Electrocardiographic Studies of Romidepsin Demonstrate Its Safety and Identify a Potential Role for $K_{ATP}$ Channel


Abstract

**Purpose:** Romidepsin is a histone deacetylase inhibitor (HDI) approved for the treatment of both cutaneous and peripheral T-cell lymphoma (CTCL and PTCL). During development, a thorough assessment of cardiac toxicity was conducted.

**Experimental Design:** A phase II single-agent nonrandomized study of romidepsin was conducted in patients with CTCL or PTCL who had progressed after at least 1 prior systemic therapy.

**Results:** Results for the first 42 patients enrolled on the NCI 1312 phase II study of romidepsin in CTCL or PTCL showed no cardiac toxicity based on serial electrocardiograms (ECG), troponins, and MUGA scans/echocardiograms. The cardiac assessments reported herein confirm the safety of romidepsin among 131 enrolled patients, while supporting a role for electrolyte replacement. Heart rate increased an average 11 bpm following romidepsin infusion; there was no evidence of increased arrhythmia. Criteria for potassium/magnesium replacement were met before 55% of 1365 romidepsin doses; an association with hypoalbuminemia was confirmed. We propose a mechanism for ST segment flattening and depression, the most common ECG abnormalities observed: HDI-induced alteration of the activity or expression of $K_{ATP}$ channels. In addition, examination of the variants of the active transporter of romidepsin, ABCB1, showed a trend toward smaller heart rate changes in the peri-infusion period among wild-type than variant diplotypes.

**Conclusions:** We conclude that in the context of appropriate attention to electrolyte levels, the data support the cardiac safety of romidepsin.

Introduction

Histone deacetylase inhibitors (HDI) are a novel class of epigenetic agents, among which 2 were approved by the U.S. Food and Drug Administration (FDA) for patients with progressive or recurrent cutaneous T-cell lymphoma (CTCL): vorinostat (Zolinza, Merck) and romidepsin (Istodax, Celgene Corporation). Romidepsin is also approved for the treatment of progressive or recurrent peripheral T-cell lymphoma (PTCL). The HDIs promote acetylation of lysine residues of histone proteins present as an octomer surrounded by DNA in the nucleosome chromatin complex. They have been shown to modulate gene expression and to provoke cell-cycle arrest, cell differentiation, and cell death.

Clinical trials with romidepsin, vorinostat, and other HDIs including belinostat, panobinostat, entinostat, and mocentinostat are ongoing, with the hope of extending the activity of HDIs into solid tumors. During the development of this class of agents, concerns were raised about cardiac toxicity due to observed electrocardiograph (ECG) changes, although some preclinical data have suggested that in the setting of cardiac hypertrophy, HDAC inhibition may reduce atrial arrhythmia inducibility (1). Phase I and II clinical trials of HDIs have shown nondiagnostic ECG changes including T-wave flattening and ST segment depression as well as QT interval prolongation and tachyarrhythmias (2–10). No clear mechanistic explanation has been identified. Several sudden deaths occurring early in the development of romidepsin led to amendment of the protocol to exclude patients with significant underlying cardiac disease at risk for sudden death and also to avoid concomitant medications that prolong the QT interval or inhibit CYP3A4. A strict electrolyte replacement regimen was also instituted during conduct of the protocol, coupled with continued ECG monitoring, and development continued without further incident.

Piekarz and colleagues previously reported cardiac studies for 42 of the first 43 patients enrolled on the NCI 1312 phase II trial of romidepsin in TCL (2). Serial ECGs, serial...
Patients and Methods

Patients with relapsed or refractory CTCL or PTCL were enrolled in a phase II trial evaluating the safety and efficacy of romidepsin, [NCI protocol 1312]. The patients and methods and results for the CTCL and the PTCL cohorts (11, 12) have been reported in detail. The trial was registered at ClinicalTrials.gov, NCT00007345, and approved by the NCI Institutional Review Board. All participants gave written informed consent. All toxicities were graded according to the NCI Common Toxicity Criteria, version 2.0. At the NIH site, ECGs were obtained before commencement of therapy, before the infusion of each dose, within 1 hour after completion of the infusion, and on the day following treatment. An additional ECG was obtained on day 2 of the first cycle. ECGs were recorded using a HP Pagewriter XLI or a GE Marquette MAC 1200 and recorded at 25 mm/s, with an amplitude of 10 mm/mV and with 60 Hz filtering. ECGs were analyzed using Pagewriter A.0.4.01 electrocardiogram analysis software (Philips Medical Systems). In this program, the QT interval measurement is obtained by averaging the 5 longest QT intervals with a T- or T’-wave amplitude of >0.15 mV. T-wave and ST segment abnormalities were assessed by either R.L. Piekarsz or S.E. Bates and graded using NCI Common Toxicity Criteria version 2.0.

Translational Relevance

Romidepsin is a histone deacetylase (HDAC) inhibitor U.S. Food and Drug Administration (FDA)-approved for the treatment of cutaneous and peripheral T-cell lymphoma and continues in development. Cardiac safety has been intensively investigated for romidepsin and other HDAC inhibitors, generating confidence in the safety of the class. However, ST and T-wave changes in the electrocardiograph (ECG) of unclear etiology are observed in a majority of patients. This report documents a consistent increase in heart rate in all patients treated without evidence of a pro-arrhythmic effect. A high fraction of patients with T-cell lymphoma require potassium and magnesium supplementation. We present a mechanistic hypothesis for the ST segment and T-wave changes observed on serial ECGs, explaining why electrolyte replacement would be important. The report affirms the safety of romidepsin in the context of attention to potassium and magnesium supplementation; the data are important for both the academic and clinical communities, among them physicians prescribing romidepsin.
conducted, comprising 3 individual genotypes and 3 diplo-
type organizations (accounting for the coinheritance of
ABCB1 alleles), tests that were \( P < 0.05 \) were considered
marginally significant. The \textit{a priori} level of nominal signif-
ificance was set at \( P < 0.01 \) rather than accounting for a strict
Bonferroni adjustment as repeated measures of heart rates
were correlated and ABCB1 alleles were coinherited (i.e.,
correlated). Data are reported as mean \( \pm SD \) unless other-
wise indicated.

Results

**Patient characteristics**

Table 1 illustrates the patient characteristics for all 131
patients treated in the study and for the 63 patients
treated at the NIH Clinical Center only. The CTCL cohort
was almost twice as large as the PTCL cohort. The
majority of patients had an Eastern Cooperative Oncol-
y Group (ECOG) performance status of 1. Thirty-three
percent of patients received 3 to 5 cycles and 40% received
6 or more cycles of romidepsin. Fifty-one
percent of all patients in the trial had received prior
doxorubicin (52% of the NIH cohort). Excluding the
differences in ECOG performance status, the NIH cohort
is mostly representative of all patients in the trial. In all,
the 131 patients received 3,358 doses of romidepsin in
1,198 cycles; the 63 patients enrolled at the NIH received
1,292 doses in 475 cycles.

**ECG changes**

ECGs for the 63 patients treated at the NIH site were
analyzed; in total, 3,633 electrocardiograms were
reviewed. Table 2 summarizes the commonly observed
T-wave (grade I) or ST segment (grade II) abnormalities
noted on ECGs following romidepsin treatment. On exam-
inng ECGs from all doses given (Table 2), 42% had grade I
and 1% had grade II abnormalities immediately after
completion of romidepsin infusion and on the day fol-
donished infusion 66% had grade I and 4% had grade II
abnormalities. ECG abnormalities observed with the
administration of the first dose of romidepsin are detailed
in the middle panel of Table 2. Last, considering the worst
ECG grade obtained at any time during the study for each of
the 63 patients, 73% had grade I and 25% had grade II
abnormalities at some point during the treatment protocol
(Table 2). These results were comparable to the findings in
the original analysis of 42 patients (2).

**Heart rate changes following romidepsin**

A new observation was that the heart rate modestly but
consistently increased in patients receiving romidepsin.
Supplementary Figure S1A–S1C show heart rate increases
following days 1, 8, and 15 dosing in cycle 1. To determine
whether the heart rate increase varied by baseline heart
rate, the change in heart rate was also analyzed in sets of 10,
commencing with patients whose baseline heart rates were
in the 40 seconds. Notably, the magnitude of heart rate
increase did not vary with baseline (Supplementary Fig.
S2A–S2C). Median baseline heart rate was 85 \( \pm 16 \) bpm,
increasing to 96 \( \pm 15 \) bpm on cycle 1 day 1. Cycle 2 day 1
values showed a similar increase (81 \( \pm 15 \) bpm to 93 \( \pm 15 \)
bpm postinfusion). Supplementary Figures S3–S8 show
similar patterns for cycle 1 and 2. Analysis was conducted
both including and excluding the patients who received
\( \beta \)-blockers.

Fourteen patients were treated with \( \beta \)-blockers. Nine
patients were noted to be on \( \beta \)-blockers on cycle 1 day 1
before receiving romidepsin: 8 for hypertension and 1 for
long QT and atrial flutter. Five patients were commenced on
\( \beta \)-blockers between cycle 1 day 2 and cycle 3 day 15 for
tachycardia or arrhythmias observed during monitoring;
these patients had pre-existing tachyarrhythmias on pre-
enrollment Holter monitoring. Patients receiving \( \beta \)-block-
ers before romidepsin start had lower heart rates (74 \( \pm 15 \)
bpm vs. 85 \( \pm 16 \) bpm; \( P = 0.019 \)). \( \beta \)-Blockers did not
prevent the heart rate increase following romidepsin. Values
after infusion on cycle 1 day 1 were 86 \( \pm 16 \) bpm versus 96 \( \pm
15 \) bpm; \( P = 0.053 \), respectively. Figure 1 shows the heart
rate changes examined in quintiles for cycle 1 day 1 exclud-
ing patients on \( \beta \)-blockers. Cycle 2 day 1 values were similar,
with and without \( \beta \)-blockers showing similar increases in
magnitude (pre-infusion, 64 \( \pm 10 \) bpm vs. 81 \( \pm 15 \) bpm;\( P = 0.0008 \), respectively, and after infusion, 75 \( \pm 12 \) bpm vs.
93 \( \pm 15 \) bpm; \( P = 0.0011 \), respectively). We therefore
accounted for \( \beta \)-blocker therapy in subsequent analyses of
heart rate.

**Holter monitor and telemetry results in NIH cohort**

Baseline 24-hour holter data were available in 56 of the
63 patients. Cardiac rhythm monitoring with telemetry
totaling 24 hours during and after romidepsin infusion was
available for 59 patients; 19 of these patients also had
concurrent holter monitoring. Table 3 summarizes the
holter and telemetry data for baseline and after cycle 1
day 1. Frequent atrial premature complexes (APC) were defined
as >200 APCs on 24-hour holter based on 2 studies showing
increased incidence of arrhythmia and stroke with >200
APCs/24 h (14, 15). Frequent premature ventricular com-
plexes (PVC) were defined as >1,000/24 h (16). On the
baseline holter monitor recorded before commencing romi-
depsin infusion, 23 patients had grade I supraventricular
tachycardia (SVT), 5 patients had grade I ventricular tachy-
cardia (VT), and 3 patients had accelerated idioventricular
rhythm (AIVR). In addition, one patient had grade II atrial
flutter and was cardioverted before commencing on proto-
col. All events reported on telemetry and holter monitoring
during and for 24 hours after the first romidepsin infusion
were grade 1, apart from one patient considered to have
grade III arrhythmia—frequent runs of SVT, VT, and AIVR
after romidepsin infusion in a patient who also had fre-
quent APCs, frequent PVCs, and AIVR on baseline holter
before romidepsin. This patient had electrophysiologic
testing within 24 hours of receiving his third dose of
romidepsin, which showed no evidence of increased
susceptibility to arrhythmia. One patient with grade I tri-
geminy was found to have concomitant hypomagnesemia

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Electrocardiographic Effects of Romidepsin
Table 1. Baseline characteristics and cycle and dose data for all 131 patients treated on NCI 1312 or only for the NIH cohort

### Data for all 131 patients on NCI 1312

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Total</th>
<th>CTCL</th>
<th>PTCL</th>
</tr>
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<tr>
<td>Gender</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Male</td>
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<td>57</td>
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<td>Female</td>
<td>49</td>
<td>27</td>
<td>22</td>
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<tr>
<td>Diagnosis</td>
<td>131</td>
<td>84</td>
<td>47</td>
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<tr>
<td>Prior doxorubicin</td>
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<td></td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>66</td>
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<tr>
<td>No</td>
<td>65</td>
<td>62</td>
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<td>ECOOG</td>
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<tr>
<td>1</td>
<td>71</td>
<td>47</td>
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</tr>
<tr>
<td>2</td>
<td>16</td>
<td>11</td>
<td>5</td>
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<tr>
<td>Cycles administered</td>
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</tr>
<tr>
<td>1–2</td>
<td>36</td>
<td>14</td>
<td>22</td>
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<tr>
<td>3–5</td>
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<td>34</td>
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</tr>
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<td>≥6</td>
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<td>Median (range)</td>
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<tr>
<td>Age</td>
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<td>59.6 (27.6–84.3)</td>
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<td>4.5 (1–79)</td>
<td>3 (1–82)</td>
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<tr>
<td>Doses</td>
<td>12.0 (1–244)</td>
<td>12.5 (2–158)</td>
<td>9 (1–244)</td>
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### Data for NIH cohort

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>Total</th>
<th>CTCL</th>
<th>PTCL</th>
</tr>
</thead>
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<td>Female</td>
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<td>13</td>
<td>10</td>
</tr>
<tr>
<td>Diagnosis</td>
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<td>Prior doxorubicin</td>
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<td>0</td>
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<tr>
<td>ECOOG</td>
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<td>5</td>
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<tr>
<td>1</td>
<td>46</td>
<td>32</td>
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<td>6</td>
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<tr>
<td>Cycles administered</td>
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<tr>
<td>1–2</td>
<td>19</td>
<td>8</td>
<td>11</td>
</tr>
<tr>
<td>3–5</td>
<td>19</td>
<td>16</td>
<td>3</td>
</tr>
<tr>
<td>≥6</td>
<td>25</td>
<td>18</td>
<td>7</td>
</tr>
<tr>
<td>Median (range)</td>
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<tr>
<td>Age</td>
<td>53.3 (27.6–80.3)</td>
<td>55.5 (28.2–80.3)</td>
<td>57.6 (27.6–79.4)</td>
</tr>
<tr>
<td>Cycles</td>
<td>4 (1–79)</td>
<td>4 (1–79)</td>
<td>2 (1–29)</td>
</tr>
<tr>
<td>Doses</td>
<td>12 (1–158)</td>
<td>12 (2–158)</td>
<td>6 (1–81)</td>
</tr>
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</table>

### Total doses

<table>
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<tr>
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<th>Total doses</th>
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</thead>
<tbody>
<tr>
<td>Data for all 131 patients on NCI 1312</td>
<td>3359</td>
</tr>
<tr>
<td>Data for NIH cohort</td>
<td>2067</td>
</tr>
<tr>
<td>Total doses</td>
<td>1292</td>
</tr>
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</table>
(magnesium 0.59 mmol/L) and hypokalemia (potassium 3.4 mmol/L).

Cardiac adverse events in the entire population

Supplementary Table S2 outlines the cardiovascular adverse events reported for all 131 patients on the trial. The majority of events were grade I. Two deaths were observed before institution of electrolyte monitoring; both were unexpected and considered episodes of sudden cardiac death, occurring in one patient with multivalvular cardiac disease and in a second patient with severe atherosclerotic disease. Fourteen episodes of SVT occurring in 13 patients were reported during or after the romidepsin infusion; 6 of these were atrial fibrillation: 2 of which were grade I, 2 grade II, and 2 grade III. Among the patients with atrial fibrillation, grade I hypomagnesemia was seen in one patient and magnesium below the replacement level was seen in another. Potassium levels below the replacement level were seen in 3 patients. One episode of grade III atrial flutter was reported in a patient whose potassium and magnesium were both above the replacement levels. Troponin levels were routinely measured in the NIH patients within 48 hours before romidepsin administration, on day 1, and before, and 1 day after each treatment cycle; 2,353 were collected over all cycles. Sixteen events of elevated troponin I were observed in 11 patients. Eleven of these were normal on repeat assay or subsequent measurement within 24 hours and were thought to be lab errors, except for 1 patient with a lymphomatous myocardial wall mass (17) who had repeatedly elevated troponin levels.

Potassium and magnesium monitoring

Given that electrolyte monitoring and supplementation was required in patients enrolled on the clinical trial, we quantified the incidence of hypomagnesemia and hypokalemia requiring intervention as defined per protocol. Serum electrolyte measurements for 1,365 doses administered to 128 of 131 patients (between cycles 1 and 6 assessed) were analyzed. Only 10 patients (7.6%) on the trial never required electrolyte replacement as defined in the protocol during their course of

Table 2. T-wave and ST-segment changes at baseline and following romidepsin administration for patients in the NIH cohort (n = 63)

<table>
<thead>
<tr>
<th>T-wave and ST-segment abnormalities by time after administration of romidepsin</th>
<th>No. of ECGs</th>
<th>Grade 0 [n, (%)]</th>
<th>Grade I [n, (%)]</th>
<th>Grade II [n, (%)]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pretreatment</td>
<td>1,101</td>
<td>898 (82)</td>
<td>192 (17)</td>
<td>11 (1)</td>
</tr>
<tr>
<td>Immediate posttreatment</td>
<td>1,061</td>
<td>604 (57)</td>
<td>446 (42)</td>
<td>11 (1)</td>
</tr>
<tr>
<td>Day after treatment</td>
<td>1,024</td>
<td>309 (30)</td>
<td>672 (66)</td>
<td>43 (4)</td>
</tr>
<tr>
<td>Unscheduled</td>
<td>447</td>
<td>246 (55)</td>
<td>181 (40)</td>
<td>20 (5)</td>
</tr>
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</table>

T-wave and ST-segment abnormalities associated with the first dose of the first cycle

<table>
<thead>
<tr>
<th>No. of ECGs</th>
<th>Grade 0 [n, (%)]</th>
<th>Grade I [n, (%)]</th>
<th>Grade II [n, (%)]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pretreatment</td>
<td>63</td>
<td>59 (94)</td>
<td>3 (5)</td>
</tr>
<tr>
<td>Immediate posttreatment</td>
<td>61</td>
<td>48 (79)</td>
<td>12 (20)</td>
</tr>
<tr>
<td>Day 2</td>
<td>62</td>
<td>27 (44)</td>
<td>33 (53)</td>
</tr>
<tr>
<td>Day 3</td>
<td>49</td>
<td>26 (53)</td>
<td>22 (45)</td>
</tr>
</tbody>
</table>

T-wave and ST-segment abnormalities observed at any time point

<table>
<thead>
<tr>
<th>No. of patients</th>
<th>Grade 0 [n, (%)]</th>
<th>Grade I [n, (%)]</th>
<th>Grade II [n, (%)]</th>
</tr>
</thead>
<tbody>
<tr>
<td>Cycle 1 only</td>
<td>63</td>
<td>8 (13)</td>
<td>49 (78)</td>
</tr>
<tr>
<td>All cycles</td>
<td>63</td>
<td>1 (-2)</td>
<td>46 (73)</td>
</tr>
</tbody>
</table>
therapy. Figure 2 shows the number of doses requiring magnesium and potassium replacement. Fifty-five percent (746 of 1,365) of doses of romidepsin administered between cycles 1 and 6 were associated with pretreatment levels meeting criteria for protocol-mandated supplementation. Magnesium repletion was required more often than potassium repletion [529 doses (39%) vs. 428 doses (31%)].

Because both our data and previous reports suggested that hypokalemia and hypomagnesemia were common in CTCL (18) and because we observed frequent hypoalbuminemia, we tested the association of these findings. Using the NIH lower limit of normal for albumin, 3.7 mg/dL, we asked whether potassium or magnesium below or equal to the lower limit of normal (potassium <3.5 mmol/L or magnesium ≤0.75 mmol/L) or below the protocol-defined replacement levels (potassium <4.0 mmol/L or magnesium <0.85 mmol/L) was associated with low albumin. This was analyzed initially by the Wilcoxon-rank sum test. Doses from cycles 1–3 and days 1, 8, and 15 were analyzed. Results, as shown in Supplementary Table S3, showed that 28 of 98 analyses revealed a $P < 0.05$. Similar results were obtained with Fisher’s exact test and the Jonckheere–Terpstra trend test (Supplementary Table S3), indicating that higher albumin levels were associated with higher potassium or magnesium levels. However, many of the $P$ values were rather large, indicating a weak association. To further examine the association, a set of repeated measures of ANOVA was conducted on the albumin data by histology (CTCL or PTCL or both), with data restricted to the first 3 cycles. Starting with the initial model, a backward selection process was conducted on the model in a hierarchical manner. The results are summarized in Supplementary Table S4. Again, association of albumin with potassium and magnesium was found. Variation with cycle and day was also noted in some of the models. Together, the results of these 4 statistical analyses show that hypoalbuminemia, aggravated by the disease process in TCL, although not the only factor, is associated with low potassium and magnesium.

**ABCB1 genotyping**

Genotype data for the ABCB1 (P-glycoprotein) transporter was available for 61 patients (ref. 13; Supplementary Table S4). We previously noted that several ABCB1 polymorphisms and diplotypes conferred a reduced impact on the QT interval on cycle 1 day 1 following romidepsin administration (13). As observed in Fig. 3, the data (excluding patients who were on β-blockers) point to a similar trend with romidepsin-induced heart rate changes (C1D1 post-baseline), although most trends are not statistically significant ($P_{trend} < 0.18$; Fig. 3). Nonetheless, those carrying genotypes and diplotypes with increasing numbers of variant alleles have smaller heart rate changes than their counterparts carrying more wild-type alleles. Similarity between heart rate changes versus the various genotypes was expected given that all 3 ABCB1 loci are in significant linkage disequilibrium. No apparent relationship was observed when the same SNPs and diplotypes were compared with heart rate changes.

![Figure 2. Number of doses of romidepsin during which replacement of magnesium or potassium or both electrolytes was required per protocol. Only 619 doses (45%) did not require potassium or magnesium replacement.](image)

***Table 3.*** Holter monitor and telemetry results in NIH cohort

<table>
<thead>
<tr>
<th></th>
<th>Frequent APCs &gt;200/24 h</th>
<th>Frequent PVCs &gt;1,000/24 h</th>
<th>SVT</th>
<th>VT</th>
<th>AIVR</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>[n, (%)] (14,15)</td>
<td>[n, (%)] (16)</td>
<td>[n, (%)]</td>
<td>[n, (%)]</td>
<td>[n, (%)]</td>
</tr>
<tr>
<td>Baseline Holter pre-romidepsin</td>
<td>56</td>
<td>10 (18)</td>
<td>5 (9)</td>
<td>23 (41)</td>
<td>5 (9)</td>
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<tr>
<td>Telemetry during and for 24 h after romidepsin infusion$^b$</td>
<td>59</td>
<td>0 (0)$^c$</td>
<td>0 (0)$^c$</td>
<td>6 (10)$^c$</td>
<td>6 (10)$^c$</td>
</tr>
<tr>
<td>Cycle 1 day 1 Holter during and for 24 h after romidepsin infusion$^b,d$</td>
<td>19</td>
<td>5 (8)$^c$</td>
<td>0 (0)$^c$</td>
<td>6 (10)$^c$</td>
<td>3 (5)$^c$</td>
</tr>
</tbody>
</table>

$^a$One event was due to line misplacement.

$^b$Some patients had both SVT and VT.

$^c$Percentages calculated using denominator of 59.

$^d$This is a subset of patients who had both telemetry and holter monitoring during cycle 1 day 1, often triggered by ectopy observed on the baseline holter.
data obtained on cycle 2 day 1 of romidepsin treatment ($P > 0.05$).

**Discussion**

Romidepsin is an HDI approved by the FDA for treatment of CTCL and PTCL. While the drug functions as an inhibitor of deacetylase activity, the downstream events that emerge are pleiotropic. The HDIs as a class cause cell-cycle arrest, gene induction, reactive oxygen species production, and apoptosis. The relative importance of these various activities likely depends on the context of pre-existing genetic aberrations. Clinically, the HDIs have similar efficacy and toxicity profiles; activity in TCL; fatigue, nausea, and vomiting the most common adverse events; transient thrombocytopenia and neutropenia; and ECG effects including minor QT prolongation and ST and T wave changes. The mechanisms underlying these cardiac effects have not been worked out although there was speculation that acetylation of the hERG channel might cause QT prolongation (19, 20). In recently reported data, intensive investigation of romidepsin has suggested minimal impact on the QT interval, no evidence of myocardial damage, and no cumulative effects (21, 22). We have theorized that the more interesting cardiac effect may be the ST and T wave change observed in as many as 75% of patients receiving romidepsin. This report summarizes our studies of the cardiac effects of romidepsin on heart rate and ST and T wave changes, including cardiac rhythm monitoring, which did not suggest an increase in the incidence of arrhythmias following romidepsin. However, the monitoring did suggest that the patient population with TCL is one with frequent ectopy at baseline. Finally, our data call attention to the preponderance of patients whose potassium and magnesium levels met protocol criteria for electrolyte replacement.

Notably, atrial fibrillation has been observed in the development of almost every HDI. It was observed as a dose limiting toxicity in the romidepsin phase I, occurring at 24.9 mg/m$^2$ on the day 1 and 5 schedule (a dose well above the approved 14 mg/m$^2$ dose; ref. 8). Atrial fibrillation or flutter was observed during the Phase I trials of panobinostat (LBH589), LAQ824, belinostat, plitidepsin, and CHR-3996; often occurring in more than one patient for any given agent, and occasionally below doses that were defined as the maximum tolerated dose (7, 23–27). This is in contrast to the QT interval–associated arrhythmia torsades de pointes, which was observed with only one agent, LAQ824, a cinnamic hydroxamate no longer in clinical development (28). Recent evaluation of romidepsin data suggests a minimal effect on the QT interval using the Fridericia (QTcF)
correction, considered a better assessor of the QT interval than the Bazett (QTcB) correction at faster heart rates (2, 22).

Romidepsin is the most potent HDI currently in development largely targeting class I HDACs. Extensive cardiac safety data have been gathered, in part because it was the first potent and effective HDI in development and in part because of the early observation of ECG changes. During early development, 4 unexpected deaths were observed in other trials in addition to 2 in the NCI 1312 study that were suggestive of sudden death. Upon review, it was recognized that each of the deaths occurred in patients with risk factors for cardiac events including severe valvular pathology, severe atherosclerotic heart disease, sarcoidosis, and uncontrolled hypertension in a patient with neuroendocrine tumor (2, 11, 24, 29). Notably, sudden death has also been reported in patients receiving other HDIs (30–32). After these observations, the protocol was amended to exclude patients with significant cardiac disease and to require potassium and magnesium supplementation to maintain levels in a high normal range, as electrolyte deficiencies increase arrhythmia risk (33–36). Hypomagnesemia is a known risk factor for cardiac arrhythmia and sudden cardiac death (SCD; refs. 34, 35). Data from the Nurses’ Health Study reported in 2011 showed an inverse linear relationship between plasma magnesium level and SCD in which each 0.25 mg/dl increment in plasma magnesium was associated with a 41% lower risk (35). Hypokalemia is also a well-documented risk factor for cardiac arrhythmia and SCD (36, 37). Consistent with a previous report of hypomagnesemia in patients with advanced-stage CTCLs (18), we observed that half of doses required supplementation with either magnesium or potassium.

The most reproducible electrocardiographic effect of the HDIs appears to be ST segment flattening and depression and not QT prolongation. ST shift is an unusual drug effect, and in cardiology practice, this ECG finding is most frequently associated with ischemia, particularly occurring in the subendocardial region of the left ventricle. Certain drugs can induce cardiac ischemia and ST depression by inducing vasospasm of the coronary arteries, with cocaine and amphetamines being the most notorious culprits. These drugs may also result in myocardial necrosis. In addition, some anticancer drugs, such as 5-fluorouracil (5FU), are known to cause coronary vasospasm (38). Importantly, objective evidence of ischemia was sought and excluded in the case of romidepsin (2). Subjects with ST depression did not manifest mechanical dysfunction or enzyme release by troponin assay, a particularly sensitive measure. Instead, the evidence is clear that ST depression occurs in the absence of significant ischemia.

The mechanism underlying ST segment shifts induced in ischemia is somewhat controversial but likely involves activation of the ATP-sensitive potassium conductance (K\textsubscript{ATP}) in the ischemic region due to energy deprivation (39). Often considered to be cardioprotective, opening of K\textsubscript{ATP} channels can also promote arrhythmias. In response to a decrease in the ATP:MgADP ratio, opening of just 1% of the K\textsubscript{ATP} channels shortens the action potential by 50% (40, 41).

When channel opening occurs in one region of the heart but not another, a voltage gradient can develop between the deenergized subendocardium with shortened action potentials and adjacent tissue with normal action potentials, resulting in arrhythmogenic dispersion in repolarization. In addition, the shortened action potential duration and refractory period can promote phase II re-entry (42).

The dual capability of K\textsubscript{ATP} channels to promote reentry (e.g., atrial fibrillation) and ST segment shifts may suggest that HDIs affect the activity or expression of K\textsubscript{ATP} channels. In cardiac myocytes, K\textsubscript{ATP} channels are composed of 2 different subunits—a pore-forming Kir6.2 and a regulatory member of the ATP-binding cassette family, either SUR1 or SUR2A (43). HDIs are known to affect the expression of ATP-binding cassette (ABC) proteins, and in recently reported data, Flagg and colleagues found that HDIs downregulate SUR1 but upregulate SUR2 in the transformed HL-1 atrial cell line (44). Furthermore, we recently reported romidepsin-induced increased trafficking to the cell surface of an ABC protein ABCG2 due to changes in both expression and protein processing (45). In mice, K\textsubscript{ATP} channel structure is chamber-specific (atrial SUR1+Kir6.2 vs. ventricular SUR2A+Kir6.2; refs. 46, 47), however, both combinations appear to exist in both chambers in the human heart (48). Increased activity of the K\textsubscript{ATP} channel in the atrium has been associated with an increased incidence of atrial fibrillation, an arrhythmia that is rarely seen with QT prolonging drugs (48). Although functional data are needed to determine whether the effects of HDI on SUR2 expression results in increased channel activity, these data support a hypothetical explanation for the reported observations, that is, differential downregulation of SUR protein in the ventricle (resulting in ST depression) and upregulation in the atria (resulting in atrial fibrillation).

Subjects carrying variant alleles in the gene encoding an active transporter of romidepsin, ABCB1 (P-glycoprotein, MDR-1), are thought to express higher levels of ABCB1 in the cardiac endothelium (49). Consistent with these data, we recently showed that ABCB1 variant carriers have a modestly lower risk of developing QT prolongation following romidepsin than similar subjects carrying wild-type alleles (13). We previously reported that ABCB1 expression directly limits intracardiac exposure in a murine model and that mice lacking Abcb1-type P-glycoprotein are consequently more susceptible to romidepsin-induced ECG changes. In our current data, there were no clinically significant differences in heart rate changes between genotypes, but a trend to lower heart rate change in the variants did appear to be consistent with the QT observations.

Taken together, these studies again support the cardiac safety of romidepsin in the treatment of CTCL and PTCL and support its continued development in solid tumors. The studies also highlight the frequency with which low potassium and magnesium levels are observed in the TCL patient population. Mechanistically, it is not a simple matter to tie together the ST–T wave changes, the heart rate increase, and the observation of atrial fibrillation occurring as a dose-limiting toxicity. We have here hypothesized that...
some effects are linked to the variable penetration of romidepsin (and equally the other HDIs) through the myocardium from the perfusing vessels. A gradient could result that altered repolarization and thereby affected the ST and T wave, as well as the potential for ectopy. An increase in heart rate due to increased catecholamines, a decrease in electrolytes due to disease, and an alteration in repolarization due to increased activity of SUR in one part of the myocardium relative to another could create a perfect storm that in a patient with a hypertrophic myocardium or a poorly perfused myocardium could induce arrhythmia. This is a hypothesis that argues for, as recommended in the FDA-approved package insert, careful monitoring of potassium and magnesium and close observation of patients with known underlying cardiac disease (50).

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