metabolizing enzymes (UGT1A1*28), and plasma pharmacokinetic studies (PK) were performed. Using a 2-stage design, patients were enrolled to 12 disease strata (10 solid tumor and 2 acute leukemia). Response was assessed after cycle 1, then every other cycle.

Results: A total of 139 children and adolescents (median age, 10 years) were enrolled, 137 were evaluable for response. Five objective responses were observed (2 complete responses and 3 partial responses). The most frequent toxicity was myelosuppression. The median alisertib trough concentration on day 4 was 1.3 μmol/L, exceeding the 1 μmol/L target trough concentration in 67% of patients. No correlations between PG or PK and toxicity were observed.

Conclusions: Despite alisertib activity in pediatric xenograft models and cogent pharmacokinetic-pharmacodynamic relationships in preclinical models and adults, the objective response rate in children and adolescents receiving single-agent alisertib was less than 5%.

Introduction

The Aurora kinase family is essential in the regulation of chromosome segregation and cytokinesis during mitosis (1). Aurora A kinase (AAK) plays an integral role in mitotic entry, DNA damage checkpoint recovery, and centrosome and spindle maturation (1). Aurora A is overexpressed in many adult tumors including bladder, breast, lung, and head and neck cancers (1–4); as well as pediatric malignancies (5–8). The Aurora A kinase gene (AURK) has 2 common polymorphisms; the phe31Ile polymorphism, which alters the kinase function and is associated with tumorigenesis or advanced cancers (9, 10); and the Va571Ile polymorphism, which in combination with phe31IIe may be associated with an increased risk of cancer- or treatment-related adverse events (11–14).

Alisertib (MLN8237) is a potent and selective AAK inhibitor previously investigated alone and in combination with chemotherapy as a potential treatment for patients with relapsed/refractory peripheral T-cell lymphoma as well as advanced solid tumors (15–18). In preclinical models, antitumor activity and maximum pharmacodynamic effect were associated with alisertib concentrations exceeding 1 μmol/L (18, 19). In adults, the recommended dose of alisertib is 50 mg twice daily for 7 days with associated dose-limiting toxicities (DLT) of neutropenia and stomatitis (20). The maximum concentration (C_{max}) and area under concentration time curve (AUC) following administration of 50-mg enteric coated tablets were 2.9 μmol/L and 20.9 μmol/L•h, respectively; and the steady state trough concentration (C_{min}) exceeded 1 μmol/L (21).

[1]Division of Oncology and Center for Childhood Cancer Research, Children's Hospital of Philadelphia, Pennsylvania. [2]Department of Pediatrics, Perelman School of Medicine at the University of Pennsylvania, Philadelphia, Pennsylvania. [3]Mayo Clinic, Rochester, Minnesota. [4]Children's Cancer Institute, Lowy Cancer Research Centre, UNSW Sydney, Sydney, Australia. [5]Greehey Children's Cancer Research Institute, University of Texas Health Science Center San Antonio, San Antonio, Texas. [6]Cancer Therapy Evaluation Program, NCI, Bethesda, Maryland. [7]Children's Oncology Group, Monrovia, California. [8]Department of Preventive Medicine, University of Southern California, Los Angeles, California. [9]Dana-Farber/Boston Children's Cancer and Blood Disorders Center, Boston, Massachusetts. [10]Texas Children's Cancer Center, Baylor College of Medicine, Houston, Texas. [11]Division of Pediatric Oncology, University of Minnesota Cancer Center, Minneapolis, Minnesota.

Note: Supplementary data for this article are available at Clinical Cancer Research Online (http://clincancerres.aacrjournals.org/).

Corresponding Author: Yael P. Mossé, Children's Hospital of Philadelphia, Colket Translational Research Building, 3501 Civic Center Blvd-Office 3056, Philadelphia, PA 19104-4318. Phone: 215-590-0965; Fax: 267-426-0685; E-mail: mosse@chop.edu

Clin Cancer Res 2019;25:3229–38

doi: 10.1158/1078-0432.CCR-18-2675

Published OnlineFirst February 18, 2019; DOI: 10.1158/1078-0432.CCR-18-2675

Translational Relevance

The Aurora kinases are a family of serine-threonine kinases that play an essential role in regulating chromosome assembly and segregation during mitosis and are critical for cell proliferation. Aurora A dysregulation has been implicated in cancer, and as such, is a rational therapeutic target. This phase 2 study of alisertib (MLN2837), an oral small molecular inhibitor of Aurora A kinase, was evaluated in 137 pediatric patients with relapsed/refractory solid tumors or acute leukemia. The lack of robust objective responses suggests that alisertib, as a single agent, has limited anti-tumor activity and alternative strategies, including novel combinations simultaneously targeting other oncogenic signaling pathways, should be explored to harness this pathway and simultaneously minimize toxicity. The continuous treatment schedule of alisertib, which was shown to be more effective in preclinical models than the 1-week administration schedule, was not feasible in the clinical setting, providing a potential explanation for the differential observed between preclinical and clinical anti-tumor activity.

Pharmacokinetic parameters derived from a population pharmacokinetic (PK) model based on data from 363 adults enrolled on 7 alisertib single-agent trials showed a terminal half-life of 19.3 hours and an apparent clearance (CL/F) of 4.25 L/h (22). Although the major metabolic pathway of alisertib is glucuronidation via the UDP-glucuronosyltransferase, UGT1A1, alisertib CL/F was not altered in adult subjects with UGT1A1*28 polymorphisms. Single-agent phase II trials of alisertib demonstrated modest activity in adults with ovarian cancer (23), acute myeloid leukemia (AML; ref. 24), and T- or B-cell lymphoma (17).

In the Pediatric Preclinical Testing Program (PPTP) alisertib was active in neuroblastoma (NBL) and acute lymphoblastic leukemia (ALL) xenografts with maintained complete responses observed in 3 of 7 NBL xenografts and 6 of 6 ALL xenografts; sustained concentrations $\geq$1 μmol/L were associated with response (25). Statistically significant improvement in event-free survival (EVS) in Wilms tumor (WT), rhabdomyosarcoma (RMS), and osteosarcoma (OS) xenograft models was also observed (26). In these preclinical studies, alisertib was administered 5 days per week for 3 and 6 consecutive weeks for ALL and solid tumor models, respectively. In addition, alisertib was active in p53-wild-type, therapy-refractory NBL cell lines (27), as a result of disruption of the Aurora-A/N-Myc complex resulting in inhibition of N-Myc dependent transcription (28). Alisertib has also been shown to induce cell death and augment radiation sensitivity in atypical teratoid rhabdoid (ATRT) cell lines that overexpress AAK and have mutations in SMARCB1 (SNF5/INI1), a tumor suppressor and component of chromatin remodeling (7).

Based on the preclinical antitumor activity in pediatric cell lines and xenograft models and the clinical antitumor activity in adult trials, a Children's Oncology Group phase I trial of alisertib in children and adolescents with relapsed or refractory solid tumors evaluated both once daily and twice daily schedules (29). The recommended phase II dose and schedule was 80 mg/m^2 orally, once daily for 7 days. DLTs included myelosuppression, mood alterations, somnolence, mucositis, fatigue, alopecia, elevated hepatic transaminases, agitation, and euphoria. In contrast to adults, the twice daily schedule in children resulted in a higher frequency of neutropenia and palmar-plantar erythrodysesthesia. There was marked inter-patient variability in the alisertib PK parameters in children. At the recommended dose, the C_{max} and AUC were 7.5 ± 0.1 μmol/L and 75 ± 13.5 μmol/L•h, respectively. The alisertib trough 24 hours after the first dose at the 80 mg/m^2 dose level was 1.1 μmol/L. Of 33 response evaluable children in the phase I trial, 1 with hepatoblastoma (HBL) had a PR and 8 [NBL (n = 4) and sarcoma (n = 4)] had stable disease for 5 to 35 cycles (29).

Based on the mechanism of action and preclinical activity of alisertib, a phase II trial was conducted to evaluate the objective response rate of alisertib in children and adolescents with relapsed/refractory solid tumors or acute leukemia. In parallel, and subsequent to the establishment of a dose and schedule for alisertib in a pediatric population, we selected 2 ALL xenografts against which we have previously reported single-agent alisertib efficacy at a dose resulting in drug plasma levels that are achievable in humans for additional testing to compare dosing schedules.

Patients and Methods

Patient population

Patients with relapsed or refractory cancer were enrolled in 1 of 12 strata based on histology, including 2 strata for NBL; one for those with measurable disease by CT or MRI and another for those with disease evaluable by MIBG scintigraphy, but no measurable disease. Patients with NBL limited to the bone marrow were not eligible. Patients with other solid tumors were required to have measurable disease as defined by RECIST 1.1 (30). Strata for patients with sarcoma included rhabdomyosarcoma (RMS), non-RMS soft tissue sarcoma (NR-STS), OS, or Ewing sarcoma (EWS)/peripheral PNET. Additional solid tumor strata included enrollment of patients with WT, hepatoblastoma (HBL), malignant germ cell tumors (GCT), and rhabdoid tumors (central nervous system atypical teratoid rhaboid or other malignant rhabdoid tumors) with loss of INI1 by IHC or molecular analysis. Patients with hematologic malignancies without CNS involvement who were refractory or recurrent after at least 2 prior induction chemotherapy regimens, including those with AML and at least 5% myelobasts in the bone marrow or those with ALL and greater than 25% blasts (M3) in bone marrow, were also eligible. Subjects enrolled on the COG phase I trial (ADVL0812) who received alisertib at the recommended phase II dose and who met criteria for inclusion in 1 of the 12 disease stratum defined in this trial were included in this study population by prospective design (29).

Patients were required to swallow alisertib tablets intact. Other inclusion criteria included age >12 months and <22 years; performance status of $\geq$50 by the Karnofsky scale for patients >16 years or by the Lansky scale if $\leq$16 years; adequate renal function (normal serum creatinine for age and gender); and hepatic function (total bilirubin $\leq$1.5-fold greater than the upper limit of normal, alanine aminotransferase (ALT) less than 225 U/L, and serum albumin of at least 2 g/dL) was required. In patients with solid tumors, bone marrow function for patients without known tumor infiltration of bone marrow included an absolute neutrophil count (ANC) $\geq$1,000/μL, platelet count

Published OnlineFirst February 18, 2019; DOI: 10.1158/1078-0432.CCR-18-2675

≥100,000/μL, and hemoglobin ≥8 g/dL; for patients with solid tumors and known bone marrow metastatic disease an ANC ≥750/μL, platelet count ≥50,000/μL, and hemoglobin ≥8 g/dL were required. Patients with leukemia could enroll if they were not refractory to red blood cell or platelet transfusions.

Patients were required to have recovered from the acute toxic effects of all prior treatment. Requirements for the interval of time from prior therapy were standard (29). Exclusion criteria included uncontrolled infection; pregnancy; lactation; concurrent administration of selected P-glycoprotein substrates (digoxin, cyclosporine, tacrolimus or sirolimus); or use of daily benzodiazepines, because of the potential benzodiazepine-like effects of alisertib.

This study was conducted in compliance with the Declaration of Helsinki, the International Conference on Harmonization, Guideline for Good Clinical Practice, and applicable national and local regulatory requirements. Institutional Review Boards at participating institutions approved the study. Informed consent was obtained from patients, ages 18 years or older, or from parents/legal guardians of children aged less than 18 years, with child assent when appropriate, according to institutional policies.

Treatment program

Alisertib (Millennium Pharmaceuticals, Inc.) was distributed by the NCI Cancer Therapy Evaluation Program as 10-mg enteric-coated tablets. Alisertib (80 mg/m^2) was administered orally once daily for 7 consecutive days. Cycle duration was 21 days. The dose was reduced to 60 mg/m^2 for reversible toxicity as outlined in the protocol. The maximum daily dose of alisertib was 160 mg. Adherence was monitored using daily dosing diaries completed by the patient or parent/guardian. In the absence of progressive disease or unacceptable toxicity, the maximum total duration of protocol therapy was 35 cycles, approximately 2 years.

Toxicity monitoring and dose modifications

The Common Terminology and Adverse Events (CTCAE v 4.0) criteria were used to grade toxicity. Prior to each cycle, physical examination, complete blood count (CBC), serum electrolytes, creatinine, and liver function tests were performed. During cycle 1, physical examinations and serum chemistries were performed weekly; CBCs were performed twice weekly. In subsequent cycles, CBCs were performed weekly or more frequently if hematologic toxicity occurred.

Response

Disease evaluations were performed at baseline, the end of cycle 1 and after completion of every other cycle of protocol therapy. In patients with non-CNS solid tumors, response was assessed using RECIST version 1.1 criteria (30). Response in NBL subjects with nonmeasurable but MIBG evaluable disease was assessed using the Curie Score criteria (31). Response for subjects with AML was assessed using the International Working Group Criteria (32) and for those with ALL response was defined by morphology. Response in patients with central nervous system ATRT was assessed based on the sum of the products of the longest diameter × perpendicular diameter. All objective responses were confirmed by central review.

Any eligible patient who received at least 1 dose of alisertib was considered evaluable for response provided: (i) the patient was observed on protocol therapy for at least 1 cycle and the tumor was not removed surgically prior to the time an objective response was confirmed; or (ii) the patient demonstrated a complete or partial response as confirmed by central review; or (iii) the patient demonstrated progressive disease or died while on protocol therapy. All other evaluable patients with solid tumors were considered to be nonresponders. The maximum evaluation period for determination of the overall best response was 6 treatment cycles for ADVL0921; the evaluation period for determination of overall best response for ADVL0812 was the time from enrollment to termination of protocol therapy.

Pharmacokinetics and pharmacogenomics

To characterize the PKs of alisertib in children and adolescents, blood samples (3 mL, EDTA) were required in all participants during cycle 1 prior to alisertib administration on days 1, 4 ± 1, and 7 ± 1. If consent was provided, optional sampling was performed at 1 to 2, 3 to 4, and 6 to 8 hours after the first dose. Plasma was stored at −80°C until analysis. Alisertib concentrations were measured as previously described and PK parameters were calculated using noncompartmental analyses (29, 33).

Consenting patients provided whole blood in EDTA tubes prior to day 7 of the first cycle for genotyping of patients for germline polymorphisms in UGT1A1 or aurora AAK gene (AURK, Phe31Ile, and Val57Ile). DNA was extracted by QIAamp DNA Blood Mini Kit as per manufacturer's instructions. Methods were validated with a panel of 60 Caucasian DNA samples from the Coriell Institute. Positive and negative controls were included for each analysis. For *UGT1A1 *28* (rs8175347), the number of TA repeats in the promoter region were detected and quantified by a modification of the method described by Akaba and colleagues (34). UGT1A1 polymorphisms rs4124874 and rs10929302 were evaluated with PCR amplification and dye-terminator sequencing. Specific primers were designed and validated to amplify the region for both SNPs. Forward and reverse primers are AGTTCTCTTCACCTCCTCCT and AATAAA CCCCACCTCACCAC, respectively. For *AURKA*, genotyping for the G>A polymorphism (rs1047972 in codon 57) and T>A polymorphism (rs2273535 in codon 31) was performed by amplification and detected on a Bio-Rad CFX384 Real-Time PCR detection system. The real-time PCR methods were validated against a standard PCR reaction with sequence detection of the polymorphisms. Primer and probe sequences were provided by Millennium Pharmaceuticals, Inc. The forward and reverse primer sequences for rs227353 were CTGGCCACTATTTACAGG-TAATGGA and TGGAGGTCCAAAACGTGTTCTC, respectively with probe/reporter 1 (VIC-labeled) sequence ACTCAGCAA-TTTCCTT and probe/reporter 2 (FAM-labeled) sequence CTCA-GCAAATTCCTT. The forward and reverse primer sequences for rs1047972 were CGGCTTGTGACTGGAGACA and GGGTC-TTGTGTCCTTCAAATTCTTC, respectively with probe/reporter 1 (VIC-labeled) sequence CAGCGCGTTCCTT and probe/reporter 2 (FAM-labeled) sequence CAGCGCATTCCTT. The AURKA haplotypes were determined using the Phe31Ile and val57Ile SNPs as described by Ishikawa and colleagues (14).

Statistical analysis plan

A 2-stage design was used to evaluate alisertib antitumor activity in 7 primary strata: NBL with RECIST measurable disease, NBL with MIBG only evaluable disease, OS, EWS, RMS, ALL, and AML. For each of the 2 NBL strata, 14 patients were enrolled at the first stage. If no patients experienced a complete or partial response, alisertib was considered inactive in that stratum, and further enrollment to that stratum was terminated. If 1 or

Published OnlineFirst February 18, 2019; DOI: 10.1158/1078-0432.CCR-18-2675

more patients achieved an objective response, 10 additional patients were enrolled to the stratum. Alisertib was active if ≥4 of 24 patients in an expanded stratum experienced an objective response. With this design, alisertib was identified as inactive with probability 0.96 if the true response rate was 5% and as active with probability 0.91 if the true response rate was 25%. For the other 5 secondary strata (NRSTS, HBL, GCT, WT, and rhabdoid tumors), at the first stage for each stratum, 10 patients were enrolled. If no patients experienced an objective response, alisertib was considered inactive in that stratum and enrollment was terminated. If 1 or more patients experienced an objective response, 10 additional patients were enrolled to that stratum. Alisertib was considered active if ≥3 of 20 patients in an expanded stratum experienced an objective response. With this design, alisertib would be identified as inactive with probability 0.93 if the true response rate was 5%, and was identified as active with probability 0.88 if the true response rate was 25%. Because of the rarity of tumors in the secondary strata, enrollment to the study was designed to be closed, irrespective of enrollment numbers, when the evaluation of the 7 primary strata was completed. If sufficient enrollment was obtained, the 2-stage design used for the non-NBL stratum was applied to the secondary stratum.

Xenograft studies

Subsequent to the prior preclinical evaluation in which alisertib was administered on a twice-daily schedule for 5 days and repeated for 3 weeks (25), the maximum tolerated dose in pediatric patients was determined to be once daily for 7 days (29). An experiment was designed to compare the preclinical and clinical schedules. ALL xenografts were generated as described previously (26, 35). ALL-8 and ALL-19 xenograft cells, generated from 2 patients with relapsed ALL, were inoculated into NOD/SCID mice and engraftment monitored by weekly flow cytometric enumeration of the percentage of human CD45 (%huCD45) cells in murine peripheral blood (PB). When the %huCD45 cells reached a median of >1% for the entire cohort, mice were randomized to receive treatment with alisertib or vehicle control. Alisertib was administered using 2 alternative schedules: schedule A, twice daily for 7 days; or schedule B, twice daily for 5 days repeated for 3 weeks. In both cases, the dose used was 10.4 mg/kg, administered by oral gavage as a suspension in 10% cyclodextrin. Groups of 4 to 6 mice were euthanized at days 0, 7, and 21 posttreatment initiation and at the end of the evaluation period (day 42) to assess leukemic infiltration of PB, bone marrow, and spleens. An additional experimental endpoint for each mouse was when the %huCD45 cells in PB reached 25% (deemed an event). Mice were euthanized if morbid or if they experienced weight loss ≥20%. EFS, Treated-Control (T-C), T/C, and overall response measure (ORM) estimations were carried out according to established methodology (35). Individual mice were assigned an ORM depending on the leukemic growth characteristics observed in the 42 days following treatment according to the established criteria used for evaluating single agents, and the median ORM was used to obtain the group score.

Results

Patients

Characteristics for all patients are presented in Table 1. All patients ($n = 139$) were eligible. Two patients, 1 with RECIST-measurable NBL and 1 with AML, were not evaluable for response due to rapid progression of disease prior to the start of protocol therapy. The median number of treatment cycles for 137 response-evaluable patients was 2 (range, 1–35). A total of 500 cycles of alisertib were delivered. Three patients completed 35 cycles (24 months) of protocol therapy.

Toxicity

During cycle 1, 18 patients (13%) experienced dose-limiting toxicity including myelosuppression, mucositis, febrile neutropenia, enterocolitis, diarrhea, depression, hypersomnia, photophobia, tumor lysis syndrome, hyperbilirubinemia, and/or electrolyte abnormalities. The frequency of alisertib-related grade 3 and 4 toxicities is shown in Table 2. Alisertib-related grade 3 or 4 toxicities that occurred in ≥10% of delivered cycles ($n = 500$) were anemia (13.6%), lymphopenia (12.2%), neutropenia (51.8%), thrombocytopenia (20.8%), and leukopenia (33%). During cycle 1, 2 patients had fatal adverse events possibly related to alisertib: a patient with pelvic soft tissue sarcoma experienced grade 5 respiratory failure and a patient with hepatoblastoma experienced a fatal hepatic hemorrhage.

Response

Five objective responses were observed. Two patients had complete responses, 1 patient with MIBG-only evaluable NBL (Fig. 1) and 1 patient with WT. Three patients had partial

Table 1. Patient characteristics at enrollment

	Total	NBL measurable	NBL evaluable	AML	ALL	EWS	RMS	NR-STS	OS	WT	HBL	GCT	MRT
Eligible (*N*)	139	25	24	11	10	10	10	10	10	10	8	7	4
Age (years)[a]	10 (2–21)	9 (3–20)	8 (4–19)	10 (3–15)	13 (7–19)	14 (6–20)	12 (4–21)	15 (9–20)	17 (12–21)	10 (3–18)	6 (3–14)	11 (3–19)	5 (2–7)
Female (%)	45%	48%	29%	55%	30%	30%	70%	50%	30%	70%	38%	57%	50%
Race, *n* (%)													
Caucasian	80 (58%)	19 (76%)	15 (63%)	2 (18%)	5 (50%)	8 (80%)	7 (70%)	4 (40%)	7 (70%)	5 (50%)	4 (50%)	4 (57%)	-
Black	21 (15%)	2 (8%)	4 (17%)	2 (18%)	2 (20%)	-	1 (10%)	1 (10%)	2 (20%	2 (20%)	2 (20%)	-	3 (75%)
Other	6 (4%)	0 (0%)	2 (8%)	1 (9%)	-	-	-	-	1 (10%)	-	1 (10%)	1 (14%)	-
Not specified	32 (23%)	4 (16%)	3 (12%)	6 (55%)	3 (30%)	2 (20%)	2 (20%)	5 (50%)		3 (30%)	1 (10%)	2 (29%)	1 (25%)
Ethnicity, *N* (%)													
Hispanic	30 (22%)	3 (12%)	1 (4%)	4 (36%)	2 (20%)	1 (10%)	1 (10%)	3 (30%)	3 (30%)	4 (40%)	3 (38%)	4 (57%)	1 (25%)
Non-Hispanic	97 (70%)	19 (76%)	22 (92%)	5 (46%)	8 (80%)	8 (80%)	7 (70%)	6 (60%)	7 (70%)	5 (50%)	5 (62%)	2 (29%)	3 (75%)
Not reported	12 (8%)	3 (12%)	1 (4%)	2 (18%)	-	1 (10%)	2 (20%)	1 (10%)	-	1 (10%)	-	1 (14%)	-

[a]Median (range).

MRT, malignant rhaboid tumor (2 patients with CNS atypical teratoid rhabdoid tumors and 2 patients with extracranial malignant rhabdoid tumors).

Published OnlineFirst February 18, 2019; DOI: 10.1158/1078-0432.CCR-18-2675

Table 2. Frequency of alisertib-related grade 3 or 4 toxicity in all cycles ($n = 500$)

CTCAE class	Toxicity	Grade 3, *n* (%)	Grade 4, *n* (%)	Grade 5, *n* (%)
Hematologic	Anemia	63 (12.6%)	5 (1%)	
	Febrile neutropenia	18 (3.6%)		
	Lymphopenia	47 (9.4%)	14 (2.8%)	
	Neutropenia	124 (24.8%)	137 (27.4%)	
	Thrombocytopenia	54 (10.8%)	50 (10%)	
	Serum amylase increased	1 (0.2%)		
	Leukopenia	117 (23.4%)	48 (9.6%)	
Eye disorders	Photophobia	1 (0.2%)		
Gastrointestinal	Diarrhea	2 (0.4%)		
	Enterocolitis	1 (0.2%)		
	Oral mucositis	19 (3.8%)		
	Oral pain	5 (1%)		
	Nausea	2 (0.4%)		
	Vomiting	2 (0.4%)		
Investigations (laboratory)	ALT increased	17 (3.4%)		
	AST increased	10 (2%)		
	Hyperbilirubinemia	3 (0.6%)		
	GGT increased	1 (0.2%)		
	INR increased	1 (0.2%)		
Infection	Infection	1 (0.2%)		
	Pneumonia	1 (0.2%)		
	Urinary tract infection	1 (0.2%)		
Metabolism/ nutrition	Anorexia	1 (0.2%)		
	Dehydration	6 (1.2%)		
	Hyperuricemia		1 (0.2%)	
	Hypoalbuminemia	1 (0.2%)		
	Hypocalcemia	1 (0.2%)		
	Hypokalemia	4 (0.8%)	1 (0.2%)	
	Hyponatremia	3 (0.6%)		
	Hypophosphatemia	1 (0.2%)		
	Tumor lysis syndrome		1 (0.2%)	
Psychiatric	Depression		1 (0.2%)	
Neurological	Dizziness	14 (2.8%)		
	Hypersomnia	1 (0.2%)		
Hepatobiliary	Hepatic hemorrhage			1 (0.2%)
Respiratory	Respiratory failure			1 (0.2%)
Skin/ dermatological	Palmar-plantar erythrodysesthesia	2 (0.4%)		

responses, 1 each with RECIST-measurable NBL, MIGB evaluable NBL and HBL. The patient with HBL and partial response was previously reported in the phase I trial (29). Unlike the phase I study (ADVL0812), prolonged stable disease was not considered as a response in this trial and was not centrally reviewed. No responses were achieved in the other primary disease strata (OS, EWS, RMS, ALL, AML). The objective responses are summarized in Table 3 and includes the number of cycles of alisertib administered for patients with a best response of stable disease. Accrual to secondary strata (HBL, WT, GCT, NRSTS) was discontinued due to insufficient response in the primary strata. At the time the study was closed, accrual to the first stage was not completed for HBL, GCT, or rhabdoid tumors.

Pharmacokinetics and pharmacogenomics

Forty-five patients provided consent for optional PK samples on ADVL0912 and 2 patients from ADVL0812 had trough levels obtained on day 4. The alisertib PK parameters were highly variable (Supplementary Table S1). There was no correlation between age or gender and alisertib C_{min} or AUC. The alisertib C_{max} exceeded 1 μmol/L in 98% (44/45) of patients participating in detailed PK studies on day 1. The median C_{min} on day 4 was 1.6 μmol/L, exceeding 1 μmol/L target trough concentration in 67% (26/39) of patients. The median C_{min} on day 7 was 0.9 μmol/L, exceeding 1 μmol/L in 41% of patients (11/27).

A total of 87 patients underwent genotyping for AURKA and UGT1A1 (Table 4). There was no relationship between cycle 1 toxicity and either the Phe31Ile or Val57Ile SNPs in the AURKA genotype or in the AURKA haplotypes (Table 5). There was no relationship between ≥ grade 2 toxicities and AURKA genotype for the Ile31Phe SNP or the AURKA haplotypes. However, patients that were heterozygous (WV) for the Val57Ile SNP appeared to have fewer ≥ grade 2 toxicities (Table 5). Given the small patient cohort, it is not possible to determine if this is clinically significant. In addition, there were no relationships between treatment response and AURKA genotype for either SNP.

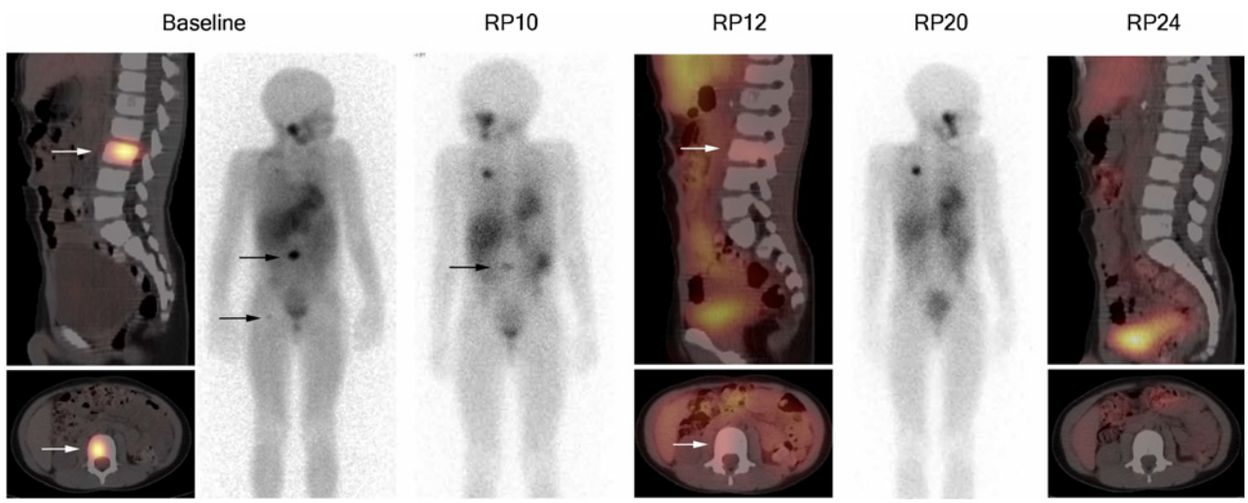

Figure 1.
Sagittal and axial SPECT/CT images from ^{123}I-MIBG examination performed at baseline, reporting periods (RP) 12 and 24; ^{123}I-MIBG anterior projection planar images at baseline, RP10 and RP 20. Each reporting period is 1 cycle. Arrows indicate MIBG-avid NBL in the L3 vertebral body and proximal right femur. The other areas of MIBG positivity on planar imagines include physiologic uptake in liver, salivary glands, renal collecting system, GI tract, and excretion in bladder. Uptake projecting of the thorax at RP 10 and 10 is residual tracer at the port injection site.

Published OnlineFirst February 18, 2019; DOI: 10.1158/1078-0432.CCR-18-2675

Table 3. Responses to alisertib

Stratum	Response evaluable	Responder		Nonresponders		
		Complete response	Partial response	Stable disease, *n* patients, median (range) cycles administered	Nonresponders	Progressive disease
NBL (measurable)	24		1	2 (6, 13 cycles)	5	16
NBL (MIBG evaluable)	24	1	1	9 [13 (5–35) cycles]	4	9
ALL	10			3 (1, 2, 2 cycles)	1	6
AML	10				10	
EWS	10			3 (4, 5, 5 cycles)	2	5
Rhabdo-myosarcoma	10			1 (15 cycles)	2	7
NR-STS	10			1 (5 cycles)	2	7
OS	10				2	8
Wilms tumor	10	1		1 (31 cycles)	1	7
Hepatoblastoma	8		1[a]	1 (5 cycles)	2	4
Germ cell tumor	7			2 (4 and 5 cycles)	1	4
Rhabdoid tumors	4				1	3
Total	137	2	3	23	33	76

[a]Reported previously in ADVL0812 publication (29).

Paired data for UGT1A1 phenotype and day 4 C_{min} was obtained in 32 patients. There did not appear to be a difference in day 4 C_{min} between the intermediate (IM) and poor metabolizer (PM) phenotypes, therefore, the data for these patients were pooled (Table 6). The mean ± SE alisertib trough concentration for the extensive metabolizer group (EM, $n = 16$) was 1.01 ± 0.23 μmol/L (95% CI, 0.53–1.49 μmol/L) and for IM/PM ($n = 16$) was 2.06 ± 0.30 μmol/L (95% CI, 1.42–2.70 μmol/L). The difference in the population means, 1.05 μmol/L (95% CI, 0.28–1.82 μmol/L), was statistically significant (P value of pooled t test, 0.0091). The mean alisertib trough concentration for patients with and without ≥grade 2 toxicities were 1.50 ± 0.25 μmol/L (95% CI, 0.96–2.04 μmol/L) and 1.58 ± 0.35 μmol/L (95% CI, 0.83–2.32 μmol/L), respectively. The difference in the population means, 0.08 μmol/L (95% CI, −0.79–0.94 μmol/L), was not statistically significant. Although alisertib trough concentrations were statistically significantly higher for IM/PM patients, there did not appear to be a relationship with the occurrence of ≥ grade 2 toxicity (P value = 0.86). Furthermore, we found no significant interaction between EM (yes/no) and ≥ grade 2 adverse events (yes/no) (P value = 1.0; Table 6).

Xenograft studies

For the T-lineage ALL-8, based on serial PB parameters, leukemia progression was significantly delayed compared to vehicle control for both of the treatment schedules, resulting in increased EFS (Fig. 2; Table 7). Leukemia progression was delayed by an additional 12.1 days in mice treated with schedule B compared with schedule A ($P = 0.004$), with T/C values 3.6 and 2.2, respectively. However, neither of the treatment schedules induced an objective response. Data are summarized in Table 7 (Supplementary Table S2). Engraftment levels for ALL-8 engrafted mice detected in the 3 compartments analyzed at autopsy (blood, bone marrow, and spleen) are shown in Supplementary Fig. S1 and Figs. 2 and 3, respectively. Treatment with both alisertib schedules limited leukemia progression to a similar extent by day 7 in all organs analyzed. These effects were not complete, with 5% to 10% human cells in the spleen and approximately 20% human cells in the bone marrow as the lowest levels achieved. schedule B was more effective than schedule A in reducing leukemia levels measured at day 21 in the 3 compartments analyzed, but the differences between the treatments were transient and after drug treatments ceased there was a rapid progression of the disease (day 42).

For the B-lineage ALL-19, based on serial PB parameters, leukemia progression was not significantly different from that of controls for schedule A (Fig. 3; Table 7), However, schedule B significantly delayed ALL-19 progression by 24.6 days relative to controls, which was 17.6 days greater than schedule A ($P = 0.048$) with T/C values 4.7 and 2.0, respectively. Furthermore, although treatment on schedule A resulted in progressive disease (PD), schedule B induced a complete response (CR). Data are summarized in Table 7 and Supplementary Table S2. Engraftment levels for ALL-19 engrafted mice detected in the 3 compartments analyzed at autopsy (blood, bone marrow,

Table 4. AURKA, UGT1A1[c]28, and UGT1A1 PBREM genotype distribution

Gene	RsSNP ID	Description	Genotype[a] (WW/WV/VV)
AURKA	rs2273535	91 A>T, Ile31Phe	51/27/8[b]
	rs1047972	169 G>A, Val57Ile	63/21/2[b]
UGT1A1[c]28	rs8175347	TA repeat (5,6,7,8)	39/32/11
UGT1A1 PBREM	rs4124874	-3279 T>G	23/42/22
	rs10929302	-3156 G>A	46/33/8

[a]W, wild-type allele; V, variant allele; for UGT1A1.
[b]One sample could not be genotyped with the available DNA.
Five samples could not be genotyped with the available DNA.
[c]28- WW = 66 or 57, WV = 67 or 68, and VV = 77 or 78.

Table 5. Toxicity versus AURKA genotype

AURKA genotype		Cycle 1 DLT		Grade 2+ toxicity	
		Yes	No	Yes	No
91 A>T	Total	13	73	72	14
	WW	8	43	42	9
	WV	5	22	22	5
	VV	0	8	8	0
169 G>A	Total	13	73	72	14
	WW	10	53	54	9
	WV	3	18	17	4
	VV	0	2	1	1
Haplotype	Total	13	73	72	14
	H1	5	25	26	4
	H2	8	48	46	10

Published OnlineFirst February 18, 2019; DOI: 10.1158/1078-0432.CCR-18-2675

Table 6. Toxicity versus UGT1A1 metabolizer phenotype and alisertib trough concentration (C_{min}) on day 4

	EM		I/E, I, P	
	Grade ≥2: No ($n = 7$)	Grade ≥ 2: Yes ($n = 9$)	Grade ≥ 2; No ($n = 8$)	Grade ≥ 2: Yes ($n = 8$)
Alisertib, C_{min} (μmol/L) mean ± SD	0.81 ± 0.43	1.17 ± 1.15	2.25 ± 1.53	1.87 ± 0.82
Alisertib, C_{min} (μmol/L) median (range)	0.99 (0.2–1.24)	0.53 (0.08–3.51)	2.22 (0.6–5.65)	1.80 (0.97–3.10)

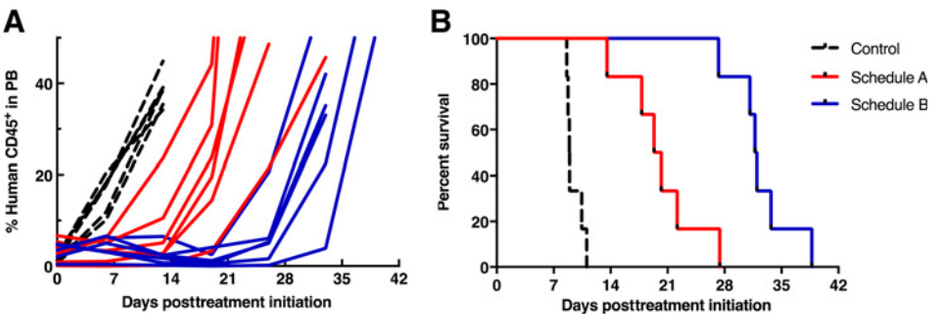

Figure 2.
Percentage of huCD45 cells in PB over time (**A**), and event-free survival curves (**B**) for ALL-8 engrafted NOD/SCID mice treated with alisertib at 10.4 mg/kg twice daily for 7 days (schedule A, red), or twice daily for 5 days repeated for 3 weeks (schedule B, blue) in relation to vehicle-treated controls (dotted line).

and spleen) are shown in Supplementary Fig. S1 and Figs. 2 and 3, respectively. They followed a similar pattern to those of ALL-8, with the exception of being higher at day 7, particularly in the bone marrow.

Discussion

In this phase II study, antitumor activity of alisertib was evaluated in 137 children and adolescents in 7 primary and 5 secondary disease strata. We demonstrated that children achieved target concentrations established in adults and preclinical models. The higher alisertib trough concentration in patients with intermediate and poor metabolizer UGT1A1 phenotypes compared with the extensive metabolizer phenotype was statistically significant. However, we did not find a difference in frequency of toxicity among these groups. Evaluation of AURK somatic mutation status and AAK expression in archival tumor specimens from children enrolled on this study was not performed; therefore, the impact of enrollment stratification by somatic mutation or expression in tumors of children enrolled on this trial is unknown. Pharmacogenomic profiling of germline AURKA in adults has focused on cancer susceptibility and early adverse reactions (14). In this study, we evaluated germline AURKA SNPs and did not find correlation with toxicity during cycle 1 or response.

Despite striking efficacy in pediatric xenograft models in which objective responses were reported in 80% of solid tumor pediatric solid tumor models and all leukemia models (26), the objective response rate in children and adolescents receiving single-agent alisertib on this trial was less than 5%. In patients receiving 50 mg twice a day, the C_{max} and AUC0–24 h were 1.3 and 40 μmol/L h, respectively (25). At the recommended phase II dose of 50 mg twice a day for 7 days, average trough concentrations exceeded 1 μmol/L, the efficacious concentration estimated in previous preclinical work. In mice receiving alisertib at 10 mg/kg, the C_{max} and AUC0–24 h were 16 and 39 μmol/L h, respectively, with the 12 hours level being 1.2 μmol/L (25). These data suggest that continuous drug exposure above 1 μmol/L throughout each 24-hour dosing period which can only be achieved with twice-daily dosing in mice, is crucial for antitumor activity. The initial preclinical studies evaluating alisertib in pediatric patient-derived xenograft models used a dose and schedule that was used in preclinical assessment of adult cancer models in an attempt to mirror pharmacokinetic–pharmacodynamic relationships expected to be tolerated in humans. However, the continuous treatment schedule was too myelosuppressive in adult phase I studies and a 7-day on, 14-day off regimen was adopted to minimize toxicity. Thus, we hypothesized that the dose and schedule used in our trial might account for the discordance between the preclinical and clinical activity observed.

Concurrent with this trial, we tested alisertib in ALL xenograft models utilizing an intermittent dose and schedule, and found significantly less efficacy compared with the previously tested schedule. This demonstrates an important point in the clinical translation of new agents. Although careful consideration is made to maximize the clinical relevance and translatability of preclinical oncology studies, numerous variables can influence the extent to which a drug against a given target will cause toxicity. This study highlights the critical importance of performing reverse translational studies to rigorously reproduce results when dose or

Table 7. Leukemic growth delay summary and clinical scoring for the 2 treatment schedules for xenografts ALL-8 and ALL-19

		Median EFS (days)					
	Treatment schedule[a]	Control	Treated	EFS T-C (days)	EFS T/C	*P* value[b]	Median response score
ALL-8	Schedule A	8.9	19.8	**10.9**	**2.2**	0.002	PD2
	Schedule B	8.9	31.9	**23.0**	**3.6**	0.002	SD
ALL-19	Schedule A	6.6	13.6	7.0	2.0	0.584	PD2
	Schedule B	6.6	31.2	**24.6**	**4.7**	0.002	CR

[a]Schedule A, twice daily × 7 days; schedule B, twice daily × 5 days × 3 weeks.
[b]Statistically significant results.

Published OnlineFirst February 18, 2019; DOI: 10.1158/1078-0432.CCR-18-2675

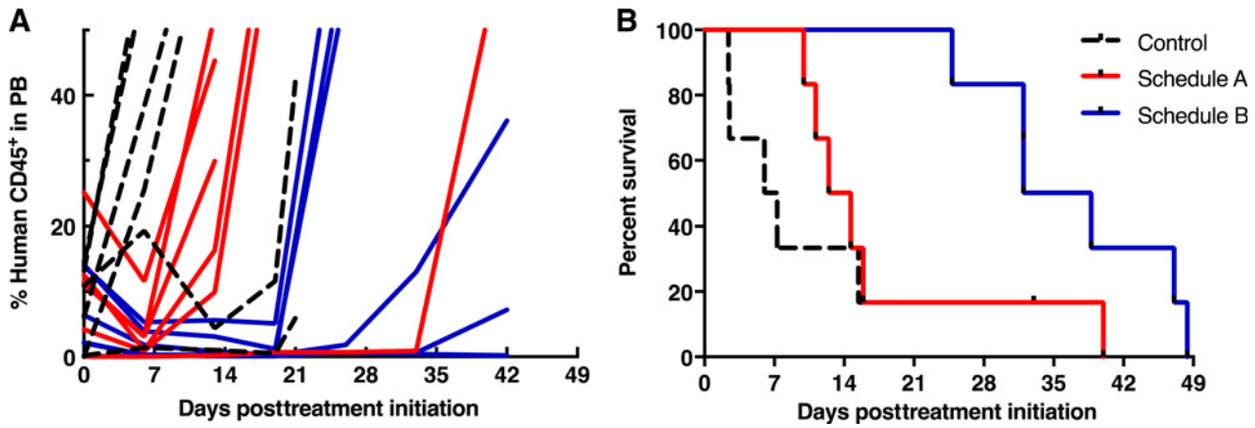

Figure 3.

Percentage of huCD45+ cells in PB over time (**A**), and event-free survival curves (**B**) for ALL-19 engrafted NOD/SCID mice treated with alisertib at 10.4 mg/kg twice daily for 7 days (schedule A, red), or twice daily for 5 days repeated for 3 weeks (schedule B, blue) in relation to vehicle-treated controls (dotted line).

schedule significantly change as a result of early-phase human clinical trials. The continuous treatment schedule of alisertib, which was shown to be more effective in preclinical models than the 1-week administration schedule, was not feasible due to toxicity to pursue in the clinical setting, providing a potential explanation for the differential observed between preclinical and clinical antitumor activity. To increase our confidence in the translation of results from preclinical studies, there needs to be continued efforts to redesign preclinical experiments as we learn from the corresponding human experience.

Identifying applicable preclinical cancer models remains a major challenge in augmenting the effectiveness of drug development and predicting success in the clinic. All models are limited and interrogating the complexity of human cancers in the laboratory remains a challenge that contributes appreciably to attrition in drug development. In recent years, patient-derived xenografts obtained by direct implants of human tumors in immunodeficient mice and then passaged directly from mouse to mouse have emerged as an important platform for translational oncology research (36). The ability of these models to predict clinical outcomes is being optimized through murine humanization strategies to improve the reach of these models as reliable tools for exploring tumor intrinsic and extrinsic heterogeneity, clonal evolution under the selective pressure of our therapies, discovery of integral biomarkers and predictability of drug response in the clinic (36). Numerous challenges and limitations remain, including the lack of a proper anatomical and metastatic niche, engraftment failure of certain tumor subtypes, access to imaging technologies for robust tumor visualization, and hurdles to achieve complete human immune system reconstitution (36).

To improve the efficiency of this 2-stage phase II trial, this trial was prospectively designed to include data from 21 patients on the phase I single-agent alisertib trial (ADVL0812), who were treated at the recommended dose and met eligibility criteria for this trial, including 5 patients with NBL-measurable disease, 12 patients with NBL-MIGB evaluable disease, 2 patients with non-RMS soft tissue sarcoma, and 2 patients with hepatoblastoma. Given the rarity of relapsed pediatric cancers, trial designs that improve efficiency are essential.

Clinical trials evaluating alisertib in combination with cytotoxic agents have shown antitumor activity in children and adolescents with relapsed solid tumors including NBL (37, 38). Given the lack of objective response observed in this comprehensive single-agent clinical trial of alisertib as well as dose-limiting myelosuppression, alternative strategies, including novel:novel combinations simultaneously targeting other oncogenic signaling pathways or exploiting the pro-apoptotic machinery (39), should be explored to harness this pathway and simultaneously minimize toxicity.

Disclosure of Potential Conflicts of Interest

D.T. Teachey is a consultant/advisory board member for Janssen. No potential conflicts of interest were disclosed by the other authors.

Authors' Contributions

Conception and design: Y.P. Mossé, E. Fox, D.T. Teachey, R.B. Lock, M. Krailo, S.L. Berg, S.M. Blaney, B.J. Weigel

Development of methodology: Y.P. Mossé, E. Fox, J.M. Reid

Acquisition of data (provided animals, acquired and managed patients, provided facilities, etc.): Y.P. Mossé, E. Fox, D.T. Teachey, J.M. Reid, S.L. Safgren, H. Carol, P.J. Houghton, M. Krailo, S.D. Voss, B.J. Weigel

Analysis and interpretation of data (e.g., statistical analysis, biostatistics, computational analysis): Y.P. Mossé, E. Fox, D.T. Teachey, J.M. Reid, H. Carol, R.B. Lock, P.J. Houghton, M.A. Smith, D. Hall, D.A. Barkauskas, M. Krailo, S.D. Voss, S.M. Blaney, B.J. Weigel

Writing, review, and/or revision of the manuscript: Y.P. Mossé, E. Fox, D.T. Teachey, J.M. Reid, S.L. Safgren, H. Carol, R.B. Lock, P.J. Houghton, M.A. Smith, D. Hall, D.A. Barkauskas, M. Krailo, S.D. Voss, S.L. Berg, S.M. Blaney, B.J. Weigel

Administrative, technical, or material support (i.e., reporting or organizing data, constructing databases): D.T. Teachey, J.M. Reid, D. Hall

Study supervision: Y.P. Mossé, M. Krailo, B.J. Weigel

Acknowledgments

This work was funded by National Institutes of Health National Cancer Institute (NIH/NCI) National Clinical Trials Network (NCTN) Operations Center grant U10CA180886 and Statistics and Data Center grant U10CA180899 to the Children's Oncology Group; National Institutes of Health National Cancer Institute NOI-CM-42216 and NOI-CM-91001-03; St Baldrick's Foundation; and Cookies for Kids Foundation.

The costs of publication of this article were defrayed in part by the payment of page charges. This article must therefore be hereby marked *advertisement* in accordance with 18 U.S.C. Section 1734 solely to indicate this fact.

Received November 26, 2018; revised December 20, 2018; accepted February 14, 2019; published first February 18, 2019.

Published OnlineFirst February 18, 2019; DOI: 10.1158/1078-0432.CCR-18-2675

References

1. Gautschi O, Heighway J, Mack PC, Purnell PR, Lara PN Jr, Gandara DR. Aurora kinases as anticancer drug targets. Clin Cancer Res 2008;14: 1639–48.
2. Nigg EA. Mitotic kinases as regulators of cell division and its checkpoints. Nat Rev Mol Cell Biol 2001;2:21–32.
3. Lapenna S, Giordano A. Cell cycle kinases as therapeutic targets for cancer. Nat Rev Drug Discov 2009;8:547–66.
4. Sen S, Zhou H, Zhang RD, Yoon DS, Vakar-Lopez F, Ito S, et al. Amplification/overexpression of a mitotic kinase gene in human bladder cancer. J Nat Cancer Inst 2002;94:1320–9.
5. Borges KS, Moreno DA, Martinelli CE Jr., Antonini SR, de Castro M, Tucci S Jr, et al. Spindle assembly checkpoint gene expression in childhood adrenocortical tumors (ACT): overexpression of Aurora kinases A and B is associated with a poor prognosis. Pediatr Blood Cancer 2013;60: 1809–16.
6. Hartsink-Segers SA, Zwaan CM, Exalto C, Luijendijk MWJ, Calvert VS, Petricoin EF, et al. Aurora kinases in childhood acute leukemia: the promise of aurora B as therapeutic target. Leukemia 2013;27:560–8.
7. Venkataraman S, Alimova I, Tello T, Harris PS, Knipstein JA, Donson AM, et al. Targeting Aurora Kinase A enhances radiation sensitivity of atypical teratoid rhabdoid tumor cells. J Neuro-Oncol 2012;107:517–26.
8. Romain C, Paul P, Kim KW, Lee S, Qiao J, Chung DH. Targeting Aurora kinase-A downregulates cell proliferation and angiogenesis in neuroblastoma. J Pediatr Surg 2014;49:159–65.
9. Ewart-Toland A, Briassouli P, de Koning JP, Mao JH, Yuan J, Chan F, et al. Identification of Stk6/STK15 as a candidate low-penetrance tumor-susceptibility gene in mouse and human. Nature genetics 2003;34: 403–12.
10. Dicioccio RA, Song H, Waterfall C, Kimura MT, Nagase H, McGuire V, et al. STK15 polymorphisms and association with risk of invasive ovarian cancer. Cancer Epidemiol Biomarkers Prev 2004;13:1589–94.
11. Egan KM, Newcomb PA, Ambrosone CB, Trentham-Dietz A, Titus-Ernstoff L, Hampton JM, et al. STK15 polymorphism and breast cancer risk in a population-based study. Carcinogenesis 2004;25:2149–53.
12. Tang W, Qiu H, Jiang H, Wang L, Sun B, Gu H. Aurora-A V57I (rs1047972) polymorphism and cancer susceptibility: a meta-analysis involving 27,269 subjects. PLoS One 2014;9:e90328.
13. Xu L, Zhou X, Jiang F, Xu L, Yin R. STK15 rs2273535 polymorphism and cancer risk: a meta-analysis of 74,896 subjects. Cancer Epidemiol 2014;38: 111–7.
14. Ishikawa A, Suga T, Shoji Y, Kato S, Ohno T, Ishikawa H, et al. Genetic variants of NPAT-ATM and AURKA are associated with an early adverse reaction in the gastrointestinal tract of patients with cervical cancer treated with pelvic radiation therapy. Int J Radiat Oncol Biol Phys 2011;81: 1144–52.
15. Barr PM, Li H, Spier C, Mahadevan D, LeBlanc M, Ul Haq M, et al. Phase II intergroup trial of alisertib in relapsed and refractory peripheral T-cell lymphoma and transformed mycosis fungoides: SWOG 1108. J Clin Oncol 2015;33:2399–404.
16. Melichar B, Adenis A, Lockhart AC, Bennouna J, Dees EC, Kayaleh O, et al. Safety and activity of alisertib, an investigational aurora kinase A inhibitor, in patients with breast cancer, small-cell lung cancer, non-small-cell lung cancer, head and neck squamous-cell carcinoma, and gastro-oesophageal adenocarcinoma: a five-arm phase 2 study. Lancet Oncol 2015;16: 395–405.
17. Friedberg JW, Mahadevan D, Cebula E, Persky D, Lossos I, Agarwal AB, et al. Phase II study of alisertib, a selective Aurora A kinase inhibitor, in relapsed and refractory aggressive B- and T-cell non-Hodgkin lymphomas. J Clin Oncol 2014;32:44–50.
18. Manfredi MG, Ecsedy JA, Chakravarty A, Silverman L, Zhang M, Hoar KM, et al. Characterization of Alisertib (MLN8237), an investigational small-molecule inhibitor of aurora A kinase using novel in vivo pharmacodynamic assays. Clin Cancer Res 2011;17:7614–24.
19. Yang JJ, Li Y, Chakravarty A, Lu C, Xia CQ, Chen S, et al. Preclinical drug metabolism and pharmacokinetics, and prediction of human pharmacokinetics and efficacious dose of the investigational Aurora A kinase inhibitor alisertib (MLN8237). Drug Metab Lett 2014;7: 96–104.
20. Cervantes A, Elez E, Roda D, Ecsedy J, Macarulla T, Venkatakrishnan K, et al. Phase I pharmacokinetic/pharmacodynamic study of MLN8237, an investigational, oral, selective aurora a kinase inhibitor, in patients with advanced solid tumors. Clin Cancer Res 2012; 18:4764–74.
21. Falchook G, Kurzrock R, Gouw L, Hong D, McGregor KA, Zhou X, et al. Investigational Aurora A kinase inhibitor alisertib (MLN8237) as an enteric-coated tablet formulation in non-hematologic malignancies: phase 1 dose-escalation study. Invest New Drugs 2014;32:1181–7.
22. Venkatakrishnan K, Zhou X, Ecsedy J, Mould DR, Liu H, Danaee H, et al. Dose selection for the investigational anticancer agent alisertib (MLN8237): Pharmacokinetics, pharmacodynamics, and exposure-safety relationships. J Clin Pharmacol 2015;55:336–47.
23. Matulonis UA, Sharma S, Ghamande S, Gordon MS, Del Prete SA, Ray-Coquard I, et al. Phase II study of MLN8237 (alisertib), an investigational Aurora A kinase inhibitor, in patients with platinum-resistant or refractory epithelial ovarian, fallopian tube, or primary peritoneal carcinoma. Gynecologic Oncology 2012;127:63–9.
24. Goldberg SL, Fenaux P, Craig MD, Gyan E, Lister J, Kassis J, et al. An exploratory phase 2 study of investigational Aurora A kinase inhibitor alisertib (MLN8237) in acute myelogenous leukemia and myelodysplastic syndromes. Leukemia Res Rep 2014;3:58–61.
25. Carol H, Boehm I, Reynolds CP, Kang MH, Maris JM, Morton CL, et al. Efficacy and pharmacokinetic/pharmacodynamic evaluation of the Aurora kinase A inhibitor MLN8237 against preclinical models of pediatric cancer. Cancer Chemother Pharmacol 2011; 68:1291–304.
26. Maris JM, Morton CL, Gorlick R, Kolb EA, Lock R, Carol H, et al. Initial testing of the aurora kinase a inhibitor MLN8237 by the pediatric preclinical testing program (PPTP). Pediatr Blood Cancer 2010;55: 26–34.
27. Michaelis M, Selt F, Rothweiler F, Löschmann N, Nüsse B, Dirks WG, et al. Aurora kinases as targets in drug-resistant neuroblastoma cells. PloS One 2014;9:e108758.
28. Brockmann M, Poon E, Berry T, Carstensen A, Deubzer HE, Rycak L, et al. Small molecule inhibitors of aurora-a induce proteasomal degradation of N-myc in childhood neuroblastoma. Cancer Cell 2013;24: 75–89.
29. Mosse YP, Lipsitz E, Fox E, Teachey DT, Maris JM, Weigel B, et al. Pediatric phase I trial and pharmacokinetic study of MLN8237, an investigational oral selective small-molecule inhibitor of Aurora kinase A: a children's oncology group phase I consortium study. Clin Cancer Res 2012;18: 6058–64.
30. Eisenhauer EA, Therasse P, Bogaerts J, Schwartz LH, Sargent D, Ford R, et al. New response evaluation criteria in solid tumours: revised RECIST guideline (version 1.1). Eur J Cancer 2009;45:228–47.
31. Ady N, Zucker JM, Asselain B, Edeline V, Bonnin F, Michon J, et al. A new 123I-MIBG whole body scan scoring method–application to the prediction of the response of metastases to induction chemotherapy in stage IV neuroblastoma. Eur J Cancer 1995;31A:256–61.
32. Cheson BD, Bennett JM, Kopecky KJ, Büchner T, Willman CL, Estey EH, et al. Revised recommendations of the international working group for diagnosis, standardization of response criteria, treatment outcomes, and reporting standards for therapeutic trials in acute myeloid leukemia. J Clin Oncol 2003;21:4642–9.
33. Lipsitz E, Moorthy G, Mosse Y, Fox E, Adamson PC. A sensitive and selective liquid chromatography/tandem mass spectrometry method for determination of MLN8237 in human plasma. J Chromatogr B Analyt Technol Biomed Life Sci 2010;878:2369–73.
34. Akaba K, Kimura T, Sasaki A, Tanabe S, Ikegami T, Hashimoto M, et al. Neonatal hyperbilirubinemia and mutation of the bilirubin uridine diphosphate-glucuronosyltransferase gene: a common missense mutation among Japanese, Koreans and Chinese. Biochem Mol Biol Int 1998;46: 21–6.
35. Houghton PJ, Morton CL, Tucker C, Payne D, Favours E, Cole C, et al. The pediatric preclinical testing program: description of models and early testing results. Pediatr Blood Cancer 2007;49:928–40.
36. Byrne AT, Alferez DG, Amant F, Annibali D, Arribas J, Biankin AV, et al. Interrogating open issues in cancer precision medicine with patient-derived xenografts. Nat Rev Cancer 2017;17:254–268.
37. DuBois SG, Marachelian A, Fox E, Kudgus RA, Reid JM, Groshen S, et al. Phase I study of the aurora a kinase inhibitor alisertib

Published OnlineFirst February 18, 2019; DOI: 10.1158/1078-0432.CCR-18-2675

in combination with irinotecan and temozolomide for patients with relapsed or refractory neuroblastoma: a NANT (New Approaches to Neuroblastoma Therapy) trial. J Clin Oncol 2016; 34:1368–75.
38. Dubois SG, Mosse YP, Fox E, Kudgus RA, Reid JM, McGovern R, et al. Phase 2 trial of alisertib in combination with irinotecan and temozolomide for patients with relapsed or refractory neuroblastoma. Clin Cancer Res 2018; 24:6142–9.
39. Ham J, Costa C, Sano R, Lochmann TL, Sennott EM, Patel NU, et al. Exploitation of the apoptosis-primed state of MYCN-amplified neuroblastoma to develop a potent and specific targeted therapy combination. Cancer Cell 2016;29:159–72.

Published OnlineFirst February 18, 2019; DOI: 10.1158/1078-0432.CCR-18-2675

Clinical Cancer Research

A Phase II Study of Alisertib in Children with Recurrent/Refractory Solid Tumors or Leukemia: Children's Oncology Group Phase I and Pilot Consortium (ADVL0921)

Yael P. Mossé, Elizabeth Fox, David T. Teachey, et al.

Clin Cancer Res 2019;25:3229-3238. Published OnlineFirst February 18, 2019.