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Clinical Activity of a Novel Multiple Tyrosine Kinase and Aurora Kinase Inhibitor, ENMD-2076, Against Multiple Myeloma: Interim Phase I Trial Results

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A B S T R A C T

Background: Despite recent improvements, multiple myeloma (MM) remains incurable, indicating the need for continued investigation of novel agents. ENMD-2076 is a novel, orally active molecule that has been shown to have significant activity against aurora and multiple receptor tyrosine kinases. Recently, we demonstrated that ENMD-2076 has significant pre-clinical *in vitro* and *in vivo* activity against MM cell lines and primary myeloma cells (Wang *et al., Br J Haematol*, 2010). Furthermore, ENMD-2076 inhibited critical pathways for MM cell survival and proliferation, including PI3K/AKT pathway with downregulation of survivin and XIAP, and Aurora A and B kinases, inducing G2/M cell cycle arrest, inhibiting angiogenesis, and the FGFR3 pathway. We present the interim results of a phase I clinical trial of ENMD-2076 in patients with relapsed and refractory MM.

Methods: An open label, single agent, dose-escalation, safety and tolerability trial of ENMD-2076 is currently conducted in heavily pre-treated, relapsed and refractory MM patients who have previously failed standard therapy. Using a 3+3 design, dose escalation with ENMD-2076 is currently being studied at the doses: 150, 225, 325 mg/day orally for 28 days. Patients are redosed in subsequent 28-day cycles according to safety, tolerability, and absence of progression. Pharmacokinetics and pharmacodynamic studies, including effect on phosphorylated histone 3 (pH3) in purified bone marrow MM cells, effect on the PI3K pathway in peripheral blood mononuclear cells (PBMC), and circulating endothelial cell precursors are being investigated.

Results: Currently, dose-escalation for the first three dose levels has been completed. Nine patients of median age 54 (range, 48-78) years were treated. There were 5 males and 5 females. The median number of prior regimens was 3 (range, 2-5), with 8 patients having failed high-dose melphalan and autologous stem cell transplantation. The most commonly observed toxicities included grades 1-2 anorexia (n=2), nausea (n=2), diarrhea (n=3), fatigue (n=1), asymptomatic elevation of amylase (n=3) and lipase (n=1), anemia (n=2), leucopenia (n=1), thrombocytopenia (n=2), and heavy proteinuria (n=1). Grades 3 toxicities included hypertension (n=1), asymptomatic elevation of lipase (n=2), and thrombocytopenia (n=1). No dose-limiting toxicity was observed with all toxicities resolving promptly upon interruption or discontinuation of dosing. All patients treated on dose level 1 had progression of disease on treatment, 1 patient in dose level 2 had stabilization of disease, and 2 patients on dose level 3 had stable disease although with 21% and 19% reduction in serum M-protein after the first cycle, and 1 patient in dose level 3 had a partial response.

Conclusion: In the ongoing phase I clinical trial, ENMD-2076 appears safe and well–tolerated at the doses tested to date. ENMD-2076 may hold promise as a treatment for MM and further study is warranted.

INTRODUCTION

ENMD-2076 is a novel, orally active molecule that has been shown to have significant activity against Aurora kinases and multiple tyrosine kinases, including Flt3, c-Kit, VEGFR2, FGFR1, FGFR3, JAK2.

We have previously shown that ENMD-2076 has *in vitro* activity againstMM cells,and *in vivo* against human MM xenografts *(Br J Haematol,* 2010).

ENMD-2076 induces early apoptosis of MM cells with caspase activation, cleavage of PARP, and modulates the expression of anti- and pro-apoptotic proteins to favor cell death. In addition, ENMD-2076 inhibits the PI3K/Akt pathway, Aurora kinases A and B, signaling through FGFR3 and VEGFR, and induces G_2/M cell cycle arrest.

Based on our preclinical studies, we initiated a phase I trial of ENMD-2076 in relapsed and refractory MM patients.

STUDY OBJECTIVE

Determine the maximum tolerated dose, safety and tolerability of ENMD-2076 as a single agent in patients with relapsed and refractory MM.

PATIENTS AND METHODS

Open-label, phase 1, 3+3 dose escalation design

Treatment:

Dose levels of ENMD-2076:

Cohort 1: 150 mg/day

Cohort 2: 225 mg/day

Cohort 3: 325 mg/day

Cycle 1: Daily ENMD-2076 for 28 days, followed by 7 days rest for PK monitoring

Cycles 2 and beyond: ENMD-2076 daily for 28 days with dose-modification for grades 3-4 toxicity.

Eligibility Criteria:

Progression of MM with at least 2 lines of prior treatment that must have included lenalidomide and bortezomib.

Age ≥18 years

ECOG performance status 0-2

Adequate organ function:

ANC >1.5x10^9/l and platelets >75x10^9/l

Serum Cr <1.5 mg/dl

AST, ALT, and bilirubin <2.5x ULN

RESULTS

1. Patient Characteristics

	n=10
Age, median (range) years	54 (48-78)
Gender, male:female, n	5 : 5
Lines of prior treatment, median (range)	3 (2-5)
Prior autologous transplantation, n	8

2. Toxicity**

	Toxicity Grade	150 mg/day (N=3)	225 mg/day (N=3)	325 mg/day (N=4)
Gastrointestinal				
Anorexia	1	-	1	1
Nausea	1	-	-	1
	2	-	1	-
Mucositis	1	-	-	1
Dyspepsia	1	-	-	2
Constipation	2	-	1	-
Diarrhea	1	-	-	2
	2	1	-	-
Elevated amylase*	1	-	-	2
	2	-	1	-
Elevated lipase*	2	-	-	1
	3	-	1	1
Elevated AST/ALT*	1	-	-	1
Cardiovascular				
Hypertension	3	-	-	1
Edema	1	-	-	1
Epistaxis	2	-	-	1
Renal				
Albuminuria	2	-	-	1
Hematological				
Anemia	2	-	-	2
Neutropenia	2	-	-	1
Thrombocytopenia	2	-	-	2
	3	-	-	1
Constitutional				
Fatigue	1	-	-	1
Tremor	2	-	-	1

*All elevations of amylase, lipase, and transaminases were asymptomatic

** Toxicity during cycle 1 of treatment

RESULTS

3. Disease Response

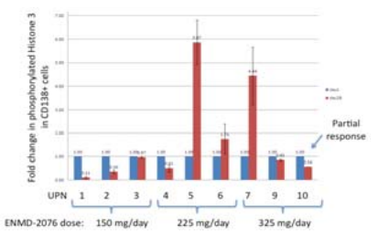
Cohort 1 (150 mg/day): 3/3 progression of MM

Cohort 2: (225 mg/day): 2/3 progression of MM
1/3 stable disease

Cohort 3: (325 mg/day): 1/4 progression of MM
2/4 stable disease (21% and 19% reduction of M-protein)
1/4 Partial response (PR)*

* Patient achieving PR is a 78 year old man with 3rd progression of MM with normal karyotype, achieved >75% reduction in volume of subcutaneous plasmacytomas and 60% reduction in urinary kappa light chain excretion.

4. Change in p-H3 in CD138+ marrow cells



CD138 immunomagnetic selection was performed on aspirated bone marrow cells and co-stained for CD138 and p-H3 for immunofluorescence microscopy. The proportion of CD138+ cells that were also positive for p-H3 were counted. Day 28 counts are normalized to baseline (day 1). Patient #9 had a 21% reduction in M-protein and patient #10 achieved a partial response. (Data for patient #8 is unavailable due to absence of day 28 sample).

CONCLUSIONS

1. **MTD of ENMD-2076 is not yet established in this ongoing trial, but dose-limiting toxicity of thrombocytopenia has been observed at 325 mg/day dose level. This cohort has been expanded to 6 patients (ongoing).**
2. **Clinical activity is noted at the 325 mg/day dose level, with 1 patient achieving a PR and 2 patients with stable disease (although with minor reduction in M-protein).**
3. **Pharmacokinetic and pharmacodynamic studies, including PI3K pathway inhibition in peripheral blood mononuclear cells and angiogenesis markers, are ongoing.**