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# Single and multiple dose evaluation of a novel MetAP2 inhibitor: Results of a randomized, double-blind, placebo-controlled clinical trial

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**Aims:** Methionine aminopeptidase 2 (MetAP2) inhibition has been shown to result in significant weight loss and improved glucose control. This Phase 1 clinical trial assessed the safety and tolerability, pharmacokinetics and preliminary efficacy of a novel MetAP2 inhibitor, ZGN-1061.

**Methods:** This clinical trial included a single ascending dose (SAD) phase in healthy subjects (BMI, 23 to <30 kg/m$^2$) and a multiple ascending dose (MAD) phase in otherwise healthy subjects (BMI, 27 to 40 kg/m$^2$). SAD phase doses, administered subcutaneously (SC), were 0.2, 0.6, 1.2, 2.4, 3.6 and 4.8 mg and the MAD phase evaluated doses of 0.2, 0.6 and 1.8 mg twice weekly SC for 4 weeks.

**Results:** The SAD phase included 39 subjects (ZGN-1061, N = 28; placebo, N = 11); 90% were male and BMI was 26.4 kg/m$^2$. ZGN-1061 was well tolerated across all doses, with the most frequent adverse events being mild headache and procedural-related irritation. There were no severe or serious adverse events. All doses of ZGN-1061 were rapidly absorbed and cleared, resulting in short duration of exposure that is anticipated to minimize potential off-drug target risks. The MAD phase included 29 subjects (ZGN-1061, N = 22; placebo, N = 7); 76% were male and BMI was 33.5 kg/m$^2$. Safety observations were consistent with SAD findings. Efficacy measures in the MAD phase indicated trends for weight change (−1.5 kg total ZGN-1061 vs −0.2 kg placebo) and other biomarker changes.

**Conclusions:** ZGN-1061 was well tolerated with no safety signals in all doses tested. In addition, the desired pharmacokinetic profile and preliminary efficacy observations with ZGN-1061 support evaluation in larger and longer clinical trials.

KEYWORDS

antidiabetic drug, antiobesity drug, appetite control, lipid-lowering therapy, pharmacodynamics, pharmacokinetics

## 1 | INTRODUCTION

Methionine aminopeptidase 2 (MetAP2) inhibitors are a promising new therapeutic approach for the treatment of diabetes, obesity and associated metabolic complications. Improvements in glycaemic control and weight loss with MetAP2 inhibition appear to be the result of effects on fat metabolism, synthesis and storage, which also influence central regulation of appetite and food intake.[1–4]

The MetAP2 inhibitor, beloranib, produces consistent and clinically meaningful weight loss in clinical trials of patients with obesity, type 2 diabetes, Prader-Willi syndrome (PWS) and hypothalamic injury-associated obesity.[5–9] In a Phase 2 clinical trial involving patients with obesity and type 2 diabetes, beloranib resulted in a 10% weight loss and a 1.4% reduction in HbA1c (placebo-corrected) following 26 weeks of treatment.[7] However, beloranib development was discontinued because of an imbalance of venous thromboembolism

(VTE) events in the beloranib-treated groups compared to placebo in beloranib clinical trials.[8] Subsequent pre-clinical evaluation indicates that the prothrombotic effect of beloranib appears to be explained, at least in part, by extended endothelial cell (EC) exposure[10] that initiates an intracellular procoagulant signaling cascade.[11]

The novel MetAP2 inhibitor, ZGN-1061, was selected for clinical development because of an improved pre-clinical safety profile, which included assessment of thrombosis potential, and in vitro and in vivo efficacy similar to that of beloranib.[10] This Phase 1 clinical trial assessed the safety (including coagulation-related biomarkers), tolerability, pharmacokinetics (PK) and preliminary efficacy of ZGN-1061 in healthy normal-weight individuals and in overweight and obese individuals.

## 2 | METHODS

### 2.1 | Clinical trial design

This was a first-in-human, Phase I, randomized, double-blind, placebo-controlled, single- and multiple-ascending dose clinical trial conducted in the Netherlands from August 2016 to April 2017. The clinical trial was conducted in conformance with the principles of the Declaration of Helsinki and used Good Clinical Practice (GCP) guidelines according to the International Conference on Harmonisation (ICH). The protocol was reviewed and approved by the Independent Ethics Committee (IEC) prior to initiating the clinical trial. All subjects provided written informed consent prior to participating. The primary objective of this trial was to evaluate the safety and tolerability of ZGN-1061. Secondary and exploratory objectives were to evaluate the PK profile and pharmacodynamic (PD) parameters.

This trial was not registered in a publicly accessible database as Phase 1 investigations do not meet the FDAAA 801 definition of an applicable clinical trial and are typically exempted from the registration requirement.

#### 2.1.1 | Single ascending dose (SAD) phase

Subjects were randomized to receive ZGN-1061 (0.2, 0.6, 1.2, 2.4, 3.6 or 4.8 mg) or placebo as a single subcutaneous (SC) 0.5 mL injection. The randomization ratio was 3:1 for ZGN-1061 dose relative to placebo for each dosing cohort. Dose escalation was based on the available safety, tolerability and PK results of ≥5 dosed subjects in the preceding group. Subjects fasted overnight prior to pre-dose procedures until 4 hours after dosing. Subjects were confined to the trial site the afternoon prior to dosing (day 0) and remained in the clinic until day 3 assessments were completed. Subjects returned for an ambulatory visit on day 10.

#### 2.1.2 | Multiple ascending dose (MAD) phase

Subjects were randomized to receive ZGN-1061 (0.2, 0.6 or 1.8 mg) or placebo twice weekly (BIW) as 0.5 mL SC injections for 4 weeks (8 total injections). The randomization ratio was 3:1 for ZGN-1061 dose relative to placebo for each dosing cohort. Dose escalation was based on safety, tolerability, PD and PK results in the preceding group. Subjects fasted overnight prior to pre-dose procedures at baseline, day 14 and day 28, and fasting continued until ≥4 hours after dosing. Subjects were confined to the trial site the morning prior to dosing (day 0) and remained until day 17 assessments were completed. Subjects returned to the trial site on days 20 and 23 and were released the following day after dosing. Subjects also returned to the trial site for ambulatory visits on days 25 and 28 (with optional overnight stay on days 27 to 29).

### 2.2 | Randomization and blinding

Randomization was conducted via a computer-generated randomization code produced by the Biostatistics department of PRA Health Sciences (The Netherlands). Double-blinding was maintained for the duration of the clinical trial; all subjects, the investigator and site personnel other than pharmacy personnel were blinded. Pharmacy personnel not involved in the conduct of the trial dispensed trial medication (placebo or ZGN-1061) in containers that differed only by subject number. An unblinded Sponsor team monitored real-time safety.

### 2.3 | Subjects

Eligible subjects were 18 to 55 years of age, with a body mass index (BMI) of 23 to <30 kg/m$^2$ (SAD phase) or 27 to 40 kg/m$^2$ (MAD phase); women were of non-childbearing potential and men were either sterile or willing to use adequate contraception, and were otherwise medically healthy, with no clinically significant medical history, physical examination results, laboratory profiles, vital signs, bilateral lower extremity venous duplex Doppler ultrasounds or electrocardiograms (ECGs), as deemed by the investigator. Tobacco use within 90 days of first dose was prohibited. Subjects with a history or presence of thrombotic disease (eg, pulmonary embolism, deep vein thrombosis, and/or superficial thrombophlebitis), including clinically significant risk factors for thrombophilia (including elevated D-dimer, anticardiolipin antibodies, lupus anticoagulant, protein C deficiency, protein S deficiency, Factor V Leiden via activated protein C resistance and prothrombin G20210A mutation), were excluded.

### 2.4 | Assessments

General safety and tolerability assessments included adverse events (AEs), clinical laboratory evaluations, vital signs, 12-lead ECG, local tolerability, physical examination, Pittsburgh Sleep Quality Index (PSQI; MAD phase only) and Leeds Sleep Evaluation Questionnaire (LSEQ). Laboratory samples were collected after a fasting period of ≥4 hours, except when collecting samples for analysis of lipids, which were collected after ≥10 hours of fasting.

Coagulation-related safety assessments included bilateral lower extremity venous duplex Doppler ultrasound (performed at screening and day 3 [SAD phase] or screening and days 15 and 28 [MAD phase]) and regular blood sampling for markers of coagulation (ie, prothrombin time, international normalized ratio, activated partial thromboplastin time and D-dimer) during the clinical trial and at follow-up. Plasma D-dimer levels were assessed at screening, pre-dose and days 1, 2, 3, 10 and follow-up (SAD phase) or at screening, pre-dose and days 0, 1 to 14, 17, 21, 24, 28, 34 and follow-up (MAD phase). In the event of repeat findings of elevated D-dimer, a bilateral lower extremity venous duplex Doppler ultrasound was performed to rule out VTE.

Blood sampling for PK characterization of ZGN-1061 was conducted during the SAD phase on day 0 at pre-dose, at 5 and 15 minutes and at 0.5, 1, 1.5, 2, 3, 4, 6, 8, 10, 12, 24, 48 and 72 hours post-dose, and at follow-up. During the MAD phase, PK samples were

collected on days 0 and 14 at pre-dose, at 5 and 15 minutes and at 0.5, 1, 2, 4, 6, 8, 10, 12, 24, 48 and 72 hours post-dose, at pre-dose on days 7, 21 and 24, on days 28 and 34, and at follow-up.

Pharmacodynamic endpoints (assessed in the MAD phase) included body weight, food consumption, blood parameters (eg, leptin, adiponectin, high-sensitivity C-reactive protein [hsCRP]) and other cardiometabolic markers. The 30-minute food consumption test, conducted at baseline and days 0, 14 and 28, consisted of a meal that contained approximately 2 kg of food for each subject (ie, more than any person could eat) and the meal was weighed before and after consumption.

All laboratory assessments were conducted by PRA Health Sciences (Groningen, The Netherlands) with the exception of analyses comprising the PD endpoints, which were assessed by Myriad RBM, Inc. (Austin, Texas).

## 2.5 | Bioanalysis

The quantitative determination of ZGN-1061 in acidified EDTA plasma samples (lower limit of quantification [LLOQ] = 0.010 ng/mL) utilized solid-phase extraction, followed by ultra-high-performance liquid chromatography with tandem mass spectroscopy (File S1 Bioanalysis Methods).

## 2.6 | Analysis

Subject demographics, subject disposition, safety parameters and PK parameters were summarized descriptively by treatment group and separately for the SAD and MAD phases. Plasma PK parameters were estimated using non-compartmental analysis as appropriate. To assess the dose proportionality, a power model was applied as appropriate to the PK parameters, including maximum observed plasma concentration ($C_{max}$) and area under the plasma concentration-time curve up to time t, where t is the last point with concentrations above the LLOQ ($AUC_{0\text{-}t}$). Statistical analysis of PD parameters was performed separately for the SAD and MAD phases using observed values by visit and treatment group and using an analysis of covariance model for the change from baseline value, with the baseline measure as a covariate and treatment as a fixed effect. All statistical analyses were performed using SAS version 9.3. Analysis of safety was conducted on the safety population, that is, all subjects who received at least 1 dose of the clinical trial drug. The PK population included all subjects who received at least 1 dose of ZGN-1061 and provided adequate blood samples for bioanalysis. The PD population included all subjects who received at least 1 dose of the clinical trial drug and were without major protocol violations.

# 3 | RESULTS

## 3.1 | Baseline demographics and subject characteristics

All 39 randomized subjects (28 to ZGN-1061 and 11 to placebo) completed the assessment period in the SAD phase. Subjects in the SAD phase were 69% white and 90% male, with a baseline (mean ±standard deviation [SD]) age of 31.3 ±11.6 years, BMI of 26.4 ±2.0 kg/m$^2$, and weight of 84.4 ±9.6 kg. In the MAD phase, 29 subjects were randomized (22 to ZGN-1061 and 7 to placebo). Of these, 27 subjects completed the trial (1 discontinued because of a work schedule and 1 because of both work and family circumstances). Subjects in the MAD phase were 79% white and 76% male. At baseline, (mean ±SD) age was 39.8 ±10.8 years, BMI was 33.5 ±3.6 kg/m$^2$, and weight was 103.0 ±17.4 kg (Table S1, Supporting Information).

## 3.2 | Pharmacokinetics

### 3.2.1 | SAD phase

ZGN-1061 dose escalation was discontinued at the 4.8 mg dose because plasma ZGN-1061 concentrations and MetAP2 enzyme occupancy and substrate (thioredoxin) levels exceeded those predicted to be in the therapeutic range, as determined by non-clinical findings and previous experience with beloranib. ZGN-1061 was rapidly absorbed and cleared at all doses tested. Mean plasma concentrations of ZGN-1061 peaked approximately 15 minutes after dosing in all treatment groups and generally approached the LLOQ within 4 hours of dosing (Figure S1 and Table S2, Supporting Information). The mean $C_{max}$ and mean $AUC_{0\text{-}t}$ of ZGN-1061 increased linearly with dose. Mean $C_{max}$ ranged from 0.5 to 18.7 ng/mL and mean $AUC_{0\text{-}t}$ ranged from 0.2 to 21.6 h × ng/mL.

### 3.2.2 | MAD phase

After repeat dosing, ZGN-1061 was rapidly absorbed and cleared at all doses tested (Figure 1 and Table S3, Supporting Information). ZGN-1061 was not detected at 12 hours or at later timepoints. The shape of the PK profile was comparable to that of the SAD phase. As observed in the SAD phase, mean $C_{max}$ and $AUC_{0\text{-}t}$ of ZGN-1061 increased linearly with increasing dose, while time to maximal plasma concentration ($t_{max}$) occurred slightly later within the first 30 minutes after dosing. There was an increase in plasma ZGN-1061 concentrations after repeat dosing, illustrated by an increase in $AUC_{0\text{-}t}$ and $C_{max}$ observed with the fifth dose profile relative to the first dose

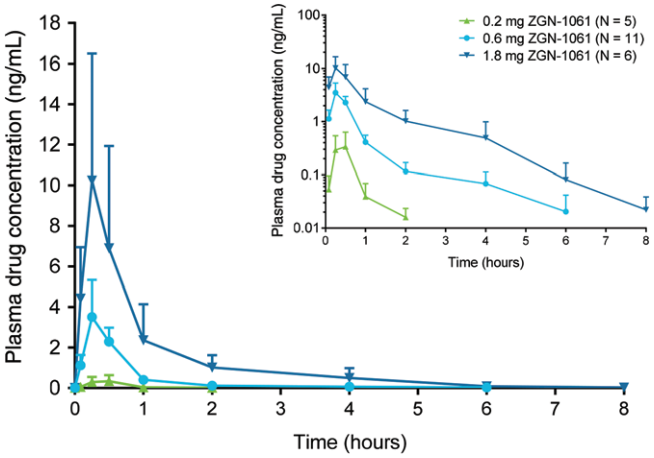


**FIGURE 1** Rapid absorption and clearance of ZGN-1061 after repeat dosing (MAD phase). Data are given as mean and SD of plasma ZGN-1061 concentrations after the 5th dose (day 14) for ZGN-1061-treated subjects in the PK population (N = 22) using linear (main figure) and logarithmic (inset) scales. ZGN-1061 was not detected at 12 hours or at later timepoints. Abbreviation: SD, standard deviation

(Table S3, Supporting Information). Exposure ratios ($R_{ss}$) of the area under the plasma concentration-time curve up to time infinity ($AUC_{0\text{-}inf}$) for the fifth compared to the first dose increased with decreasing dose and ranged from 1.4 to 2.7.

## 3.3 | General safety and tolerability

Single and repeat doses of ZGN-1061 were generally well tolerated at all doses tested in the SAD and MAD phases. There were no serious adverse events (SAEs), no severe AEs and no AEs leading to early withdrawal from the clinical trial.

### 3.3.1 | SAD phase

There were no clinically meaningful or dose-dependent changes in clinical chemistry, haematology, urinalysis, ECG, vital signs, physical examination results, local tolerability or sleep-related (LSEQ) measures. All AEs were mild in severity and there were no trends observed by ZGN-1061 dose or type of AE. When all ZGN-1061 dose groups were pooled for analysis of AEs (total ZGN-1061, N = 28), the number of subjects who experienced an AE was similar in the total ZGN-1061 (19/28; 67.9%) and placebo groups (8/11; 72.7%). The most frequent AEs in the ZGN-1061 treatment groups were headache (5/28; 17.9%) and procedural-related irritation (irritation related to ECG stickers for electrode placement) (5/28; 17.9%). The most frequent AEs in the placebo group were headache (2/11; 18.2%) and diarrhoea (2/11; 18.2%).

### 3.3.2 | MAD phase

There were no clinically meaningful or dose-dependent changes in clinical chemistry, haematology or urinalysis, ECG, vital signs, physical examination results or local tolerability measurements from baseline. All AEs were mild, with the exception of 1 moderate AE of toothache in a subject in the 0.6 mg ZGN-1061 group. The most common AEs with ZGN-1061 were mild gastrointestinal issues (similar incidence for ZGN-1061 and placebo), headache, catheter site pain and procedural-related irritation (irritation related to a bracelet worn for subject identification) (Table 1). Although transient sleep-related disturbances have been noted in long-term MetAP2 inhibitor trials with beloranib,[6] no sleep-related findings, as evaluated by the PSQI and LSEQ, were seen with ZGN-1061 in the current trial. Unlike beloranib, ZGN-1061 is not readily distributed to the central nervous system and, therefore, may not be expected to result in comparable sleep-related AEs.

## 3.4 | Coagulation-related safety

There were no VTE events and no ultrasound findings or D-dimer elevations indicative of the presence of VTE in either phase of the clinical trial. Mean values for coagulation-related biomarkers remained within

**TABLE 1** Adverse events in ≥2 subjects overall (MAD phase)

| | ZGN-1061 | | | | |
|---|---|---|---|---|---|
| | 0.2 mg (N = 5) | 0.6 mg (N = 11) | 1.8 mg (N = 6) | Total (N = 22) | Placebo (N = 7) |
| Any AE | 3 (60.0) | 9 (81.8) | 5 (83.3) | 17 (77.3) | 5 (71.4) |
| Diarrhoea | 1 (20.0) | 4 (36.4) | 2 (33.3) | 7 (31.8) | 3 (42.9) |
| Headache | 0 | 5 (45.5) | 1 (16.7) | 6 (27.3) | 0 |
| Catheter site pain | 2 (40.0) | 2 (18.2) | 0 | 4 (18.2) | 1 (14.3) |
| Application site irritation[a] | 0 | 3 (27.3) | 1 (16.7) | 4 (18.2) | 0 |
| Abdominal pain | 0 | 2 (18.2) | 1 (16.7) | 3 (13.6) | 1 (14.3) |
| Contusion | 1 (20.0) | 0 | 1 (16.7) | 2 (9.1) | 0 |
| Erythema | 0 | 0 | 2 (33.3) | 2 (9.1) | 1 (14.3) |
| Flatulence | 0 | 1 (9.1) | 1 (16.7) | 2 (9.1) | 1 (14.3) |
| Hot flush | 0 | 2 (18.2) | 0 | 2 (9.1) | 0 |
| Myalgia | 0 | 1 (9.1) | 1 (16.7) | 2 (9.1) | 0 |
| Nausea | 0 | 1 (9.1) | 1 (16.7) | 2 (9.1) | 1 (14.3) |
| Neck pain | 0 | 2 (18.2) | 0 | 2 (9.1) | 0 |
| Pollakiuria | 0 | 2 (18.2) | 0 | 2 (9.1) | 0 |
| Toothache | 0 | 2 (18.2) | 0 | 2 (9.1) | 0 |
| Ultrasound doppler abnormal[b] | 0 | 1 (9.1) | 1 (16.7) | 2 (9.1) | 0 |
| Vomiting | 0 | 2 (18.2) | 0 | 2 (9.1) | 0 |
| Abdominal distension | 0 | 0 | 1 (16.7) | 1 (4.5) | 1 (14.3) |
| Catheter site erythema | 0 | 1 (9.1) | 0 | 1 (4.5) | 1 (14.3) |
| Fatigue | 0 | 1 (9.1) | 0 | 1 (4.5) | 1 (14.3) |
| Injection site erythema | 0 | 0 | 1 (16.7) | 1 (4.5) | 1 (14.3) |
| Nasopharyngitis | 1 (20.0) | 0 | 0 | 1 (4.5) | 1 (14.3) |

Abbreviations: AE, adverse event; MedDRA, Medical Dictionary for Regulatory Activities. Data are given as percent of subjects for the safety population (N = 29) by MedDRA preferred term. AEs with onset or worsening on or after the first randomized dose of clinical trial drug are shown and are ordered by decreasing frequency in the Total ZGN-1061 group. Total includes all ZGN-1061-treated subjects.

[a] Events involved irritation as the result of a bracelet worn for subject identification.

[b] D-dimer values for these subjects were not indicative of venous thromboembolism.

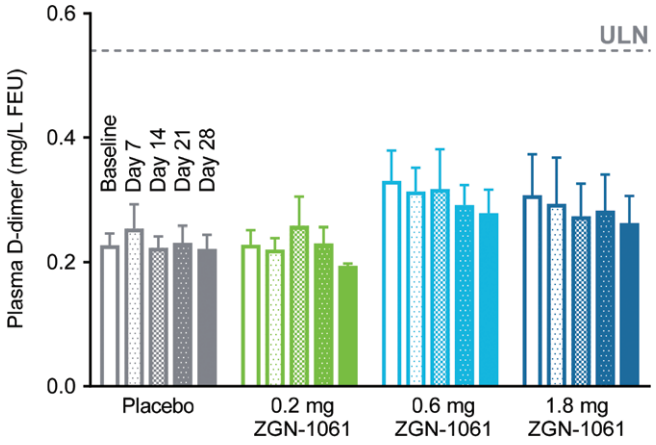


**FIGURE 2** Mean weekly D-dimer concentrations during repeat dosing of ZGN-1061 (MAD phase). Data are given as mean and SEM at each timepoint for the safety population (N = 29). Dashed line represents the ULN for the assay (0.54 mg/L FEU). Abbreviations: FEU, fibrinogen equivalent units; SEM, standard error of the mean; ULN, upper limit of normal

the normal range (Table S4, Supporting Information) and there were no clinically meaningful changes in coagulation-related biomarkers in any individual subjects.

At baseline, mean D-dimer concentrations for the 0.6 and 1.8 mg groups were slightly higher than those for the placebo and 0.2 mg groups, but were still within the normal range. There were no meaningful post-dose elevations in mean D-dimer concentrations across the dosing groups compared to baseline and placebo during the MAD phase (Figure 2). Four subjects (3 in the 0.6 mg ZGN-1061 group and 1 in the 1.8 mg ZGN-1061 group in the MAD phase) had D-dimer levels that exceeded the upper limit of normal (ULN) by >1.5-fold (0.81 mg/L FEU). None of these elevations was found to be indicative of VTE. These elevations occurred as a single isolated measurement, that is, occurred during the follow-up period, 28 days post-last dose (n = 1), or coincided with known non-VTE related causes of D-dimer elevation (sampling from blocked catheter [n = 1], tooth infection/root canal [n = 1], multiple hematomas at venipuncture sites [n = 1]).

### 3.5 | Pharmacodynamics

#### 3.5.1 | Body weight and cardiometabolic measures

Mean baseline body weight in the MAD phase was lower in the placebo group than in the ZGN-1061 treatment groups in the PD population (Table 2). A greater reduction in body weight from baseline to day 28 was observed with all doses of ZGN-1061 than with placebo (Figure 3A). The reduction in mean body weight with ZGN-1061 consisted of a gradual decline for the duration of the trial that rebounded after clinical trial drug administration was discontinued (Figure 3B).

ZGN-1061 treatment produced an apparent dose-response reduction in total amount of food consumed during the 30-minute meal challenge, with the highest dose of ZAF-1061 associated with an approximate 40% reduction in food consumption on day 28 compared to baseline (Table 2). There were also observed trends in reductions in waist circumference, low-density lipoprotein (LDL) cholesterol and hsCRP relative to placebo (Table 2). In a subset of subjects with abnormally elevated LDL cholesterol at baseline (≥130 mg/dL, N = 13), there was a trend toward a greater reduction in LDL cholesterol in the combined group of ZGN-1061-treated subjects (baseline mean ±SD, 158.3 ±25.2 mg/dL; mean ±SD change from day 28 to baseline, −18.0 ±18.4 mg/dL, N = 10) compared with placebo (187.9 ±16.1 mg/dL; −3.9 ±21.5 mg/dL, N = 3). Similarly, the change in hsCRP was calculated in subjects with elevated baseline hsCRP (≥1.5 μg/L, N = 22). A greater reduction in hsCRP was observed in subjects in the combined ZGN-1061 treatment groups (baseline mean ±SD, 5.6 ±4.9 μg/L; mean change from day 28 to baseline, −2.7 ±3.4 μg/L, N = 18) compared with placebo (5.2 ±6.6 μg/L; −1.6 ±1.1 μg/L, N = 4). There was also a trend toward reductions in leptin and increases in adiponectin in the ZGN-1061 treatment groups compared with placebo. No treatment-group differences in ECGs, blood pressure or heart rate were observed (Table S4, Supporting Information).

## 4 | DISCUSSION

In this first-in-human clinical trial of single and multiple ascending doses of ZGN-1061 for up to 28 days, ZGN-1061 was generally well tolerated and no notable safety signals were observed. There were no VTE events, no ultrasound findings or D-dimer elevations indicative of the presence of VTE, and no clinically meaningful changes in coagulation-related biomarkers. ZGN-1061 was rapidly absorbed and cleared, consistent with pre-clinical observations[10] and the desired PK profile, to avoid extended EC exposure. Efficacy data trends support a favourable effect of ZGN-1061 on body weight and cardiometabolic measures, consistent with MetAP2 inhibition and results of pre-clinical studies.[10,12]

ZGN-1061 binds MetAP2 in an irreversible covalent interaction, which diminishes overall MetAP2 activity. Subsequent administrations of ZGN-1061 will similarly bind to and inhibit newly synthesized functional MetAP2. Rapid absorption and clearance of ZGN-1061 was observed during single and repeat SC administration with peak concentrations occurring within 30 minutes and a half-life ($t_{1/2}$) of approximately 1 hour (range, 0.4-1.3 hours) that resulted in no detection of circulating ZGN-1061 12 hours after dosing for the doses selected for further multi-dose assessment. The relatively short duration of circulating concentrations of ZGN-1061, in combination with covalent binding of MetAP2, is hypothesized to promote target engagement and prolonged pharmacologic activity, while minimizing overall drug exposure to ECs. The shorter $t_{1/2}$ of ZGN-1061, compared to the MetAP2 inhibitor beloranib ($t_{1/2}$ ranged from 6 to 12 hours), is thus thought to contribute to the improved safety profile, including reduced risk of coagulation.

Adverse events in the SAD and MAD phases were mild, for the most part, and did not result in clinical trial discontinuation. In the SAD phase, dose escalation was discontinued because exposure substantially exceeded the therapeutic range. There were no notable AEs or safety findings after single dosing of up to 4.8 mg. Repeat dosing during the MAD phase was also generally well tolerated, with mild headache and procedural-related irritation events (non-injection site-related) reported more frequently than events with placebo.

All subjects were screened for signs of VTE prior to the clinical trial and were regularly monitored for evidence of VTE by D-dimer measurements taken at screening, pre-dose, during drug administration and at follow-up. D-dimer is a fibrin degradation product that is a conventional

**TABLE 2** Change in weight and cardiometabolic measures with repeat administration of ZGN-1061 (MAD phase)

| | ZGN-1061 | | | | |
|---|---|---|---|---|---|
| | 0.2 mg (N = 5) | 0.6 mg (N = 11) | 1.8 mg (N = 6) | Total (N = 22) | Placebo (N = 6) |
| Body weight, kg | | | | | |
| Baseline | 104.1 ±19.0 | 108.2 ±20.4 | 107.6 ±11.0 | 106.9 ±16.7 | 96.3 ±22.5 |
| Change | −2.1 ±1.9 | −1.0 ±2.2 | −1.7 ±1.8 | −1.5 ±1.9 | −0.2 ±1.3 |
| Food consumption test intake, g | | | | | |
| Baseline | 1002 ±401 | 942 ±422 | 1132 ±314 | 1007 ±382 | 1012 ±362 |
| Change | −21 ±184 | −121 ±245 | −482 ±258** | −204 ±294 | −51 ±174 |
| Waist circumference, cm | | | | | |
| Baseline | 101.9 ±8.3 | 112.4 ±10.0 | 107.2 ±8.8 | 108.6 ±9.9 | 96.3 ±19.1 |
| Change | −6.30 ±4.6* | −0.8 ±7.2 | −3.2 ±2.1 | −2.9 ±5.7 | 4.4 ±10.4 |
| LDL cholesterol, mg/dL | | | | | |
| Baseline | 112.0 ±28.6 | 139.3 ±24.0 | 131.9 ±46.6 | 131.1 ±32.7 | 148.6 ±45.6 |
| Change | −15.4 ±19.5 | −6.9 ±13.8 | −12.2 ±25.8 | −10.6 ±18.7 | −7.7 ±19.1 |
| HDL cholesterol, mg/dL | | | | | |
| Baseline | 44.8 ±8.9 | 53.7 ±11.1 | 41.2 ±4.0 | 48.3 ±10.5 | 47.6 ±15.2 |
| Change | −2.3 ±3.5 | −0.0 ±3.9 | −0.0 ±2.4 | −0.6 ±3.4 | −3.2 ±6.7 |
| Triglycerides, mg/dL | | | | | |
| Baseline | 89.4 ±34.9 | 113.8 ±80.4 | 134.5 ±62.8 | 113.9 ±67.2 | 133.0 ±76.1 |
| Change | 6.0 ±42.7 | 20.3 ±42.9 | −4.1 ±45.7 | 9.4 ±42.7 | 22.3 ±60.4 |
| hsCRP, μg/mL | | | | | |
| Baseline | 2.7 ±2.2 | 4.8 ±4.0 | 6.5 ±7.3 | 4.8 ±4.8 | 3.6 ±5.6 |
| Change | −0.7 ±3.2 | −1.7 ±3.3 | −3.5 ±3.9 | −2.0 ±3.5 | −1.0 ±1.2 |
| Adiponectin, μg/mL | | | | | |
| Baseline | 2.6 ±1.1 | 3.4 ±1.5 | 2.5 ±0.8 | 3.0 ±1.3 | 3.5 ±1.7 |
| Change | 0.5 ±0.4** | 0.3 ±0.5*** | −0.2 ±0.6 | 0.2 ±0.6** | −1.0 ±0.7 |
| Leptin, ng/mL | | | | | |
| Baseline | 26.7 ±18.6 | 27.0 ±25.6 | 22.3 ±12.6 | 25.7 ±20.5 | 17.5 ±14.7 |
| Change | −9.5 ±11.4 | −7.0 ±6.1 | −6.1 ±2.5 | −7.3 ±6.8 | −4.3 ±5.3 |

Abbreviations: HDL, high-density lipoprotein; hsCRP, high-sensitivity C-reactive protein; LDL, low-density lipoprotein; PD, pharmacodynamic; SD, standard deviation. *$P$ < .05, **$P$ < .01, ***$P$ < .001 for comparison vs placebo. Data are given as mean and SD. Change is from baseline to day 28. Cardiometabolic data are for the PD population (N = 28). Weight data include only those subjects in the PD population with baseline and day 28 measurements (N = 26; N = 9 for 0.6 mg ZGN-1061). Total includes all ZGN-1061-treated subjects.

marker of clotting. Because D-dimer is markedly elevated during active clot formation and lysis, D-dimer levels within the normal range have a high predictive value for ruling out acute thrombosis.[13,14] However, an elevated D-dimer level is not a specific marker for coagulation, because increased D-dimer levels can be caused by many other events, including recent surgery, active or recent bleeding, hematomas, including those associated with phlebotomy, infection, trauma and inflammation. In this trial, all D-dimer elevations were followed by a repeat D-dimer measurement and/or lower extremity ultrasound to rule out a VTE. All instances of persistently elevated D-dimer levels were not indicative of VTE, supported by lower-extremity ultrasound, and instead coincided with clinical observations of other known causes of D-dimer elevation (eg, tooth infection/root canal, multiple hematomas).

As this was a Phase 1 clinical trial designed to monitor and assess safety and tolerability, subjects were confined to the clinic for the majority of the MAD phase, with scheduled meals and meal times, as well as limited activity. These and other clinical trial factors, such as the relatively small sample size, sequential conduct of cohorts, and lower mean baseline weight in the placebo group, limit between-group comparisons, including assessment of dose-response, and the generalizability of trial results. Further titration of the dose response and evaluation of the full extent of weight loss and other efficacy parameters is needed.

Despite these limitations, there was a consistent trend toward weight loss with ZGN-1061 compared to placebo during repeat administration in the MAD phase that rebounded after drug treatment was discontinued. ZGN-1061 administration was also accompanied by dose-dependent reductions in food intake, which warrants further evaluation. Trends towards improvements in cardiometabolic markers (eg, reductions in waist circumference, hsCRP, LDL cholesterol and leptin, and increases in adiponectin) were also observed with ZGN-1061. These changes suggest that patients treated with ZGN-1061 may experience improvement in glucose control and significant reduction in body weight as observed in pre-clinical studies of ZGN-1061 in obese mice and in clinical trials of other MetAP2 inhibitors.[5–10,12]

Results of this Phase 1 clinical trial indicate that ZGN-1061 is associated with a brief exposure profile, was observed to be generally safe with no prothrombotic signals and was well tolerated in healthy normal-weight individuals and in overweight and obese individuals. These findings, in combination with positive trends in key PD parameters, suggest that ZGN-1061 may be effective in the treatment of cardiometabolic

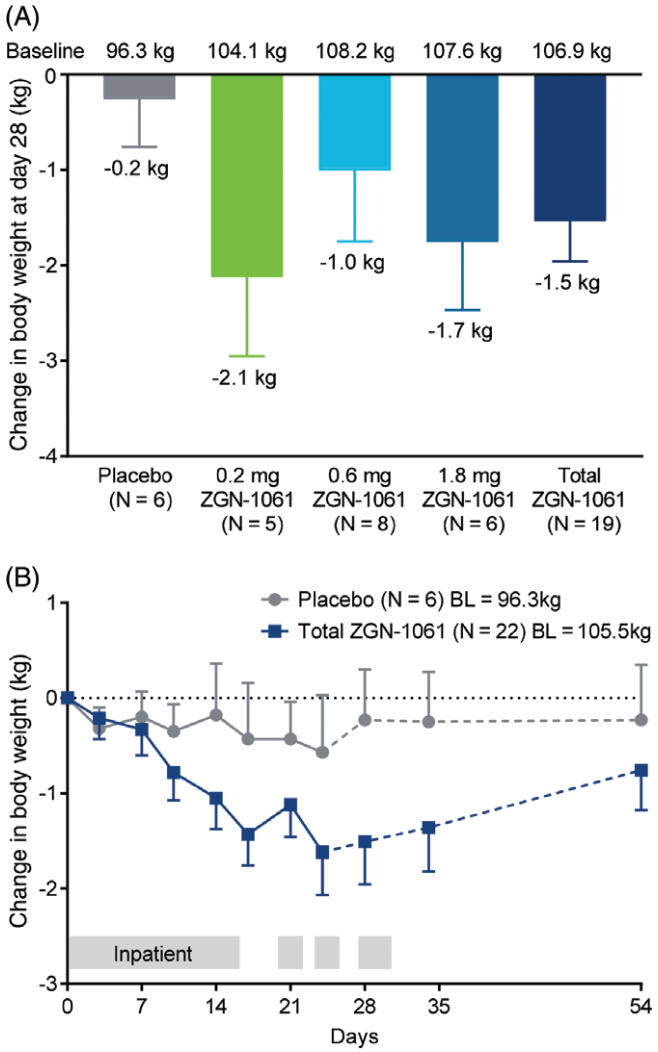


**FIGURE 3** Change in body weight with repeat administration of ZGN-1061 (MAD phase). Data are given as mean and SEM (change from baseline) or mean (baseline) for the PD population (N = 28). Total includes all ZGN-1061-treated subjects. A, Mean change in weight from baseline to day 28 in subjects with both baseline and day 28 measurements (observed values). B, Mean weight change by visit. Dashed line indicates follow-up period during which no treatments were administered. Abbreviations: BL, baseline weight; SEM, standard error of the mean

diseases such as type 2 diabetes and obesity and thus support the evaluation of ZGN-1061 in larger Phase 2 clinical trials.


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## Conflict of interest

JM, DZ, TK, PI, DK, and KT are employees of and hold stock in Zafgen, Inc.

## Author contributions

JM, DZ, TK, PI, DK, and KT designed the study, participated in data review and/or interpretation, and contributed to the writing of the report. DZ contributed to the statistical analysis and takes responsibility for the integrity of the data and the accuracy of the data analysis. All authors critically reviewed the report and approved the final version.


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## SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.


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