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Safety and Efficacy of Tinostamustine in a Subpopulation of Patients With Relapsed/Refractory Hodgkin Lymphoma From a Phase I Trial

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ABSTRACT

A significant unmet need remains for patients with Hodgkin lymphoma (HL) who fail to respond to first-line treatment or experience an early relapse. Tinostamustine, a novel alkylating deacetylase inhibitor, inhibits tumor cell growth and slows disease progression in models of hematological malignancies and solid tumors. This was a Phase I, multicenter, open-label, two-stage trial investigating the safety and efficacy of tinostamustine in patients ≥ 18 years with relapsed/refractory (R/R) hematological malignancies, including HL. Stage 1 involved dose-escalation to determine the maximum tolerated dose (MTD) of tinostamustine, optimal infusion time and recommended Phase II dose (RP2D). Stage 2 confirmed the safety and efficacy of the RP2D in expansion cohorts of selected R/R hematological malignancies. Ten patients with heavily pre-treated HL entered dose-escalation, with nine patients experiencing treatment-emergent adverse events (TEAEs) considered to be related to study treatment—primarily hematological toxicities. MTD was 100 mg/m^2 tinostamustine over 60 min and signals of efficacy were observed for patients with HL. In Stage 2, all 20 patients with HL experienced ≥ 1 TEAE, which were principally hematological or gastrointestinal. There were no tinostamustine-related deaths in either stage of the study. Overall response rate in Stage 2 was 37% (2 complete responses, 5 partial responses; 95% confidence interval [CI]: 16%, 62%) and median progression-free survival 3.8 months (95% CI: 2.2–9.4 months). Tinostamustine is a promising new therapeutic approach for the treatment of patients with R/R classical HL with limited options. This study demonstrates a predictable and manageable safety profile with signals of efficacy.

Trial Registration: ClinicalTrials.gov identifier: NCT02576496

Abbreviations: AE, adverse event; ASCT, autologous stem cell transplantation; CBR, clinical benefit rate; CI, confidence interval; C_{max}, maximum plasma concentration; CPI, checkpoint inhibitor; CR, complete response; CTCAE, Common Terminology Criteria for Adverse Events; DLT, dose-limiting toxicity; ECG, electrocardiogram; HDAC, histone deacetylase; HL, Hodgkin lymphoma; IV, intravenously; MAD, maximum administered dose; MTD, maximum tolerated dose; NE, not estimable; ORR, overall response rate; OS, overall survival; PD, disease progression; PD-1, programmed cell-death protein 1; PFS, progression-free survival; PK, pharmacokinetic; PR, partial response; QTcF, corrected QT using the Fredericia formula; R/R, relapsed/refractory; RP2D, recommended Phase II dose; SAE, serious adverse event; SD, stable disease; TEAE, treatment-emergent adverse event.

1 | Introduction

The treatment of Hodgkin lymphoma (HL) has significantly improved over time with the introduction of antibody-based immunotherapies such as brentuximab vedotin and immune checkpoint inhibitors (CPIs) [1, 2]. However, outcomes differ significantly for patients who relapse following a durable response to first-line therapy compared with those with refractory and early relapsing disease. Approximately 20%–30% of patients with advanced-stage HL will ultimately experience disease recurrence, with 10%–15% of cases refractory to first-line therapy even when treated with highly active combination regimens such as brentuximab vedotin plus adriamycin, vinblastine, and dacarbazine [3, 4]. In addition, approximately one-half of these patients will experience disease progression following the current standard of care with salvage chemotherapy and autologous stem cell transplantation (ASCT) [5]. Moreover, although single-agent brentuximab vedotin and programmed cell-death protein 1 (PD-1) blockade led to a significant improvement in overall survival for patients who fail ASCT, only a few patients can be cured with these approaches [5, 6]. It should also be noted that the routine application of ASCT has limitations, including eligibility criteria, poor hematopoietic cell collection, comorbidities, patient risk factors, and inadequate pre-transplant disease control, that can affect outcomes [5]. Therefore, for patients who progress or relapse after ASCT, and for those ineligible to the procedure, the prognosis is poor [5, 7–9]. There remains, therefore, a need for more effective therapies which can offer long-term disease control with a manageable safety profile for the population of patients with HL who are heavily pre-treated having experienced multiple relapses, including ASCT failure, and those ineligible for ASCT, or with refractory disease [5, 9].

Tinostamustine is a novel alkylating deacetylase inhibitor combining bifunctional alkylating activity and high-affinity pan-histone deacetylase (HDAC) inhibition, which improves alkylator access to cancer cell DNA resulting in increased cross-linking as well as reduced DNA repair [10–13]. In non-clinical studies, single-agent tinostamustine has shown significant antitumor activity, and slowed disease progression in both in vitro cell line experiments and animal models of myeloid and lymphoid malignancies, including HL, and solid tumors [10, 12, 14].

Here we report safety and efficacy findings from those patients with relapsed/refractory (R/R) HL enrolled into either the dose-escalation or cohort-expansion stage of the Phase I first-in-human study for tinostamustine in hematological malignancies (NCT02576496).

2 | Materials and Methods

2.1 | Study Design

This was a first-in-human, Phase I, multicenter, open-label, two-stage trial to investigate the safety, pharmacokinetic (PK) profile, and efficacy of tinostamustine in R/R hematological malignancies, including those with R/R HL for whom there were no available therapies.

Overall, 46 patients with R/R hematological malignancies were enrolled in Stage 1, 10 of whom had R/R HL. A total of 48 patients were enrolled in Stage 2 of the study, 20 of whom had R/R HL. Further details on the study design are given in Supporting Information S1. Here we report the findings from both the dose-escalation and cohort-expansion stages of the study for the subset of patients with R/R HL only.

2.2 | Stage 1: Dose Escalation

2.2.1 | Patients

Patients were ≥ 18 years with a life expectancy > 3 months, Eastern Cooperative Oncology Group performance status ≤ 2, and a diagnosis of R/R lymphoid malignancy, including HL, for which there were no available therapies.

Exclusion criteria included any central nervous system involvement, relapse within 100 days of an allogeneic or autologous bone marrow transplant, history of another malignancy within the previous 3 years, QTc interval (Fridericia's formula [QTcF]) > 450 msec at baseline, receipt of drugs known to prolong the QT/QTc interval, any serious medical condition that may interfere with adherence to trial procedures, active infections, Stage III/IV congestive heart failure, defined arrhythmias, steroid treatment, receipt of valproic acid, pregnancy, and breast feeding.

2.2.2 | Objectives

The primary objective of Stage 1 of this study was to determine the safety, tolerability, maximum tolerated dose (MTD), and PK profile of tinostamustine monotherapy. Secondary objectives included establishing the PK profile and the recommended Phase II dose (RP2D) for tinostamustine. These determinations were performed on the whole Stage 1 study population, with the findings reported here being for the subset of patients with HL within that population.

2.2.3 | Endpoints

In Stage 1, the MTD, maximum administered dose (MAD), and dose-limiting toxicities (DLTs) were determined. Definitions applied for these terms are provided in Supporting Information S1. Because of the recognized potential for very high doses of tinostamustine to cause QTc prolongation, digital 24-h 12-lead electrocardiogram (ECG) monitoring was applied in Cycle 1, alongside PK assessments to perform a tinostamustine concentration QTc analysis.

The infusion time was set at 1 h based on non-clinical findings, with the RP2D subsequently investigated at shorter infusion times. Due to increases in maximum plasma concentration (C_{max}) and toxicities at infusion times of 45 and 30 min, 1 h was established as the optimal infusion time.

2.3 | Stage 2: Expansion Cohort

2.3.1 | Patients

Inclusion and exclusion criteria were similar to those for Stage 1 of the study. The first five patients entering Cohort 2 were required to have ≥ 75,000/μL platelets, while the remaining nine patients in Cohort 2 and all six patients in Cohort 2a were required to have platelet levels ≥ 100,000/μL and no supportive treatments to improve hematologic status for 2 weeks before enrollment. In addition, to be eligible for recruitment to the HL cohorts, patients must have received at least three (Cohort 2) or two (Cohort 2a) lines of prior therapy with no other therapy with proven clinical benefit available.

2.3.2 | Objectives

The primary objectives for the study were to examine the overall response rate (ORR) and clinical benefit rate (CBR), together with the safety of the selected RP2D in selected lymphoma subtypes including HL. Secondary objectives included evaluation of time to ORR and duration of response, progression-free survival (PFS), and overall survival (OS).

2.3.3 | Endpoints

In Stage 2, ORR, CBR, PFS, and OS were assessed. Following completion of the initial HL expansion cohort, the decision was taken to extend the study further by recruitment of an additional expansion cohort; however, this second cohort was closed prematurely due to slow recruitment following the enrollment of six patients. Efficacy data are summarized in this manuscript using descriptive statistics from all patients enrolled in the two HL cohorts.

Adverse events (AEs) were assessed using the US National Cancer Institute's Common Terminology Criteria for Adverse Events (CTCAE; version 4.03) for all treated patients, with monitoring and recording of AEs conducted from the provision of patient consent to the point of discontinuation of tinostamustine. QTc prolongations were assessed according to CTCAE version 5. AEs recorded included treatment-emergent adverse events (TEAEs), AEs leading to death, serious adverse events (SAEs), and AEs resulting in trial discontinuation. In the event of a clinically significant laboratory toxicity ≥ Grade 2, more frequent laboratory tests were performed until resolution to Grade 1 or stabilization.

2.4 | Ethical Approval

The protocol, protocol amendments and the study informed consent form were reviewed and approved by the Institutional Review Board/Independent Ethics Committee prior to implementation at any given investigative center. This study was conducted in accordance with the Declaration of Helsinki principles and was consistent with the International Council for Harmonization of Technical Requirements for Pharmaceuticals for Human Use's Good Clinical Practice guidelines, applicable in the United States and the European Union regulatory requirements, and sponsor policies. Written informed consent was obtained from each participant prior to entry into the study.

2.5 | Statistical Analyses

Summaries of continuous and ordinal variables included number (n), mean, standard deviation, median, minimum, and maximum. Summaries for discrete variables included frequency counts and percentages. Two-sided 95% confidence intervals (CIs) were reported for ORR and CBR. PFS and OS were estimated using the Kaplan–Meier method. The estimated survival probabilities are presented via Kaplan–Meier curves with median survival reported including 95% CI.

3 | Results

3.1 | Stage 1: Dose-Escalation Cohort

3.1.1 | Summary of the Sub-Population of Patients With HL

A total of 10 patients with heavily pre-treated HL were enrolled from Italy ($n = 6$), the USA ($n = 3$), and Switzerland ($n = 1$), and formed the safety population, with all 10 patients receiving at least one dose of tinostamustine. Patient demographics and baseline characteristics are presented in Table 1.

3.1.2 | Safety

All 10 patients in the safety population experienced at least one TEAE, with nine patients experiencing TEAEs considered to be related to study treatment (Table 2). The majority of these were hematological toxicities, including thrombocytopenia ($n = 7$), anemia ($n = 7$), lymphopenia ($n = 3$), neutropenia ($n = 3$), leukopenia ($n = 2$), and febrile neutropenia ($n = 1$); five patients experienced nausea that was considered to be related to tinostamustine. No dose-modifying events were reported, and no tinostamustine-related renal or hepatic toxicity was observed at the studied doses.

Two patients (20%) experienced at least one serious TEAE, both of which were considered to be related to tinostamustine treatment. One patient (10%) developed Grade 3 thrombocytopenia and the second Grade 3 febrile neutropenia. Eight of the 10 patients (80%) experienced Grade 3 TEAEs (anemia, thrombocytopenia, lymphopenia, neutropenia, leukopenia, and febrile neutropenia) and four patients (40%) experienced Grade 4 TEAEs (thrombocytopenia, lymphopenia and neutropenia) (Table 3). Five patients (50%) discontinued treatment due to TEAEs, including thrombocytopenia ($n = 4$) and febrile neutropenia ($n = 1$), and three due to progressive disease. There were no deaths related to tinostamustine treatment.

Two DLTs occurred in the 120 mg/m^2 over 60 min cohort: one event of Grade 4 thrombocytopenia ≥ 7 days ($n = 1$), and one of prolonged thrombocytopenia/toxicity resulting in the delay of

TABLE 1 | Patient demographics and baseline characteristics (dose-escalation, safety population).

	Patients with HL receiving tinostamustine[a] ($n = 10$)
Mean ± standard deviation age (range), years	49.4 ± 18.0 (21–74)
Male, *n* (%)	3 (30)
Race, *n* (%)	
White	10 (100)
Median (range) time since initial diagnosis, months	37.7 (21.2–126.9)
Median (range) time since most recent R/R diagnosis, months	2.5 (0.8–26.4)
ECOG PS, *n* (%)	
0	4 (40)
1	6 (60)
Primary refractory disease, *n* (%)	6 (60)
Relapsed disease, *n* (%)	4 (40)
Previous lines of therapy, median (range)	4.5 (2–7)
Prior therapies, *n* (%)	
ABVD/BEACOPP-like chemotherapy	10 (100)
Brentuximab vedotin	9 (90)
Bendamustine	6 (60)
Checkpoint inhibitors	6 (60)
Stem cell transplantation	3 (30)
CHOP/CHOP-like chemotherapy	1 (10)
CVP/CVP-like chemotherapy	1 (10)
HDAC inhibitors	1 (10)
Immunomodulators	1 (10)
Proteasome inhibitors	1 (10)
Steroids	1 (10)
B symptoms, *n* (%)	2 (20)
Night sweats	1 (10)
Unexplained fever	1 (10)

Abbreviations: ABVD, adriamycin (doxorubicin), bleomycin, vinblastine, and dacarbazine; BEACOPP, bleomycin, etoposide, adriamycin, cyclophosphamide, vincristine, procarbazine, and prednisone; CHOP, cyclophosphamide, doxorubicin, vincristine, and prednisone; CVP, cyclophosphamide, vincristine, and prednisone; ECOG PS, Eastern Cooperative Oncology Group—performance status; HDAC, histone deacetylase; HL, Hodgkin lymphoma; R/R, relapsed/refractory.
[a]Patients with HL in the dose-escalation cohort were treated with the following doses of tinostamustine: 40 mg/m^2, 1 patient; 60 mg/m^2, 2 patients, 80 mg/m^2, 3 patients and 120 mg/m^2, 4 patients.

TABLE 2 | Number (%) of patients with TEAEs in the sub-population of patients with HL (dose-escalation, safety population).

	Patients with HL receiving tinostamustine ($n = 10$)
Any TEAE, *n* (%)	10 (100)
≥ 1 tinostamustine-related TEAE, *n* (%)	9 (90)
≥ 1 tinostamustine-related serious TEAE, *n* (%)	2 (20)
Thrombocytopenia	1 (10)
Febrile neutropenia	1 (10)
Permanent withdrawals due to TEAEs, *n* (%)	5 (50)
Deaths due to TEAEs, *n* (%)	0

Abbreviations: HL, Hodgkin lymphoma; TEAE, treatment-emergent adverse event.

TABLE 3 | Number (%) of patients with TEAEs by grade (dose-escalation, safety population).

	Patients with HL receiving tinostamustine ($n = 10$)					
	All grades	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**	**Grade 5**
Any TEAE, *n* (%)	10 (100)	9 (90)	9 (90)	8 (80)	4 (40)	0
Most common TEAEs, *n* (%)						
Hematological toxicities						
Anemia	7 (70)	0	5 (50)	4 (40)	0	0
Thrombocytopenia	7 (70)	2 (20)	6 (60)	3 (30)	3 (30)	0
Lymphopenia	3 (30)	0	0	3 (30)	2 (20)	0
Neutropenia	3 (30)	0	2 (20)	3 (30)	1 (10)	0
Leukopenia	2 (20)	0	1 (10)	1 (10)	0	0
Febrile neutropenia	1 (10)	0	0	1 (10)	0	0
Gastrointestinal toxicities						
Nausea	5 (50)	4 (40)	1 (10)	0	0	0
Vomiting	0	0	0	0	0	0
General disorders						
Pyrexia	3 (30)	3 (30)	0	0	0	0
Asthenia	3 (30)	2 (20)	1 (10)	0	0	0
Injury and procedural complications						
Infusion-related reaction	1 (10)	1 (10)	0	0	0	0
Infusion site erythema	0	0	0	0	0	0
Infusion site pain	0	0	0	0	0	0
Investigations						
QTcF prolongation	0	0	0	0	0	0

Abbreviations: HL, Hodgkin lymphoma; QTcF, corrected QT using the Fredericia formula; TEAE, treatment-emergent adverse event.

the next dose ($n = 1$). No patients in this cohort experienced Grade 3 or higher QTcF prolongation.

3.1.3 | Efficacy

Signals of efficacy were observed for patients with HL who received tinostamustine with an ORR of 60% (95% CI: 26%, 88%) and a CBR of 80% (95% CI: 44%, 97%); these included one complete response (CR), five partial responses (PRs), and two patients with stable disease (SD) following a median (range) number of cycles of tinostamustine of 6 (6–6), 5 (4–10), and 6 (6–6), respectively. Only 2 patients did not show any response to treatment, with one experiencing disease progression (PD) and one death due to PD (Figure 1). Mean ± standard deviation duration of response was 4.9 ± 3.0 months (range: 2.1–9.5 months) and mean ± standard deviation duration of SD was 3.9 ± 3.1 months (range: 1.7–6.1 months). Median PFS was 9.9 months (95% CI: 0.9 months, not estimable [NE]) and median OS was NE (95% CI: 7.8 months, NE).

Of the 10 patients treated with tinostamustine in the dose-escalation stage of the trial, nine (90%) had received brentuximab vedotin and six (60%) a CPI in a previous line of therapy. The ORR in the subset of patients who had received priory therapy with a CPI was 67% (95% CI: 22%, 96%), with one CR and three PRs achieved. Additionally, one patient achieved SD resulting in a CBR of 83% (95% CI: 36%, 100%). Mean ± standard deviation duration of response in these patients was 4.9 ± 3.2 months (range: 2.5–9.5 months) and duration of SD was 1.7 months (range: 1.7–1.7 months).

3.1.4 | Determination of RP2D

The MTD was determined to be 100 mg/m^2 tinostamustine with an infusion time of 60 min. The initial tinostamustine RP2D depended on platelet count at treatment initiation: 100 mg/m^2 intravenously (IV) over 60 min ($\geq 200 \times 10^9$/L platelets), 80 mg/m^2 IV over 60 min ($< 200 \times 10^9$/L, $> 100 \times 10^9$/L platelets), and 50 mg/m^2 IV over 60 min ($\leq 100 \times 10^9$/L platelets). Subsequently, the RP2D was defined as 100 mg/m^2 tinostamustine over 60 min for all patients with $\geq 100 \times 10^9$/L platelets, and the enrollment criteria for patients entering the expansion cohort were revised.

3.2 | Stage 2: Expansion Cohorts

3.2.1 | Summary of the Sub-Population of Patients With HL

A total of 20 patients with R/R HL were recruited from Italy ($n = 7$), France ($n = 5$), Spain ($n = 6$), and the Netherlands

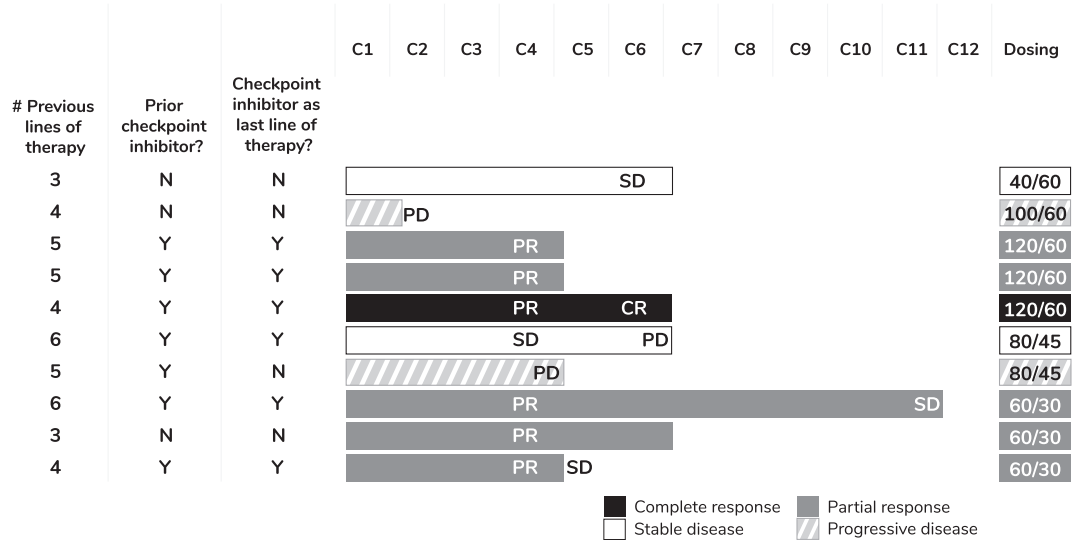

FIGURE 1 | Best overall response to tinostamustine in patients with HL (dose-escalation population). CR, complete response; HL, Hodgkin lymphoma; PD, progressive disease; PR, partial response; SD, stable disease.

($n = 2$) to the expansion cohorts and received tinostamustine treatment; 14 patients were recruited to Cohort 2 and 6 to Cohort 2a (Table 4).

3.2.2 | Safety

Overall, all 20 patients who received tinostamustine experienced at least one TEAE; 65% of these were hematological (77 events in 13/20 patients; Table 5). Nineteen patients experienced TEAEs considered to be related to tinostamustine treatment, including 13 patients who experienced hematological events of all grades. In total, 70% (14/20) of patients experienced Grade 3 TEAEs and 25% Grade 4 (5/20 patients); there were no fatal (Grade 5) TEAEs. Grade 3/4 thrombocytopenia occurred in 65% of patients (13/20) with nine of these patients experiencing Grade 3 thrombocytopenia and four experiencing Grade 4 events; five patients (25.0%) discontinued the study due to thrombocytopenia. Grade 3 anemia occurred in seven patients (35%) and Grade 3/4 neutropenia in four patients (20%) (Table 6).

Eight of the 20 patients (40%) experienced nine serious tinostamustine-related TEAEs, including thrombocytopenia ($n = 1$ patient) and infusion-related reaction ($n = 1$). One patient experienced a QTcF prolongation longer than 60 msec from baseline at 60 min after the start of study drug infusion (100 mg/m^2) on Cycle 1 Day 1. During this event, the maximum QTcF was 445 ms (baseline QTcF of 369 ms) and the patient was asymptomatic. This event was confirmed by central review, and resolved on the same day without any action being taken with regard to tinostamustine, or any other medication being required; the event was assessed as Grade 3 in severity and considered related to tinostamustine.

Six patients discontinued treatment due to primarily hematological TEAEs, including five who discontinued due to thrombocytopenia.

3.2.3 | Efficacy

Patients received a median of 3 (range 1–12) cycles of tinostamustine. One patient enrolled in Cohort 2 had no post-baseline response evaluation present and so was excluded from the Full Analysis Set but retained in the Safety Analysis Set. The ORR was 37% (7/19 patients; 95% CI: 16%, 62%), including two CRs (Figure 2). This was above the proportion of treatment successes (ORR; 30%) specified in the study protocol as indicating that tinostamustine warrants further investigation in this patient population, with a mean ± standard deviation duration of response of 5.5 ± 4.2 months (median 5.0 [range 1.2–13.3] months). The CBR was 53% (10/19 patients; 95% CI: 29%, 76%). Median PFS was 3.8 months (95% CI: 2.2, 9.4 months), and median OS was NE (95% CI: 9.9 months, NE).

3.2.4 | Efficacy in Each Cohort of Patients With HL

3.2.4.1 | Cohort 2. Five patients out of the 13 patients enrolled to Cohort 2 achieved a PR as best response (median of 6.0 [range 5–12] cycles of tinostamustine); ORR was 38% (5/13 patients; 95% CI: 14%, 68%). Four patients achieved SD (median of 4.5 [range 3–6] cycles) as best response; CBR in this cohort was 62% (8/13 patients; 95% CI: 32%, 86%). Mean ± standard deviation duration of response was 5.7 ± 4.6 months (median 5.3 [range 3.0–13.3] months). Median PFS was 6.2 months (95% CI: 2.1, 14.0 months) and median OS was NE (95% CI: 14.0 months, NE).

3.2.4.2 | Cohort 2a. Two patients out of the six patients enrolled to Cohort 2a achieved a CR as best response (median of 2.5 [range 1–4] cycles of tinostamustine); the ORR in this cohort was 33% (2/6 patients; 95% CI: 4%, 78%). CBR was 33% (2/6 patients; 95% CI: 4%, 78%) with a mean ± standard deviation duration of response of 4.7 months (median 4.7 [range 4.7–4.7] months). Median PFS was 2.5 months (95% CI: 1.6, NE months), and median OS 8.0 months (95% CI: 2.9 months, NE).

TABLE 4 | Patient demographics and baseline characteristics (cohort-expansion, safety population).

	Patients with HL receiving tinostamustine		
	Cohort 2 (n = 14)	**Cohort 2a (n = 6)**	**All patients (n = 20)**
Mean ± standard deviation age (range), years	40.9 ± 14.4 (25–74)	39.3 ± 16.1 (25–63)	40.5 ± 14.5 (25–74)
Male, *n* (%)	10 (71.4)	4 (66.7)	14 (70.0)
Race, *n* (%)			
White	14 (100)	4 (66.7)	18 (90)
Other	0	2 (33.3)	2 (10)
Median (range) time since initial diagnosis, months	48.4 (15.9–306.7)	67.6 (53.5–96.0)	62.1 (15.9–306.7)
Median (range) time since most recent R/R diagnosis, months	1.1 (0.2–26.0)	0.7 (0.2–6.6)	1.0 (0.2–26.0)
ECOG performance status, *n* (%)			
0	10 (71.4)	4 (66.7)	14 (70.0)
1	3 (21.4)	1 (16.7)	4 (20.0)
2	1 (7.1)	1 (16.7)	2 (10.0)
Refractory disease[a], *n* (%)	9 (64.3)	1 (16.7)	10 (50.0)
Median (range) number of lines of prior therapy	5 (3–9)	6 (3–9)	5 (3–9)
Prior cancer therapies, *n* (%)			
ABVD/BEACOPP-like chemotherapy	14 (100)	6 (100)	20 (100)
Brentuximab vedotin	13 (92.9)	5 (83.3)	18 (90.0)
Checkpoint inhibitors[b]	12 (85.7)	4 (66.7)	16 (80.0)
Bendamustine	10 (71.4)	3 (50.0)	13 (65.0)
Stem cell transplantation	5 (35.7)	4 (66.7)	9 (45.0)
Monoclonal antibodies[c]	3 (21.4)	1 (16.7)	4 (20.0)
CHOP/CHOP-like chemotherapy	1 (7.1)	2 (33.3)	3 (15.0)
CVP/CVP-like therapy	1 (7.1)	1 (16.7)	2 (10.0)
Kinase inhibitors	1 (7.1)	0	1 (5.0)
Radiotherapy	1 (7.1)	0	1 (5.0)
Steroids	0	1 (16.7)	1 (5.0)

Abbreviations: ABVD, adriamycin (doxorubicin), bleomycin, vinblastine, and dacarbazine; BEACOPP, bleomycin, etoposide, adriamycin, cyclophosphamide, vincristine, procarbazine, and prednisone; CHOP, cyclophosphamide, doxorubicin, vincristine, and prednisone; CVP, cyclophosphamide, vincristine, and prednisone; ECOG, Eastern Cooperative Oncology Group; HL, Hodgkin lymphoma; R/R, relapsed/refractory.
[a]According to investigator judgement.
[b]Including nivolumab, pembrolizumab, cemiplimab, and spartalizumab.
[c]Including isatuximab and ADCT-301-201 (anti-CD45).

Of the 19 evaluable patients enrolled in the expansion cohort, 15 received prior CPI therapy; 11 patients from Cohort 2 and four from Cohort 2a. The ORR in this subset of CPI-treated patients was 40% (6/15 patients; 95% CI: 16%, 68%) and CBR was 53% (8/15 patients; 95% CI: 27%, 79%), with one patient achieving a CR, five a PR, and two SD. Among the 11/14 patients in Cohort 2 who received prior CPI therapy, ORR was 45% (5/11 patients; 95% CI: 17%, 77%) and the CBR was 64% (7/11 patients; 95% CI: 31%, 89%), with five patients achieving a PR and two SD. In Cohort 2a, one of the four patients who received prior CPI therapy achieved a CR resulting in an ORR and CBR of 25% (95% CI: 1%, 81%). Mean ± standard deviation duration of response was 56.4 ± 4.6 months overall, and 5.7 ± 4.6 months and 8.4 ± 5.2 months in Cohorts 2 and 2a, respectively.

4 | Discussion

This study in heavily pre-treated patients with R/R HL and limited treatment options demonstrated that tinostamustine administered on Day 1 of a 21-day cycle was generally well tolerated with no unexpected AEs and no treatment-related deaths. Most observed TEAEs were hematological, with thrombocytopenia being the main TEAE leading to treatment discontinuation. Following the implementation of modified hematologic criteria, the rate of thrombocytopenia within the study was markedly reduced and subsequent cycles were delayed in those patients in whom platelet levels decreased more than 35% compared with baseline. While patient numbers were small, promising efficacy in terms of tumor response was evident in some of these difficult-to-treat patients who had been

TABLE 5 | Number (%) of patients with treatment-emergent adverse events (TEAEs) following treatment with tinostamustine[a] (cohort expansion, safety population).

	Patients with HL receiving tinostamustine		
	Cohort 2 ($n = 14$)	**Cohort 2a ($n = 6$)**	**All patients ($n = 20$)**
Any TEAE, *n* (%)	14 (100)	6 (100)	20 (100)
≥ 1 tinostamustine-related TEAE, *n* (%)	13 (92.9)	6 (100)	19 (95.0)
≥ 1 tinostamustine-related serious TEAE, *n* (%)	6 (42.9)	2 (33.3)	8 (40.0)
Thrombocytopenia	1 (7.1)	0	1 (5.0)
ECG QTcF prolongation	0	1 (16.7)	1 (5.0)
Infusion-related reaction	1 (7.1)	0	1 (5.0)
Permanent withdrawals due to TEAEs, *n* (%)	4 (28.6)	2 (33.3)	6 (30.0)
Thrombocytopenia	3 (21.4)	2 (33.3)	5 (25.0)
Lower respiratory tract infection	1 (7.1)	0	1 (5.0)
Gamma-glutamyltransferase increased	1 (7.1)	0	1 (5.0)
Deaths due to TEAEs, *n* (%)	0	0	0

Abbreviations: ECG, electrocardiogram; HL, Hodgkin lymphoma; QTcF, corrected QT using the Fredericia formula; TEAE, treatment-emergent adverse event.
[a]Some patients experienced multiple events and were counted in all relevant categories. Patients with multiple events in the same system organ class or preferred term are counted only once in that category.

TABLE 6 | Number (%) of patients with TEAEs by grade (cohort-expansion, safety population).

	Patients with HL receiving tinostamustine in cohorts 2 and 2a ($n = 20$)					
	All grades	**Grade 1**	**Grade 2**	**Grade 3**	**Grade 4**	**Grade 5**
Any TEAE, *n* (%)	20 (100)	19 (95.0)	18 (90.0)	14 (70.0)	5 (25.0)	0
Most common TEAEs, *n* (%)						
Hematological toxicities						
Thrombocytopenia	12 (60.0)	4 (20.0)	10 (50.0)	9 (45.0)	4 (20.0)	0
Anemia	7 (35.0)	2 (10.0)	6 (30.0)	7 (35.0)	0	0
Neutropenia	4 (20.0)	0	2 (10.0)	3 (15.0)	1 (5.0)	0
Lymphopenia	1 (5.0)	0	0	0	1 (5.0)	0
Leukopenia	1 (5.0)	0	1 (5.0)	0	0	0
Febrile neutropenia	0	0	0	0	0	0
Gastrointestinal toxicities						
Nausea	14 (70.0)	14 (70.0)	2 (10.0)	0	0	0
Vomiting	9 (45.0)	8 (40.0)	4 (20.0)	0	0	0
General disorders						
Pyrexia	7 (35.0)	7 (35.0)	0	0	0	0
Asthenia	3 (15.0)	2 (10.0)	1 (5.0)	0	0	0
Injury and procedural complications						
Infusion-related reaction	3 (15.0)	2 (10.0)	2 (10.0)	1 (5.0)	0	0
Infusion site erythema	2 (10.0)	2 (10.0)	0	0	0	0
Infusion site pain	2 (10.0)	2 (10.0)	0	0	0	0
Investigations						
QTcF prolongation	1 (5.0)	0	0	1 (5.0)	0	0

Abbreviations: HL, Hodgkin lymphoma; QTcF, corrected QT using the Fredericia formula; TEAE, treatment-emergent adverse event.

previously exposed to the majority of standard of care treatments including brentuximab vedotin and CPIs. Preliminary signals of efficacy were observed during dose escalation, including in a patient with primary refractory HL. In the expansion cohort, an ORR of 37% (7/19 patients) and a median PFS of 3.8 months were observed, revealing signals of efficacy in this heavily pre-treated patient population, and exceeding the pre-specified proportion of treatment successes required to

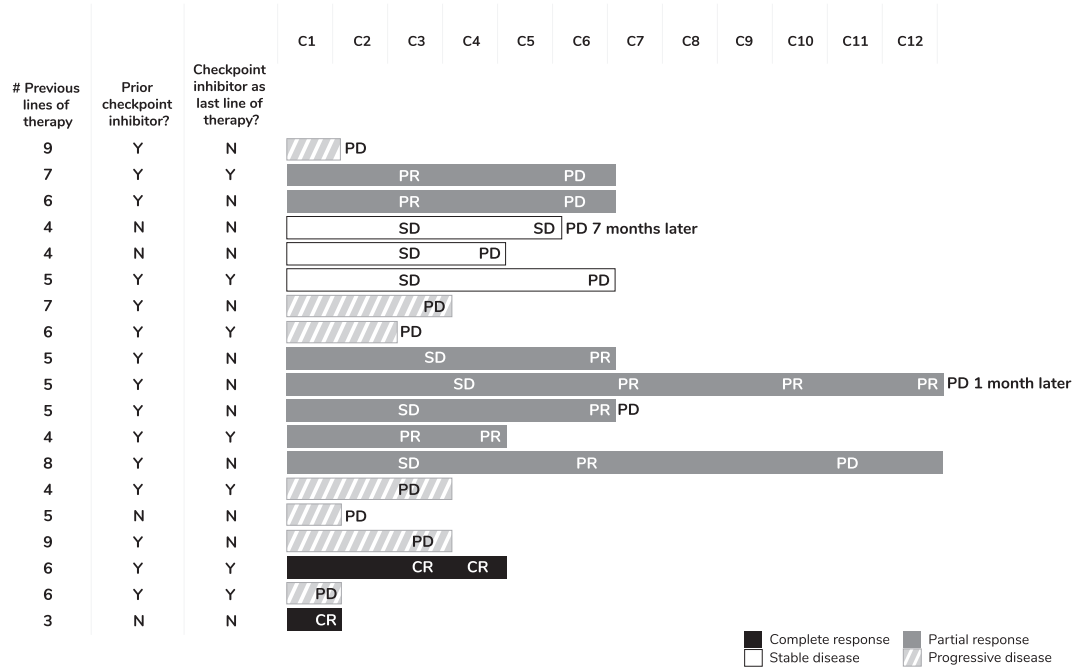

FIGURE 2 | Best overall response to tinostamustine in patients with HL (cohort-expansion population, Full Analysis Set[a]). [a]One patient enrolled in Cohort 2 had no post-baseline response evaluation present and so was excluded from the Full Analysis Set but retained in the Safety Analysis Set. CR, complete response; HL, Hodgkin lymphoma; PD, progressive disease; PR, partial response; SD, stable disease.

indicate that further investigation of tinostamustine is warranted. These findings also align with those noted with other single-agent treatments of R/R HL, including bendamustine (ORR: 55.5%), gemcitabine (ORR: 22%; median time to progression 6.4 months) and mocetinostat (disease control rate 34.8%) [15–17]. In addition, in patients who had received prior CPI therapy, an ORR of 40% (6/15 patients) was observed. These findings accord with a previous study which demonstrated a re-sensitization to chemotherapy in some patients with HL following anti-PD-1 therapy [18].

Despite advances in HL therapy, a high level of unmet medical need remains, particularly for patients with R/R disease [7–9, 19, 20]. A review article by Ma and colleagues noted that combination therapy using targeted agents such as brentuximab vedotin together with more conventional chemotherapeutic agents may be the way forward in improving survival time and quality of life for patients with R/R HL [8]. Several early phase studies have investigated novel combinations of immunotherapies and targeted therapies, and their potential application to ASCT as a means of improving outcomes for these difficult-to-treat patients [21–23]. Moreover, potential synergy between CPIs and HDAC inhibitors has been noted, together with suggestions that chemotherapy may influence the tumor microenvironment and the likelihood of response to CPI-based regimens [24]. For example, HDAC inhibitors have been shown to counteract resistance to PD-1 blockade in patients with relapsed or refractory HL and to up-regulate programmed cell death ligand one expression on tumor cells [25, 26].

Furthermore, combinations of brentuximab vedotin or CPIs with chemotherapy have been demonstrated to provide an efficient bridging strategy enabling patients with R/R HL to progress to potentially curative allogeneic stem-cell transplantation [9].

Limitations of this study include the relatively small patient population; however, as a hypothesis-generating study, tinostamustine was found to have a manageable safety profile, and the primary endpoint for the cohort-expansion phase was met with > 30% of patients demonstrating treatment success, thus warranting further investigation of tinostamustine as a therapeutic option in R/R HL.

5 | Conclusion

In conclusion, tinostamustine is a promising new therapeutic approach for the treatment of patients with R/R classical HL with limited options. This study demonstrates a manageable safety profile, with signals of efficacy indicating that further studies in larger patient populations are warranted to fully explore the potential of tinostamustine.

Author Contributions

K.H. and T.J. contributed to the design of the study. A.S., A.P., H.G., F.M., O.T., P.M., J.M.Z., R.D.F. and P.L.Z. were involved in patient care and data collection. N.M. carried out the data analysis. K.H., N.M. and T.J. evaluated the study results. All authors contributed to the drafting and critical review of the manuscript.

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Conflicts of Interest

A.S.: Honoraria from Takeda, BMS/Celgene, MSD, Gilead Kite, Janssen, Sanofi, GenMab, AbbVie, Roche, Jazz Pharma, Pierre Fabre, Astra Zeneca, GSK, and Novartis; Speakers bureau for Takeda; Advisory boards for Takeda, BMS/Celgene, MSD, Gilead Kite, Novartis, Autolus, Janssen, Sanofi, GenMab, AbbVie, Pierre Fabre, and Mundipharma; Research support from Takeda (to the Spanish Lymphoma Cooperative Group–GELTAMO). A.P.: Honoraria from Roche, Beigene, Incyte, Eli-Lilly, Sobi, MSD, BMS for educational lectures, speaker bureau and advisory board activities. H.G.: Honoraria: Gilead, Roche, BMS, AbbVie; Consultancy: Roche, Gilead, AbbVie. F.M.: Consultancy for Roche, Novartis, BMS, and AbbVie; Advisory boards for Modex therapeutics; Scientific lectures for Takeda, Chugai, and AstraZeneca. O.T.: No conflicts of interest to disclose. P.M.: No conflicts of interest to disclose. J.M. Z.: Research support from Takeda for Hodgkin MRD Consortium; Research support from Roche. R.D.F.: No conflicts of interest to disclose. K.H.: Consultant with Mundipharma Research Ltd. N.M.: Employee of Mundipharma Research Ltd. T.J.: Employee of Mundipharma Research Ltd. P.L.Z.: Consultancy for MSD, EUSA Pharma, and Novartis; Speakers' bureau for Celltrion, Gilead, Janssen-Cilag, BMD, Servier, MSD, AstraZeneca, Takeda, Roche, EUSA Pharma, Kyowa Kirin, Novartis, Incye, and Beigene; Advisory boards for Secura Bio, Celltrion, Gilead, Janssen-Cilag, BMS, Servier, Sandoz, MSD, AstraZeneca, Takeda, Roche, EUSA Pharma, Kyowa Kirin, Novartis, ADC Therapeutics, Incyte, and Beigene.

Data Availability Statement

The data that support the findings will be available following an embargo from the date of publication to allow for commercialization of research findings.

Peer Review

The peer review history for this article is available at https://www.webofscience.com/api/gateway/wos/peer-review/10.1002/hon.70000.

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Supporting Information

Additional supporting information can be found online in the Supporting Information section.