A Phase I First In Human Pharmacokinetic and Pharmacodynamic Study of Serdemetan

In Patients With Advanced Solid Tumors

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Disclosures:
Dr. Beijsterveldt was an employee of Johnson & Johnson Pharmaceutical Research & Development, L.L.C. at the time of the study. Drs. Platero, Knoblauch, Zhuang, Yuan and Hall are employees of Johnson & Johnson Pharmaceutical Research & Development, L.L.C. Dr. Tabernero, Schöffski, Capdevila, Dirix, Lopez-Marín and Cervantes have no conflict of interest.
All authors met ICMJE criteria and all those who fulfilled those criteria are listed as authors. All approved submission to this journal.

**Previous presentations:**

Data from this study were presented at 20th EORTC-NCI-AACR symposium on “Molecular Targets and Cancer Therapeutics”, October 21-24, 2008, Geneva, Switzerland; and the 45th annual meeting of the American Society of Clinical Oncology, May 29 - June 2, 2009, Orlando, Florida and at the 46th annual meeting of the American Society of Clinical Oncology, June 4-8, 2010, Chicago, Illinois.
Statement of translational relevance:

This first in-human study described the safety profile and the maximum tolerated dose (MTD) of serdemetan, a novel tryptamine derivative initially characterized as a p53 inducer agent, in patients with advanced stage/refractory solid tumors. Extensive pharmacodynamic (PD) and pharmacokinetic (PK) analyses, as well as PK/PD modeling helped to identify a minimal biologically-effective dose. The MTD of serdemetan was 350 mg. PD analyses of paired baseline and on-treatment skin and tumor biopsies provided evidence of p53 induction. There was a dose-related increase in p53 induction in skin biopsies at doses ≥150 mg/day. Evidence existed for an exposure-effect relationship for p53 induction in skin biopsies. Outcomes of PD assays from skin and tumor biopsies were concordant in several patients. Due to exposure-related QTc toxicity observed with serdemetan, the development of derivatives lacking this liability is currently under consideration.
Abstract:

Purpose:
Originally isolated on the basis of its ability to induce p53, serdemetan demonstrated potent activity in various preclinical models, inducing S-phase arrest and apoptosis in TP53 wild-type and mutant tumors. This study evaluated the safety and tolerability of serdemetan, determined the pharmacokinetic and pharmacodynamic profiles, and identified a recommended phase II dose.

Patients and Methods:
Patients (71) with refractory solid tumors were allocated to dose-escalating cohorts (3+3 patients each) and received oral serdemetan once-daily in 21-day cycles, to determine the maximum tolerated dose (MTD) and dose-limiting toxicities (DLT). Plasma was collected for pharmacokinetic analyses. Paired baseline and on-treatment skin and tumor biopsies were performed; blood samples were collected for pharmacodynamic analyses, including p53 and macrophage inhibitory cytokine-1 (MIC-1) induction.

Results:
The MTD of serdemetan was determined to be 350 mg once-daily. During this study, Grade 3 QTc prolongation was the most common DLT and nausea (66.2%) was the most frequent treatment-emergent adverse event. Serdemetan was rapidly absorbed after oral administration and exhibited dose proportional pharmacokinetics. At steady-state, mean maximum plasma concentration ($C_{\text{max}}$) was 2330 ng/mL and mean area under plasma concentration curve (AUC$_{0-24h}$) was 43.0 µg.h/mL, with serdemetan 300 mg/day. There was a dose- and exposure-dependent p53 induction. One patient with breast cancer showed a partial response, 22 (38.6%) patients had stable disease.
Conclusions: Serdemetan treatment was associated with p53 induction in both tumor and surrogate tissue pharmacodynamic studies, and modest clinical activity. Although serdemetan was well tolerated with dose-proportional pharmacokinetics, exposure-related QTc liability was observed.
Introduction

Tumor suppressor proteins act through a variety of mechanisms to control the process of cell proliferation, including suppression of cell division, induction of apoptosis, and identification and repair of damaged DNA (1). Deactivation of the tumor suppressor protein p53 is critical for the development of many tumors (2). Serdemetan (JNJ-26854165) is a novel tryptamine derivative that was originally isolated in a p53-activating screen designed to detect increased cellular expression of p53. While initial studies suggested activity as an inhibitor of human double minute-2 oncogene (HDM2), subsequent studies demonstrated anti-proliferative activity in both TP53 wild-type and mutant tumor models, with equivalent potencies regardless of p53 status (3). Serdemetan inhibits proliferation of cell lines derived from multiple solid tumor types, with IC50 in the low micromolar range, and has demonstrated potent in vivo antitumor activity in non-small cell lung, breast, colon, prostate and glioblastoma cancer xenograft models. It induced p53-mediated apoptosis in acute leukemia cells with wild-type TP53 (3). Micro-array analysis of a wild-type TP53 tumor cell line response to various anti-cancer drugs demonstrated that serdemetan induces a gene expression profile similar to that of chemotherapeutic agents known to interfere with DNA synthesis and induce S-phase arrest (3). While the exact mechanism of action remains under investigation, the observed activities of serdemetan in preclinical models suggest that it may have efficacy in the treatment of cancer.

The primary objectives of this first-in-human, phase 1 study were to explore the safety and pharmacokinetic (PK) profiles of serdemetan, and to determine its dose limiting toxicities (DLTs) and maximum tolerated dose (MTD) in patients with advanced malignancies. A key secondary objective was to explore the pharmacodynamic (PD) effects on the p53-dependent
pathways in sequential tumor and skin biopsies as well as in blood samples in order to better characterize the recommended phase II dose for further development.

**Patients and Methods**

**Study population**

Patients of either sex, ≥18 years of age, with advanced, refractory solid malignancies were eligible for the study. All patients were required to have histological or cytological confirmation of malignancy, an Eastern Cooperative Oncology Group (ECOG) performance status score ≤2, and adequate bone marrow, liver, and renal function.

Patients were excluded from the study if they had central nervous system metastasis, uncontrolled heart disease or hypertension, or psychiatric illness incompatible with study participation. After the initial instances of QTc prolongation were observed, stricter criteria excluding patients at increased risk for QTc prolongation (clinically significant rhythm or conduction abnormality, congenital long QT syndrome, QTc at screening >450 msec, structural heart disease, liver impairment, and family history of long QT syndrome or sudden death before age 40 years) were introduced by protocol amendment. Recent, ongoing, or expected medical therapy with amiodarone or warfarin excluded patients from participation in the study.

An Independent Ethics Committee at each study site approved the protocol. This study was conducted in accordance with the Declaration of Helsinki, consistent with Good Clinical Practices and applicable regulatory requirements. All patients provided written informed consent.
to participate in the study. A Data Review Committee (DRC) was installed to ensure optimum study conduct.

**Study design**

This phase 1 study was conducted from December 2006 to February 2010 at 5 study centers in Belgium and Spain. It included a 14-day screening phase, an open-label treatment phase consisting of 21-day cycles, and an end-of-study visit within 14 days after the last dose.

The study was divided into two parts, a dose escalation phase (Part 1) and an expansion phase (Part 2). Serdemetan was administered orally, starting with a 4 mg/day dose, with subsequent doses escalated in a 3+3 dose escalation scheme, to determine the MTD. The starting dose was initially calculated at 8 mg, based on the human equivalent dose of one-sixth of the lowest dose in the rat (the most sensitive species) that does not cause severe irreversible toxicity, while daily dosing was deemed optimal for a proposed HDM2 inhibitor. The starting dose was subsequently reduced to 4mg daily, at the request of Belgium Health Authorities. Up to three additional patients were added if one patient exhibited a DLT within the first cycle of therapy (DLT period). Further dose escalation was halted if at least 2 out of a maximum of 6 patients within a cohort exhibited a DLT. Intrapatient dose escalations were allowed to minimize early cohort exposure to sub-therapeutic doses.

To minimize exposure of patients to dose levels predicted to lack therapeutic potential, a pharmacokinetically guided dose escalation (PGDE (4-5)) was initially pursued, in which dose increments of up to 500% were allowed. Once the drug-drug interaction safe level (DDISL) of
50 ng/mL maximum plasma concentration ($C_{\text{max}}$) had been achieved, an adapted “modified Fibonacci” scheme (6-7) was pursued, in which dose increases of 10 to 100% were allowed. The DRC convened after every dose cohort had completed the DLT period, and at additional time points if necessary, to review all available safety, PK, and PD data. If unacceptable toxicity occurred in patients after the DLT period, dose de-escalations were permitted.

During Part 2 of the study, additional patients were enrolled to increase the number of patients evaluated for safety, PK, and PD profiles of serdemetan, at doses and schedules that were candidates for phase II studies. At least 12 evaluable patients for each dose or schedule were to be investigated.

During cycle 1, study investigations (PK, PD, and toxicity assessments) were carried out on days -1, 1, 3, 7, 10, 14 and 21; interim safety evaluations were performed on days 3, 7, 10 and 14. Interim safety evaluations were performed on days 3, 10 and 21 of cycle 2 and on day 21 of subsequent cycles. During cycle 2, PD sampling was carried out on day 21. Tumor response assessments were performed on day 21 of every alternate treatment cycle, from cycle 2 onwards.

Serdemetan solution (Johnson & Johnson Pharmaceutical Research and Development, Division of Janssen Pharmaceutica, N.V., Beerse, Belgium) was supplied in 3 concentrations: 0.5, 5 and 20 mg/mL containing 2%, 5% or 20% hydroxypropyl-beta-cyclodextrin (HP-β-CD) respectively. Liquid formulation was initially used to allow for maximum dose flexibility during early dose escalation. Serdemetan solution was used for doses up to 300 mg/day during once-daily dosing schedule and capsules were used for doses above 300 mg/day, for both once-daily and twice...
daily dosing schedules. Immediate release, hard gelatin capsules of serdemetan were supplied in strengths of 10, 25, 100, and 200 mg. Medications metabolized by the CYP3A4, 2D6, 2C8 or 2C9 enzymes were used with caution. Drugs known to prolong the QT interval were prohibited.

**Safety evaluations**

Safety assessments included monitoring for treatment-emergent adverse events (TEAEs), DLTs, clinical laboratory tests (hematology, coagulation, blood chemistry, and urinalysis), vital signs, physical examinations, and carotid duplex ultrasound scan (pre-clinical studies had suggested carotid artery contraction). Electrocardiograms (ECG) were obtained at each specified clinic visit and analysis, and multiple gated acquisition (MUGA) scans were performed at the conclusion of each cycle to monitor cardiac function while on study.

Adverse events were evaluated in accordance with National Cancer Institute Common Terminology Criteria, Version 3.0 (8), and were monitored up to 30 days after the last dose of study drug.

**Pharmacokinetic evaluations**

*Blood and urine sample collection*

Venous blood samples (3 mL) were collected on days 1 and 21 at 0.5, 1, 2, 3, 4, 5, 6, 8 (or immediately before the 2nd daily dose for twice-daily schedule), and 24 hours postdose, during cycle 1. Predose blood samples were collected within 15 minutes before dosing on days 1, 3, 7, 10 and 14 in cycle 1, and on day 21 of each subsequent cycle. Several blood sample collections were performed immediately after ECG recordings. Urine samples were collected only during
cycle 1, on days 1 and 21, within 30 minutes before dosing and for the intervals 0 to 4, 4 to 8, 8 to 12, and 12 to 24 hours.

Food effect and drug-drug interactions were also investigated as part of this study, but will be reported separately.

**Bioanalytical procedures**

Plasma and urine concentrations of serdemetan were determined using a validated liquid chromatography-mass spectrometry method at the department of Bioanalysis, Johnson & Johnson Pharmaceutical Research & Development, Belgium. The lower limit of quantification was 0.5 ng/mL.

**Pharmacodynamic evaluations**

The study was designed to investigate parameters of serdemetan activity, based upon its demonstrated ability to induce p53 (and based upon its originally proposed mechanism of action as an HDM2 inhibitor). These parameters included p53 and Ki67 levels detected via immunohistochemical staining, and serum levels of macrophage inhibitory cytokine-1 (MIC-1) protein. Skin biopsies (at baseline and on day 21 of cycle 1) and tumor biopsies (on day 21 of cycle 1) were collected from patients expected to achieve serum C<sub>max</sub> concentrations greater than 100 ng/mL and 195 ng/mL, respectively, for analysis of p53 and Ki67. Levels of MIC-1 were evaluated using enzyme-linked immunosorbent assay (ELISA) of venous blood samples (5 mL), collected at baseline and on days 3, 10, and 21 in cycle 1, on day 21 in subsequent cycles, and at the end of treatment.
Immunohistochemistry staining (IHC) was performed using standard technique. Antibodies used were p53 mouse monoclonal clone (clone DO-1) (Santa Cruz Biotechnology) and Ki67 antigen mouse monoclonal clone (clone MIB-1) (N1633, Dako). MIC-1 was evaluated using standard ELISA methods, where we used the MIC-1 ab Ab26G6H6 for capture. For imaging and quantification, Ki67 and p53-stained slides of skin and tumor were imaged with a Mirax digital slide scanner (3DHistech) equipped with a 20x objective. The virtual images were observed with the Mirax Viewer software (3DHistech). Region of interest (ROI) of the epidermis or tumors were selected at a magnification of 40x. The selected ROI were saved as BMP files and further analysed with Axiovision image analysis software (version 4.6, Zeiss).

**Efficacy**

Efficacy was evaluated by CT scan imaging after every 2 treatment cycles, with comparison to baseline scans. More frequent evaluation was allowed, if clinically indicated. The tumor assessments were performed in accordance with Response Evaluation Criteria in Solid Tumors (RECIST) v1.1(9).

**Statistical analysis**

The PK analysis set included patients who completed the PK assessments on day 1 of cycle 1. The safety analysis set included all patients who received at least one dose of serdemetan. Descriptive statistics was used to summarize safety and efficacy results.
The plasma concentration time-curves were used to estimate $C_{\text{max}}$, time to reach maximum plasma concentration ($t_{\text{max}}$), area under the concentration-time curve (AUC) from time 0 to 24 hours ($\text{AUC}_{24h}$) for day 1 and 21, and additionally the AUC from time 0 to infinity ($\text{AUC}_{\infty}$) for day 1. For all evaluated doses, the effective half-lives ($t_{1/2}$) were estimated on the basis of the accumulation ratio between day 1 and 21. The relationship between plasma concentrations and QTcF (corrected QT using Fridericia’s formula) values was evaluated on the basis of all data for which blood samples were taken immediately after ECG recording.

Results

Patient disposition and baseline characteristics

The majority (59.2%) of patients was male, most (98.6%) were white and 97.2% of patients had an ECOG performance status score of 0 or 1 at the time of study entry (Table 1). The most frequent tumor types were colorectal cancers, sarcomas, and melanomas.

Of the 71 enrolled patients, 62 were treated in the dose escalation phase (Part 1): Fifty one patients were on a once-daily dosing schedule, and 11 were on a twice-daily dosing schedule. Nine patients were treated in the expansion phase (Part 2). A summary of the dose escalations, and the observed dose limiting toxicities, is provided in Table 2.

The median (range) number of administered cycles was 2 (1-21). Of the 71 enrolled patients, 14 patients (19.7%) received one treatment cycle, 33 (46.5%) received 2 cycles, 12 (16.9%) received 3 cycles, 4 (5.6%) received 4 or 5 cycles, and 8 (11.3%) received 6 or more cycles. Of
the 14 patients who did not receive 2 cycles, the reported reasons for discontinuation included disease progression (6), adverse event (5), and subject choice (3).

Forty-seven (66.2%) patients discontinued the study due to disease progression, 10 (14.1%) discontinued by their own choice, 9 (12.7%) discontinued after an adverse event, 3 (4.2%) discontinued due to unsatisfactory therapeutic benefits, 1 (1.4%) discontinued due to new primary malignancy, and 1 (1.4%) discontinued due to close out of the trial.

Safety
The first DLT observed during dose escalation occurred in the 300 mg daily cohort, when 1 patient of the first 4 treated (one patient did not complete the first cycle of therapy) experienced a Grade 3 QTc prolongation (Table 2). When subsequent expansion of this cohort to 3 additional patients failed to demonstrate any additional DLTs, dose escalation proceeded until the 400 mg daily dose was reached, at which time 2 of the 3 patients treated experienced DLTs. As a result, the 350 mg daily dose cohort was further expanded with 4 patients. Although there were no observed DLTs in any patient in this expansion cohort, one patient did require dose de-escalation. This fact, and concern over the potential of QTc prolongation as toxicity, prompted further expansion of the 300 mg daily dose cohort with 9 additional patients, all of which were treated without any additional observed DLTs. Thus, with the absence of any observed DLTs at the 350 mg dose (Table 2), 350 mg daily was identified as the protocol-defined MTD.

In light of the observed QTc prolongation in the daily dosing schedule, a twice-daily dosing escalation was also performed in the expectation that a lowered $C_{\text{max}}$ might mitigate the risk of
this toxicity. Two cohorts were treated on a twice-daily dosing schedule (150 mg and 200 mg), with the resulting observation of QTc prolongation as a DLT at both doses. The MTD for this dosing schedule was determined to be 150 mg twice-daily, based upon the additional finding of a Grade 3 rash in the 200 mg twice-daily cohort.

The DLTs observed during the study were Grade 3 QTc prolongation (n=4), Grade 3 rash and itching (n=1), and Grade 3 tremor (n=1) (Table 2). No Grade 4 TEAEs were reported during the DLT period.

Most (98.6%) patients reported 1 or more TEAEs during the study. A summary of the most frequently observed (>10% incidence) TEAEs, and their frequency of Grade 3 and 4 severity, is provided in Table 3. The most frequently occurring Grade 3 TEAEs reported as possibly related to the study drug were: QTc prolongation (n=6 [8.5%]); asthenia/fatigue (n=9 [12.7%]); and diarrhea, decreased appetite, and abnormal hepatic function (each in 2 [2.8%] patients). With the exception of QTc prolongation (discussed below), the presence of an exposure-related risk of these toxicities was difficult to determine, due to the small number of patients enrolled into the majority of dose cohorts (data not shown). Grade 4 TEAEs reported after the DLT period included asthenia, hypocalcaemia, hypercalcaemia, hypophosphatemia, spinal cord compression, abnormal hepatic function, and dyspnea, of which only hypophosphatemia was reported as possibly related to the study drug. There appeared to be minimal, if any, effect on cardiac left ventricular function, as only 1 of the 72 study patients (dosed at 300mg daily) exhibited a decrease in LVEF of ≥10% (a decrease from 82% at baseline to 57% at the end of Cycle 2).
Lab chemistries were independently investigated for evidence of toxicity. Grade 4 toxicities were seen in one patient with hypocalcaemia, 8 patients with gamma-glutamyltransferase (GGT) elevations, and one patient with hypophosphatemia. The abnormalities in serum GGT and alkaline phosphatase (ALP) levels were notable for both severity and frequency (Grade 3 or 4 toxicities in >10% of patients). Serum GGT abnormalities of Grade 3 and 4 severity were reported in 26 patients (Grade 3: n=18 [25.7%] and Grade 4: n=8 [11.4%]), 24 of which had started the study with abnormally elevated GGT levels (toxicity \( \geq \) Grade 2). ALP of Grade 3 was seen in 7 (10%) patients, 2 of whom had no worsening from baseline. This level of toxicity, however, was not reflected in other indicators of hepatobiliary damage, including total bilirubin (Grade 3: n=1; 1.4%), and the liver transaminases, alanine aminotransferase (Grade 3: n=4; 5.7%) and aspartate aminotransferase (Grade 3: n=3; 4.3%).

The predominant hematologic toxicity reported in this study was lymphopenia with 11 (16.4%) patients showing Grade 3 toxicity and 3 (4.5%) patients showing Grade 4 toxicity, with the majority (12) of these patients exhibiting worsening lymphocyte counts after initiation of serdemetan therapy. Other Grade 3 hematologic toxicities were reported in only 2 (3%) patients: one patient experienced Grade 3 anemia during the first cycle, after starting the study with anemia of Grade 1, and the second patient experienced an episode of pancytopenia (Grade 3 neutropenia, anemia, and leukopenia, and Grade 2 thrombocytopenia), during the 5\(^{th}\) treatment cycle. There were no instances of Grade 4 toxicities observed in these hematological parameters.

Twelve (16.9%) patients discontinued the study due to TEAEs, of which 6 discontinued due to TEAEs considered related to study drug (QTc prolongation [n=4]; abnormal hepatic function
Anemia and thrombocytopenia [n=1]). Ten (14.1%) patients experienced QTcF increases >60 msec from baseline, of which 5 patients showed QTcF >500 msec. The relatively frequent observation of QTc prolongation prompted a more thorough examination of this toxicity. Correlation of QTc changes with corresponding plasma levels of serdemetan suggested a positive correlation (Fig. 1C).

Eight (11.3%) patients died during the study due to general physical health deterioration (n=3); central nervous system metastases (n=1); lung infection and pyrexia (n=1); dyspnea (n=1); pneumonia (n=1); or intestinal obstruction (n=1). All deaths were considered either not related, or unlikely related, to study drug by the investigators.

**Pharmacokinetics**

Serdemetan was rapidly absorbed after oral administration, with C\text{max} generally observed 2-3 hours after administration of the solution, and 3-4 hours after administration of capsules (Supplementary Table 1, Fig. 1A and 1B). At 24 hours after once-daily dosing, levels were less than half of their peak values. Plasma concentrations reached steady-state after 10 days of dosing, with no further accumulation observed through day 21. After a single dose, and at steady-state, serdemetan exposure as expressed by C\text{max} and AUCs increased dose-proportionally. At steady-state, C\text{max} and AUC\text{0-24 h} were 3 to 4 times higher than after single dose indicating an effective t\text{1/2} in the order of 2 to 3 days. The mean steady-state C\text{max} was 2,330 ng/mL after 300 mg once-daily dosing and slightly lower after 150 mg twice-daily dosing. Overall, inter-patient variability (CV%) was low (<50%) for all exposure parameters for both dosing schedules. Less than 10% of the daily dose of serdemetan was recovered as parent drug in urine. The mean renal clearance was 1 L/h.
Changes in QTcF were directly related to serdemetan plasma concentrations (Fig. 1C). Steady-state $C_{\text{max}}$ concentrations after 300 mg once daily dosing caused QTcF changes of on average $>25$ msec.

**Pharmacodynamics**

Biomarker studies provided evidence of serdemetan activity in both surrogate and tumor biopsy tissue. The p53 levels in skin biopsies increased on day 21, compared with day 1. The increase was exposure-related (Fig. 2), with higher levels of nuclear p53 staining observed in skin biopsies taken from patients with higher AUC exposures. Conversely, Ki67 levels in skin biopsies decreased from day 1 to 21, but this was observed at the 300 mg/day dose only. Evidence of response in tumors was less robust. Significant increases in p53 expression were only observed in 8 patients out of 13 from whom the tumor biopsies were collected. No significant changes in Ki67 levels were observed relative to baseline in these tumor specimens. A dose dependent increase in serum MIC-1 levels was measured as a potential PD marker of serdemetan induced p53 activation. Serdemetan treatment reliably resulted in increase of MIC-1 serum levels at day 21 compared to baseline, however the magnitude of the effect was not observed to be dose dependent.

**Efficacy**

Of the 71 patients treated in this study, 57 were evaluable for efficacy, having had both a baseline and at least one post-treatment study performed for comparison. Of the 14 non-evaluable patients, 6 were reported to have discontinued as a result of an adverse event, 6 as a
result of disease progression, and 2 by patient choice. One patient with breast cancer, receiving serdemetan 150 mg twice-daily, showed a partial response (this patient also reported 102% increase in p53 levels in tumor biopsies) (Fig. 3B). Seven additional patients exhibited a decrease in tumor size, but did not meet criteria for partial response. Twenty-two (38.6%) patients had stable disease as their best overall response (evaluated as per RECIST guideline), with four of these patients exhibiting prolonged stable disease with extended courses of serdemetan therapy (of 126, 196, 308, and 420 days in patients with angiosarcoma, breast cancer, Hurthle cell carcinoma, and ependymoma, respectively). Thirty-four (59.6%) patients had progressive disease as their best overall response. The maximum tumor reduction in all patients receiving doses above 150 mg/day, the threshold that resulted in p53 induction in skin biopsies, is shown in Fig. 3A.

Discussion

Serdemetan is a novel tryptamine derivative that was originally isolated in a chemical screen, based upon its ability to induce p53 expression. Initial pre-clinical studies suggested that its mechanism of action included the inhibition of HDM2, but continued investigations, conducted in parallel with this trial, recently disproved this hypothesis by demonstrating that serdemetan was similarly active in cell lines and xenografts lacking p53 function, or in HDM2-deficient experimental models. Although it’s exact mechanism of action remains under investigation, its ability to induce S phase arrest and apoptosis in a wide range of tumor models, independent of p53 status, has been repeatedly demonstrated.

This first-in-human study of serdemetan evaluated incremental doses of serdemetan in order to identify the DLTs, and the MTDs, for both once-daily and twice-daily dosing schedules. Safety,
PK and PD (using biomarker studies) profiles of serdemetan were characterized; efficacy data for antitumor activity was also collected.

Serdemetan was rapidly absorbed after oral administration when given as a solution or capsule. Capsules showed a lag time of 0.5 to 1 hour, followed by rapid absorption that was similar to absorption from the solution. Steady-state exposure was achieved within 10 days of daily administration of serdemetan. The elimination of serdemetan was slower than anticipated, as indicated by the overall effective t1/2 of 2-3 days, in contrast to pre-clinical studies, where the elimination t1/2 was 3-8 hrs in rats, mice and dogs (data not shown).

The MTD of serdemetan was found to be 350 mg for once-daily schedule and 150 mg for twice-daily schedule. The main DLT reported was Grade 3 QTc prolongation (observed in 4 patients). Grade 2 QTc prolongation was observed in 10 additional patients, identifying QTc prolongation as the primary safety concern associated with serdemetan therapy. QTc prolongation was directly correlated with serdemetan plasma concentration. The other DLTs included rash, pruritis, and tremor. Though 8 deaths occurred during the study, all were considered to be unlikely related to serdemetan therapy by the study investigator.

Lymphopenia was observed in the majority of patients and >20% patients experienced Grade 3 or 4 severity. The TEAEs of anemia, thrombocytopenia, and neutropenia were minimal, indicating that serdemetan has little effect on bone marrow, and therefore might be used in combination with existing cytotoxic chemotherapies. Although >30% patients experienced Grade 3 or 4 elevations in GGT, the majority of these patients had elevated GGT levels at baseline,
suggesting that true incidence rate for this toxicity may be lower than that was observed. Although the MTD was determined to be 350 mg daily, as defined by the protocol, the next lower dose of 300 mg daily was recommended for subsequent phase II studies, in light of the observed dose-dependent nature of the QTc prolongation (Figure 1C), and the similar PK profiles of the 350 mg and 300 mg doses observed in this study (Figure 1B).

Biomarker analyses performed before and after the serdemetan therapy showed dose-dependent increase in p53 levels in skin biopsies, from doses 150 mg/day onwards. Though the number of samples available for tumor biomarker analysis was limited, a trend towards p53 induction in tumors was observed. Thus, the observation of serdemetan-induced p53 expression in human tumors in this study is consistent with pre-clinical studies demonstrating similar activity in both cell lines and xenograft models. Serum levels of MIC-1, a marker of p53 activation, were seen to rise after treatment with serdemetan, however the increase was not dose-dependent, and therefore MIC-1 levels were considered to be a poor pharmacodynamic marker for serdemetan.

Serdemetan showed a modest clinical activity based on the results of antitumor assessment, with one patient with advanced breast cancer exhibiting a partial response. Interestingly, a 102% increase in p53 staining was observed in the tumor of this patient. The disease remained stable in approximately 40% of patients. Some of these patients with stable disease received extended courses of serdemetan therapy, at doses that reliably induced p53 in skin biopsies. These findings, in addition to the other observed minor responses, suggest a direct effect of serdemetan on at least a subset of tumors.
In summary, this first-in-human study identified the MTD of serdemetan in patients with solid tumors in advanced stage or refractory to available therapy. Though serdemetan showed evidence of clinical efficacy in this patient population with limited treatment options, the identification of exposure-related QTc toxicity is of concern. The development of serdemetan derivatives lacking this liability is currently under consideration.
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References


Table 1  Baseline demographic and clinical characteristics

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<td>Melanoma</td>
<td>8 (11.3)</td>
</tr>
<tr>
<td>Breast</td>
<td>7 (9.9)</td>
</tr>
<tr>
<td>Renal</td>
<td>3 (4.2)</td>
</tr>
<tr>
<td>Ovary</td>
<td>2 (2.8)</td>
</tr>
<tr>
<td>Pancreatic</td>
<td>2 (2.8)</td>
</tr>
<tr>
<td>Other</td>
<td>18 (25.4)</td>
</tr>
</tbody>
</table>

*One patient was described as ‘mixed race’; #Eastern Cooperative Oncology Group
Table 2 Overview of dose escalation and dose limiting toxicities

<table>
<thead>
<tr>
<th>Dose Cohort (mg)</th>
<th>Dosing Schedule</th>
<th>Patients Treated (n)</th>
<th>DLT (n)</th>
<th>DLT Adverse Event¹</th>
</tr>
</thead>
<tbody>
<tr>
<td>4</td>
<td>Daily</td>
<td>4</td>
<td>0</td>
<td>QTc prolongation</td>
</tr>
<tr>
<td>8</td>
<td>Daily</td>
<td>3</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>20</td>
<td>Daily</td>
<td>3</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>40</td>
<td>Daily</td>
<td>4</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>60</td>
<td>Daily</td>
<td>4</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>90</td>
<td>Daily</td>
<td>4</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>150</td>
<td>Daily</td>
<td>4</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>225</td>
<td>Daily</td>
<td>7</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>300</td>
<td>Daily</td>
<td>16²</td>
<td>1</td>
<td>QTc prolongation</td>
</tr>
<tr>
<td>350</td>
<td>Daily</td>
<td>8³</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>400</td>
<td>Daily</td>
<td>3</td>
<td>2</td>
<td>Rash QTc prolongation</td>
</tr>
<tr>
<td>150</td>
<td>Twice Daily</td>
<td>8⁴</td>
<td>1</td>
<td>QTc prolongation</td>
</tr>
<tr>
<td>200</td>
<td>Twice Daily</td>
<td>3</td>
<td>2</td>
<td>Tremor QTc prolongation</td>
</tr>
</tbody>
</table>

¹ All DLTs were of Grade 3 severity
² Includes 7 patients from original dose escalation and 9 patients from subsequent cohort expansion
³ Includes 4 patients from original dose escalation and 4 patients from subsequent cohort expansion
⁴ Includes 4 patients from original dose escalation and 4 patients from subsequent cohort expansion
Table 3 Treatment-emergent adverse events experienced by >10% of patients

<table>
<thead>
<tr>
<th>Treat-Emergent Adverse Event</th>
<th>Total patients N (%)</th>
<th>Grade 3 n (%)</th>
<th>Grade 4 n (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Nausea</td>
<td>47 (66.2)</td>
<td>1 (1.4)</td>
<td>0</td>
</tr>
<tr>
<td>Vomiting</td>
<td>34 (47.9)</td>
<td>1 (1.4)</td>
<td>0</td>
</tr>
<tr>
<td>Decreased appetite</td>
<td>21 (29.6)</td>
<td>4 (5.6)</td>
<td>0</td>
</tr>
<tr>
<td>Asthenia/Fatigue</td>
<td>28 (39.4)</td>
<td>9 (12.7)</td>
<td>1 (1.4)</td>
</tr>
<tr>
<td>Constipation</td>
<td>16 (22.5)</td>
<td>1 (1.4)</td>
<td>0</td>
</tr>
<tr>
<td>Insomnia</td>
<td>16 (22.5)</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>QTc Prolongation</td>
<td>11 (15.5)</td>
<td>6 (8.5)</td>
<td>0</td>
</tr>
<tr>
<td>Abnormal hepatic function</td>
<td>10 (14.1)</td>
<td>4 (5.6)</td>
<td>1 (1.4)</td>
</tr>
<tr>
<td>Anemia</td>
<td>9 (12.7)</td>
<td>1 (1.4)</td>
<td>0</td>
</tr>
</tbody>
</table>
Figure legends:

**Fig. 1.** Plasma concentration-time profile of serdemetan
(A) Plasma concentration-time profile of serdemetan in 4 to 400 mg dose group on day 1; bid: twice-daily; sol: serdemetan solution; cap: serdemetan immediate release capsules. (B) Plasma concentration-time profile of serdemetan in 4 to 400 mg dose group on day 21; bid: twice-daily; sol: serdemetan solution; cap: serdemetan immediate release capsules. (C) Correlation of serdemetan plasma levels with QTcF changes. Plasma levels of serdemetan were assessed on the day 21 visit of cycle 1, and immediately after ECGs were performed. Plasma levels were correlated to changes in QTcF, as determined by comparing the QTcF interval in the time-matched ECG to that on pre-treatment ECG. The resulting graph shows a positive correlation, suggesting a dose-dependent effect on the QTcF interval.

**Fig. 2.** p53 measurement in skin tissue of patients
(A) Immunohistochemistry staining of p53 (brown) of skin from patients receiving serdemetan at day 1 (left panel) or day 21 (right panel). Insert indicates the quantification of the p53 signal (mean percentage of positive nuclei). (B) Graph demonstrating correlation of drug exposure to change in p53 from baseline in skin of patients treated with serdemetan. The correlation has a R^2 value of 0.6942.

**Fig. 3.** Clinical responses to serdemetan in patients and tumors
(A) Waterfall plot showing the best tumor responses, in all patients who received daily doses of 150 mg or greater. (B) p53 immunohistochemistry staining in tumor samples from patient exhibiting confirmed partial response. Left panel shows baseline p53 staining (brown). Right panel shows p53 staining (brown) from the same patient on day 21 of cycle 1. The respective levels of p53 induction are indicated (mean percentage of positive nuclei).
Fig 1

A

Day 1

Mean seremetan plasma conc. (ng/mL)

Time (hours)

B

Day 21

Mean seremetan plasma conc. (ng/mL)

Time (hours)

C

Delta Q/F vs baseline (msec)

Seremetan plasma concentration (ng/mL)

y = 1.86 + 0.011x
Fig. 2

A

Baseline

Day 21

B

Change in p53 staining (% positive nuclei)

Patient AUC [x10^3 ng/h/mL]

R^2 = 0.6942
Fig. 3

A

B

Baseline

Day 21

32.6%

65.8%
Fig 1

A

Day 1

Mean seremetan plasma conc. (ng/mL)

B

Day 21

Mean seremetan plasma conc. (ng/mL)

C

y = 1.98 + 0.011x

Delta OTC vs baseline (msec)

Seremetan plasma concentration (ng/mL)
Fig 2

A

Baseline

Day 21

4.6 %

58.6 %

B

Change in p53 staining (% positive nuclei) vs. Patient AUC [x10^3 ng.h/mL]

R² = 0.6942
A Phase I First-In-Human Pharmacokinetic and Pharmacodynamic Study of Serdemetan In Patients With Advanced Solid Tumors

Josep Tabernero, Dirix Luc, Patrick Schoffski, et al.

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