Original Article
CLINICAL TRIALS: BEHAVIOR, PHARMACOTHERAPY, DEVICES, SURGERY

Obesity

Ascending Dose-Controlled Trial of Beloranib, a Novel Obesity Treatment for Safety, Tolerability, and Weight Loss in Obese Women

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Objective: Evaluate the safety and tolerability of beloranib, a fumagillin-class methionine aminopetidase-2 (MetAP2) inhibitor, in obese women over 4 weeks.
Design and Methods: Thirty-one obese (mean BMI 38 kg/m^2) women were randomized to intravenous 0.1, 0.3, or 0.9 mg/m^2 beloranib or placebo twice weekly for 4 weeks ($N = 7$, 6, 9, and 9).
Results: The most frequent AEs were headache, infusion site injury, nausea, and diarrhea. Nausea and infusion site injury occurred more with beloranib than placebo. The most common reason for discontinuation was loss of venous access. There were no clinically significant abnormal laboratory findings. In subjects completing 4 weeks, median weight loss with 0.9 mg/m^2 beloranib was −3.8 kg (95% CI −5.1, −0.9; $N = 8$) versus −0.6 kg with placebo (−4.5, −0.1; $N = 6$). Weight change for 0.1 and 0.3 mg/m^2 beloranib was similar to placebo. Beloranib (0.9 mg/m^2) was associated with a significant 42 and 18% reduction in triglycerides and LDL-cholesterol, as well as improvement in C-reactive protein and reduced sense of hunger. Changes in β-hydroxybutyrate, adiponectin, leptin, and fibroblast growth factor-21 were consistent with the putative mechanism of MetAP2 inhibition. Glucose and blood pressure were unchanged.
Conclusions: Beloranib treatment was well tolerated and associated with rapid weight loss and improvements in lipids, C-reactive protein, and adiponectin.

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Introduction

Obesity has profound social, medical, and financial costs, and its prevalence in developed countries is increasing to unprecedented proportions (1). Weight reduction generally improves the diseases associated with adipose dysfunction (2). However, adherence to energy restriction diets is problematic and generally unsuccessful (3) and approved medical therapies have only modest efficacy for long-term weight management (4). In most cases, toxicity and side effects have hampered the development of potential weight-loss drug candidates (5-8).

Inhibitors of methionine aminopetidase 2 (MetAP2), originally developed as anti-angiogenic agents for the treatment of cancer (9), are a novel mechanism for inducing significant and sustained weight loss in animal models of obesity at low doses that do not impact angiogenesis (10-13). Fumagillin, a natural product isolated from ***aspergillus fumigatus***, is a potent and selective MetAP2 inhibitor (14). Daily fumagillin administration normalizes weight, improves insulin sensitivity, and reduces glucose levels in mice with diet-induced obesity (DIO) (13). Although the complete mechanism for the anti-obesity actions of MetAP2 inhibitors remains to be understood, it is thought that MetAP2 inhibitors have intracellular actions that lead to reduced fat biosynthesis and increased fat oxidation and lipolysis (10). Furthermore, weight loss following fumagillin treatment in DIO mice was not associated with reduced adipose vascular density, indicating that weight loss may not be associated with an anti-angiogenic effect (13).

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Beloranib is a synthetic fumagillin analog displaying potent and selective inhibition of MetAP2 (12). Like other fumagillin analogs, continuous incubation with beloranib inhibits endothelial cell proliferation and in xenografted mice, high doses of beloranib suppress angiogenesis and tumorigenesis (15). Subcutaneous or intravenous administration of low doses of beloranib to rats and mice significantly reduces cumulative food intake and bodyweight (12). Notably, beloranib-treated animals have significantly reduced adipocyte size, and smaller epididymal and mesenteric fat pads than control.

The purpose of this study was to determine the safety, tolerability, and pharmacology of beloranib for obesity in obese adult women and to characterize the effects of beloranib on bodyweight, sense of hunger, and cardiometabolic risk factors.

Methods and Procedures

Study design and subjects

Subjects were enrolled at two study centers in Australia from January 2010 through October 2010 under the guidelines of the Declaration of Helsinki and according to the Australian Clinical Trial Notification Scheme. Institutional review boards reviewed and approved the protocol and amendments for each study center, and all subjects provided written informed consent before participating in the trial.

This double-blind, placebo-controlled, sequential dose-escalation study consisted of a screening period, a 4-week treatment period and a 10 day post-treatment follow-up. Eligible subjects were non-diabetic female, aged 18-60 years, weighed at least 50 kg, and had a stable BMI of 32-45 kg/m^2. Subjects were required to be postmenopausal or of non-childbearing potential and otherwise healthy.

The study took place in three phases. During the first phase, subjects received 0.1 mg/m^2 beloranib or placebo (Cohort 1). The starting dose of 0.1 mg/m^2 beloranib was selected because it is 1% of the maximum tolerated dose identified in a prior clinical trial (16). Subsequent doses were 0.3 mg/m^2 (Cohort 2) and 0.9 mg/m^2 (Cohort 3). Before treatment of the next cohort could begin, a Safety Review Committee provided assent after reviewing 2 weeks of safety data from the previous cohort. Qualified subjects were randomized to receive intravenous (IV) beloranib or placebo in a 3:1 ratio (within each cohort), double-blinded, administered twice weekly (BIW) for 4-weeks.

There were two inpatient visits (Days 1-2 and 26-27), as well as six outpatient visits in which beloranib or normal saline (placebo) was administered as a 1-hour IV infusion. Baseline was at Day 1. Subjects were required to return to the study center for a safety evaluation 10 days after the last visit (Day 36). A telephone interview was performed on Day 56 to follow-up on AEs. Dietary and exercise counseling were not provided.

Safety

Vital signs, physical exam, electrocardiograms (ECGs), safety laboratory tests, adverse events (AEs), concomitant medication usage, changes in hunger, bodyweight, and waist circumference were documented at each visit. Urinalysis, urine pregnancy, drug screen, and cotinine were assessed during select visits. Cardiac rhythm and QTc interval changes were also monitored via routine 12-lead ECGs during inpatient visits (Days 1 and 26) pre-infusion, immediately post-infusion, and at 1, 4, 12, and 24 hours post-infusion.

Pharmacology

Blood samples for plasma pharmacokinetic (PK) evaluation were obtained at each visit. Calculated by using typical noncompartmental analyses, beloranib plasma PK was determined with the use of a validated, Good Laboratory Practice-compliant liquid chromatography-tandem mass spectrometry LC-MS/MS method (TetraQ, Brisbane, Australia). The limit of quantitation for this method was 0.05 ng/mL. PK analysis included data reported from all subjects in the safety population who provided an adequate sample.

Bodyweight and cardiometabolic risk factors

Bodyweight was measured at approximately the same time at each visit. Waist circumference was assessed using a tape measure placed around the abdomen 1 cm above the iliac crest. Sense of hunger was assessed in the fasted state using a 10-point visual analog scale, which has been reported to be reliable in appetite research (17). Subjects were asked to rate their overall sense of hunger for the previous 2 days on a scale of 1-10, where 10 was extremely hungry and 1 was not hungry at all. On infusion days, sense of hunger was assessed pre-infusion.

Fasting blood samples were collected at all visits and on Day 36 (10 days after last infusion) to assess changes in fasting lipids, β-hydroxybutyrate, free thyroid hormones (fT3, fT4, and thyroid stimulating hormone), glucose, and insulin. CRP and leptin were analyzed by Rules-Based Medicine, Inc. (Austin, TX, USA) using multi-analyte profile (MAP) methods (CardiovascularMAP® and MetabolicMAP®). Plasma fibroblast growth factor (FGF)-21 concentrations were determined with a commercially available assay (Quantikine FGF21 Assay Kit from R&D Systems, Inc., Minneapolis, MN, USA). High molecular weight (HMW) adiponectin concentrations were determined by proteolytic digestion of the mid- and low-molecular weight forms of adiponectin followed by a specific enzyme-linked immunosorbent assay (Alpco Diagnostics, Salem, NH, USA).

Data analysis

Placebo data from each cohort were aggregated. AEs were summarized overall and by severity, seriousness, and relationship to treatment. All AE summaries were restricted to treatment-emergent adverse events (TEAEs; AEs that commenced on or after the start of first study drug administration). AEs without an onset date or time were defined as treatment-emergent. Subjects who experienced the same AE more than once were only counted once for that event. Change from baseline in continuous safety parameters, such as physical examination findings, laboratory parameters, vital signs or ECG intervals were summarized by descriptive statistics. Shifts from baseline in categorical safety parameters were summarized by frequencies and percentages. Where data were normally distributed, paired *t*-tests were performed to assess the percent change from baseline within treatment group. Where data were skewed, non-parametric Wilcoxon signed rank test was performed to assess the median percent change from baseline.

The safety population included all randomized subjects who received study drug or placebo. Summaries of efficacy data were performed on the per protocol population, which included all randomized subjects who completed the 4-week treatment phase and the 10-day follow up. All summary statistics are presented as mean

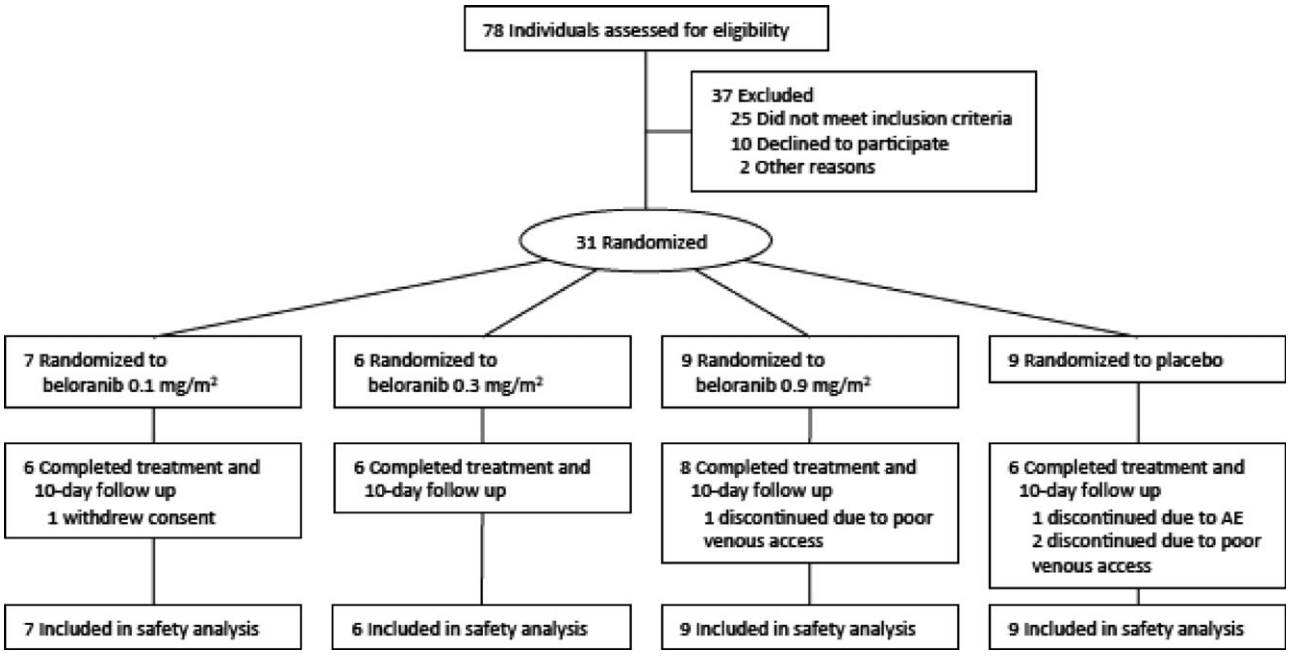

FIGURE 1 Study flow chart.

± SEM for continuous variables (weight change is also presented as median ± SEM because of skewedness) except where indicated.

Results

A total of 31 adult white females with an average BMI of 37.8 kg/m^2 and mean age range between 49.7 and 53.5 y were randomized to receive IV placebo ($n = 9$), 0.1 ($n = 7$), 0.3 ($n = 6$), or 0.9 mg/m^2 ($n = 9$) beloranib. TEAEs, bodyweight, and cardiometabolic risk factors were assessed at each visit. Treatment groups had similar demographics and baseline characteristics, with the exception of bodyweight and BMI (Table 2). Average bodyweight ranged from 96.0 kg to 105.3 kg and baseline mean BMI was higher in the 0.9 mg/m^2 group (40.3 kg/m^2) compared with the other treatment groups (36.4-37.1 kg/m^2). On average, baseline LDL-cholesterol and triglycerides tended to be high, but mean blood pressure, glucose, and other lipid values were normal (Table 2).

A total of 26 of the 31 enrolled subjects completed the study as planned, receiving all 8 infusions of study drug and completing the 10-day follow up (Figure 1). One subject in the placebo group discontinued prematurely because of a tooth abscess. Three subjects (2 placebo-, 1 beloranib-treated) discontinued because of loss of venous access. One subject (beloranib 0.1 mg/m^2) withdrew consent after five infusions for reasons unrelated to AEs.

Safety

A total of 137 TEAEs were reported by 29 (94%) of subjects; all were mild or moderate in severity and there did not seem to be any treatment group differences (Table 1). Overall, the most common TEAEs were nervous system disorders (38 events). Headache was the most frequent TEAE, with the greatest incidence occurring in the placebo group. Two subjects with a history of migraines in the 0.9 mg/m^2 cohort reported experiencing migraines of mild intensity during the study. Two subjects experienced AEs of vomiting (one in the 0.3 mg/m^2 beloranib group and one in the 0.9 mg/m^2 beloranib group) that were classified by the investigators as mild and unlikely to be related to study drug. One was associated with trial procedures (cannulation).

Six (19%) subjects experienced an AE that was considered likely related to study medication. These included nausea, dry mouth, and diarrhea ($N = 1$ for 0.1, 0.3 mg/m^2 beloranib and placebo, $n = 3$ for 0.9 mg/m^2). All were considered mild/moderate in intensity and resolved spontaneously without deviation from treatment schedule.

There were no dose-limiting toxicities associated with beloranib and no deaths. One subject in the 0.3 mg/m^2 group experienced two SAEs of moderate chest pain and back pain. Both events occurred 1 month after the final infusion of study drug and were deemed unrelated to study medication. There were no clinically significant hematology or serum chemistry findings in any subjects.

Pharmacology

Blood samples were obtained at each visit and assessed for pharmacokinetic parameters. On Day 26, plasma beloranib concentrations were generally too low in the 0.1 mg/m^2 group for determination of PK parameters other than C_{max} and T_{max} (Supporting Information Table 1). The Day 26 C_{max} for beloranib was 0.078, 0.53, and 2.1 ng/mL and the T_{max} was 1.08, 1.06, and 1.08 hours for 0.1, 0.3, and 0.9 mg/m^2 beloranib groups, respectively. The $T_{1/2}$ of beloranib was approximately 4 and 6.5 hours and the AUC_{∞} was 1.6 and 11.3 for the 0.3 and 0.9 mg/m^2 beloranib groups on Day 26. Apparent

TABLE 1 Summary of TEAEs*

	Beloranib			
	0.1 mg/m^2 $N = 7$ No (%) E	0.3 mg/m^2 $N = 6$ No (%) E	0.9 mg/m^2 $N = 9$ No (%) E	Placebo $N = 9$ No (%) E
Total TEAEs	6 (85.7) 21	5 (83.3) 33	9 (100) 42	9 (100) 41
Nervous system disorders	4 (57.1) 8	3 (50.0) 8	7 (77.8) 12	8 (88.9) 10
Dizziness	-	-	2 (22.2) 4	1 (11.1) 1
Headache	4 (57.1) 8	3 (50.0) 8	4 (44.4) 5	6 (66.7) 8
Migraine	-	-	2 (22.2) 2	-
Gastrointestinal disorders	4 (57.1) 7	3 (50.0) 5	5 (55.6) 11	4 (44.4) 7
Diarrhea	3 (42.9) 3	-	2 (22.2) 3	3 (33.3) 3
Nausea	2 (28.6) 2	3 (50.0) 3	4 (44.4) 5	2 (22.2) 2
Injury, poisoning and procedural complications	-	3 (50.0) 10	4 (44.4) 14	3 (33.3) 8
Contusion	-	3 (50.0) 5	4 (44.4) 9	2 (22.2) 5
Procedural complication	-	2 (33.3) 3	2 (22.2) 4	2 (22.2) 3
Musculoskeletal and connective tissue disorders	3 (42.9) 3	2 (33.3) 2	2 (22.2) 2	5 (55.6) 6
Musculoskeletal pain	2 (28.6) 2	-	-	-
Neck pain	-	-	-	2 (22.2) 2
Infections and infestations	-	1 (16.7) 1	1 (11.1) 1	3 (33.3) 4
Upper respiratory tract infection	-	-	-	2 (22.2) 2
Skin and subcutaneous tissue disorders	2 (28.6) 2	3 (50.0) 3	1 (11.1) 1	1 (11.1) 2

*TEAEs per System Organ Class are presented if ≥25% of subjects experienced these events. TEAEs per preferred term are presented if more than one subject experienced the same AE. No (%) indicates the number and proportion of subjects with an AE. E indicates the number of AEs.

volume of distribution (V_z) was lower at the 0.9 mg/m^2 group compared to the 0.3 mg/m^2 group.

Bodyweight

There was a dose-dependent reduction in bodyweight with beloranib (Table 2). Significant differences in bodyweight at Day 26 compared to baseline were evident with 0.3 and 0.9 mg/m^2 beloranib and placebo. Only the 0.9 mg/m^2 beloranib group experienced rapid and consistent weight loss of approximately 1 kg per week (Figure 2). Weight loss in the 0.9 mg/m^2 beloranib group was generally maintained at the follow up visit (Day 36), 10 days after the last infusion. Self-reported sense of hunger also declined by 41% with 0.9 mg/m^2 beloranib. It is possible that nausea and/or vomiting AEs can contribute to weight loss. However, exclusion of subject with more than one episode of either nausea or vomiting did not change the overall bodyweight response.

Cardiometabolic risk factors

Treatment with 0.9 mg/m^2 beloranib was associated with reductions in triglycerides and LDL-cholesterol compared to baseline (Table 2). The change in triglycerides was associated with weight loss, while the reduction in LDL appeared to be independent of weight loss (data not shown). There were no significant changes in lipid profile in the other beloranib groups (except for a 13% reduction in LDL-cholesterol with 0.3 mg/m^2 beloranib) or placebo. Glucose levels were generally unchanged. There was a nonsignificant trend for reduced blood pressure with beloranib ($P = 0.28$ and 0.34 for diastolic blood pressure in subjects treated with beloranib at 0.3 and 0.9 mg/m^2).

CRP, a marker of inflammation, was reduced rapidly during beloranib treatment. The reduction in CRP was achieved prior to appreciable weight loss, reaching a nadir at 1 week (time course not shown). Significant reductions from baseline of 42% and 68% were evident with 0.1 and 0.9 mg/m^2 beloranib and there was a nonsignificant trend ($P = 0.07$) for reduced CRP with 0.3 mg/m^2 beloranib compared to no change with placebo (Table 2).

After 4 weeks of treatment with 0.9 mg/m^2 beloranib, HMW adiponectin was increased from baseline by 85% while leptin levels were reduced by 28% and 53% in the 0.3 mg/m^2 and 0.9 mg/m^2 beloranib groups, respectively (Table 2). β-hydroxybutyrate and FGF21 concentrations were increased in the 0.9 mg/m^2 group.

Discussion

This proof-of-concept clinical trial tested the safety, tolerability, and weight-reducing effects of a novel MetAP2 inhibitor, IV beloranib (0.1-0.9 mg/m^2), for the first time in obese human subjects. Beloranib appeared to be safe. Nausea and, to a lesser extent, dizziness and migraines may have been related to beloranib. Nausea and vomiting AEs did not contribute to weight loss; vomiting was observed infrequently. The incidence of these reported TEAEs was relatively low and all were of mild to moderate intensity, self-limited, and did not result in premature withdrawal from the study. Much higher doses of beloranib (doses up to 50 mg/m^2) tested in human oncology studies conducted by another sponsor (in subjects with advanced cancer taking concomitant cytotoxic agents) have shown potential toxic effects including blood dyscrasias, electrolyte abnormalities, and liver enzyme elevations. These events were isolated to doses

TABLE 2 Percent change from baseline in bodyweight, hunger, and cardiometabolic risk factors

	Beloranib			
	0.1 mg/m^2 $N = 6$	0.3 mg/m^2 $N = 6$	0.9 mg/m^2 $N = 8$	Placebo $N = 6$
Bodyweight, % change	−0.8 (−2.0, 0.3)	−1.0 (−3.9, −0.5)[a]*	−3.5 (−4.9, −2.2)***	−0.6 (−5.5, −−0.1)[a]*
Median absolute change, kg	−0.6 (−2.9, 0.0)	−1.0 (−3.3, −0.6)*	−3.8 (−5.1, −0.9)**	−0.6 (−4.5, −0.1)*
Baseline, kg	103.9 (20.0)	100.4 (17.5)	104.9 (7.4)	94.1 (7.8)
Day 26, kg	103.0 (19.3)	99.1 (18.1)	101.3 (8.2)	92.9 (9.1)
Waist circumference, % change	−1.1 (−4.3, 2.1)	−2.5 (−5.0, −0.0)*	−2.3 (−5.4, 0.8)	−1.7 (−2.7, 6.6)[a]
Baseline, cm	118.0 (14.3)	114.2 (8.5)	124.1 (6.2)	112.5 (9.5)
Day 26, cm	116.2 (12.1)	111.2 (6.3)	121.3 (7.1)	111.8 (9.1)
Hunger, % change	−1.2 (−36.8, 34.4)	−24.1 (−48.5, 0.2)	−40.8 (−63.8, −17.9)**	−2.2 (−16.5, 12.1)
Baseline, VAS units	3.0 (2.1)	4.8 (2.4)	4.9 (1.6)	4.8 (1.6)
Day 26, VAS units	2.5 (0.8)	3.3 (1.6)	3.0 (1.9)	4.7 (1.5)
Triglycerides, % change	−4.9 (−29.0, 19.2)	−3.9 (−39.2, 31.4)	−42.0 (−60.5, −23.4)**	−15.2 (−42.7, 12.3)
Baseline, mmol/L	1.22 (0.58)	1.18 (0.55)	1.30 (0.49)	1.22 (0.50)
Day 26, mmol/L	1.07 (0.39)	1.17 (0.69)	0.80 (0.46)	1.05 (0.60)
LDL−c, % change	−3.0 (−16.5, 10.4)	−13.0 (−24.9, −1.2)*	−18.4 (−31.6, −5.2)*	2.3 (−11.1, 15.8)
Baseline, mmol/L	3.68 (1.07)	4.20 (1.03)	3.11 (0.97)	1.05 (0.60)
Day 26, mmol/L	3.55 (1.00)	3.63 (0.90)	2.44 (0.52)	3.72 (1.16)
HDL-c, % change	2.8 (−8.9, 14.5)	−6.3 (−25.2, 12.5)	6.0 (−5.5, 17.4)	0.2 (−20.7, 21.1)
Baseline, mmol/L	1.12 (0.07)	1.18 (0.39)	1.35 (0.23)	5.1 (0.5)
Day 26, mmol/L	1.15 (0.13)	1.11 (0.42)	1.42 (0.23)	5.3 (0.5)
Glucose, % change	1.2 (−7.7, 10.1)	0.8 (−7.7, 10.1)	4.2 (−2.8, 11.3)	−1.1 (−11.4, 9.3)
Baseline, mmol/L	5.08 (0.49)	5.35 (0.48)	5.14 (0.49)	4.9 (0.23)
Day 26, mmol/L	5.13 (0.52)	5.37 (0.47)	5.34 (0.47)	4.83 (0.34)
Systolic BP, % change	−1.2 (−9.2, 6.7)	−2.9 (−20.2, 14.3)	−2.2 (−11.6, 7.1)	−1.9 (−16.6, 12.9)
Baseline, mm Hg	121.8 (3.7)	133.0 (12.6)	126.4 (8.6)	127.7 (14.1)
Day 26, mm Hg	120.5 (11.3)	127.8 (14.9)	122.9 (9.6)	124.0 (11.3)
Diastolic BP, % change	−3.1 (−11.7, 5.5)	−5.8 (−18.9, 7.3)	−3.7 (−15.9, 8.5)	7.3 (−15.0, 29.6)
Baseline, mm Hg	75.2 (5.8)	81.7 (5.8)	76.9 (13.1)	75.0 (14.3)
Day 26, mm Hg	73.0 (9.9)	76.5 (7.2)	72.9 (9.8)	78.3 (6.5)
CRP, % change	−41.7 (−66.4, −16.9)*	−72.2 (−89.9, 21.8)[a]	−67.5 (−80.3, −54.7)***	16.5 (−21.0, 53.9)
Baseline, μg/mL	10.36 (3.20)	7.51 (6.17)	9.36 (7.41)	6.42 (7.25)
Day 26, μg/mL	5.94 (3.06)	2.36 (1.49)	3.33 (3.47)	5.68 (4.05)
β-hydroxybutyrate, % change	99.2 (−49.5, 248.0)	−9.4 (−33.5, 14.7)	187.5 (47.5, 327.5)*	10.1 (−9.1, 180.0)[a]
Baseline, mmol/L	0.14 (0.06)	0.17 (0.10)	0.15 (0.08)	0.13 (0.03)
Day 26, mmol/L	0.25 (0.14)	0.15 (0.07)	0.40 (0.30)	0.19 (0.12)
Leptin, % change	−11.9 (−26.0, 1.2)	−28.2 (−47.3, −9.1)*	−53.2 (−60.0, −46.4)***	−8.4 (−41.6, 24.8)
Baseline, μg/mL	71.4 (34.6)	47.4 (14.7)	65.7 (20.7)	40.4 (14.9)
Day 26, μg/mL	65.2 (37.6)	33.4 (10.6)	31.5 (13.5)	38.8 (20.4)
HMW Adiponectin, % change	–	–	85.4 (50.3, 120.4)**	−12.4 (−39.3, 14.5)
Baseline, μg/mL			4.68 (2.04)	4.48 (1.49)
Day 26, μg/mL			8.34 (3.18)	4.10 (2.20)
FGF-21, % change	–	–	123.7 (32.2, 215.1)*	41.8 (−90.0, 173.5)
Baseline, pg/mL			202.5 (142.3)	216.7 (177.4)
Day 26, pg/mL			413.2 (232.9)	176.3 (82.6)

Percent values for change from Day 26 to baseline (Day 1) are mean (95% CI), except where indicated. Baseline and Day 26 values are mean (SD). All analyses performed on the Per Protocol population. VAS hunger scores range from 1 to 10, with higher values indicating greater self-reported hunger over the 2 days preceding the assessment. HMW adiponectin and FGF-21 were only measured in 0.9 mg/m^2 and placebo groups.

[a]Because of skewness, percent change values are median change (95% CI).

*$P < 0.05$, **$P < 0.01$, ***$P < 0.001$.

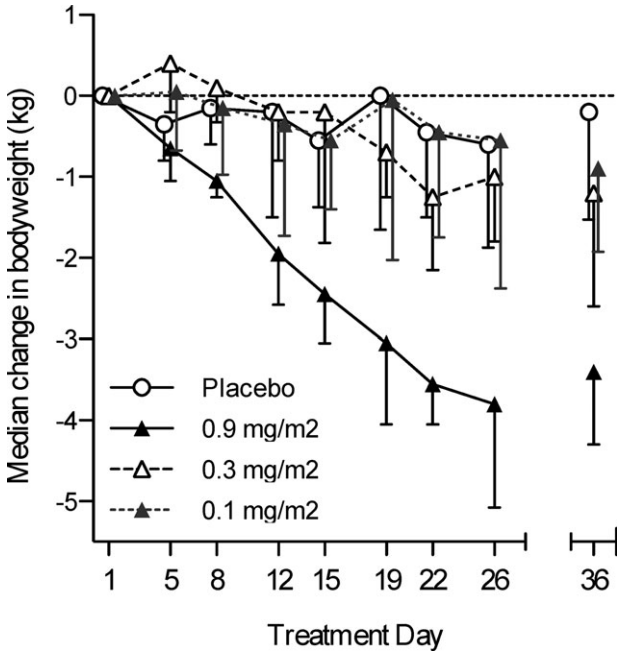

FIGURE 2 Median (interquartile range) change in bodyweight from baseline (Day 1) through 4 weeks of treatment and at follow up (Day 36). Data are for the per protocol population for 0.1 mg/m^2 beloranib ($N = 6$), 0.3 mg/m^2 beloranib ($N = 6$), 0.9 mg/m^2 beloranib ($N = 8$), and placebo ($N = 6$).

above 20 mg/m^2 per injection and have not been observed with much lower doses tested in the current study (or in animal models with human equivalent exposures). At the high doses tested in human oncology trials, no discernible anti-angiogenic effects were observed. Pharmacokinetic evaluations of beloranib revealed that T_{max} was reached at approximately 1 hour after start of infusion and $T_{1/2}$ was approximately 4 to 6.5 hours for 0.3 and 0.9 mg/m^2 beloranib, respectively. Despite the PK profile, twice weekly administration of 0.9 mg/m^2 beloranib resulted in clinically significant weight loss of approximately 1 kg/week (or 1% of bodyweight per week) lasting for the duration of treatment (4 weeks). Moreover, this weight loss was associated with meaningful improvements in cardiometabolic risk factors such as plasma TG, LDL, CRP, as well as possible improvements in systolic and diastolic blood pressure. Although this study was of limited size and duration, beloranib, as utilized in this study, results in findings consistent with clinical need for weight loss therapy with beneficial impact on cardiometabolic risk factors without any apparent clinical toxicity issues.

Although the mechanism of weight loss with beloranib is not fully elucidated, non-enzymatic actions of MetAP2 to suppress activity of extracellular signal regulated kinases 1 and 2 (ERK1/2) may be important (18). Cellular responses to MetAP2 inhibition reflective of potential ERK-related processes may include suppression of sterol regulatory element binding protein (SREBP) activity, leading to reduced lipid and cholesterol biosynthesis (19,20). Interestingly, changes in the expression patterns of hepatic and adipose tissue genes after prolonged (~9 months) fumagillin exposure suggest that MetAP2 inhibition also may alter the relative abundance of factors involved in inflammation, consistent with reduced ERK-dependent cellular processes (unpublished observations). The putative mechanism of MetAP2 inhibition leading to mobilization of adipose depot and catabolism of free fatty acids as energy source by the body is supported by the changes in plasma β-hydroxybutyrate, adiponectin, leptin, and FGF21 observed in this study. Elevation in the levels of key catabolic hormones adiponectin and FGF21, coupled with the appearance of ketone bodies (β-hydroxybutyrate), suggest MetAP2 inhibition with beloranib stimulates energy expenditure, fat utilization, and lipid excretion (21). The reduction in leptin observed in this study is also consistent with a decrease in total adipose tissue and negative energy balance (21). As would be predicted based on decreased subject-reported sense of hunger, the observed reduction in bodyweight of approximately 4 kg over 4 weeks in subjects receiving the 0.9 mg/m^2 dose is consistent with approximately 30-40% (600-800 kcal) reduction in daily food intake (22).

The effects of beloranib on bodyweight, sense of hunger and cardiometabolic risk markers may appear inconsistent with the relatively short-lived PK profile of beloranib reported here. A possible explanation for this apparent disconnect is that beloranib forms a covalent bond with MetAP2 (14), thereby irreversibly inhibiting and silencing existing enzyme until a newly produced pool of MetAP2 is generated in target tissues (e.g., liver and adipose tissue). A once weekly dosing schedule of beloranib is currently being tested in humans and may provide alternative clinical utility. Because of its high potency, beloranib can also be administered subcutaneously in a small injection volume, which is presently also being tested in obese humans.

The rate and extent of weight loss, as well as improvements in associated cardiometabolic risk factors reported in this study should be contextualized. The minimum clinically significant weight loss after 1 year of therapy (behavioral modification and/or medication) is often regarded as 3-5%, assuming risk factors such as waist circumference, blood pressure, serum lipids and inflammatory markers also improve (2,23,24). In fact, the US FDA has set 5% total bodyweight loss at 1 year as a clinical efficacy hurdle for marketing approval of drugs intended to treat obesity. This is in contrast to the approximately 20% weight loss deemed acceptable by obese women engaging in weight loss therapy (17). In the current study, beloranib resulted in median bodyweight loss of approximately 4% after 4 weeks and may have incremental lipid lowering and anti-inflammatory effects. It appears likely, but remains unknown whether these additional effects are independent of weight loss. Longer studies with beloranib will be needed to more definitively characterize its long term effects on safety, tolerability, weight, cardiometabolic risk factors, and, ultimately, on morbidity/mortality risks in obese subjects. However, the rate and extent of weight loss and associated laboratory benefits observed with the 0.9 mg/m^2 beloranib dose over 4 weeks encourage further investigation of beloranib's potential in our collective efforts to ameliorate the consequence of the obesity pandemic.

The limitations of this study are common features of early stage human clinical studies and include the following: small sample size, homogeneous population, relatively short treatment duration, lack of any diet and exercise counselling to supplement treatment effects, and no sophisticated measurement of body composition to confirm preclinical findings that MetAP2 inhibitors cause preferential reductions in adipose tissue (13). Larger studies in a more diverse population that are designed to specifically test for weight loss efficacy will better address these issues. Additional factors, including the 3-phase study design and pooled placebo group may have influenced

study outcomes. Although the 0.9 mg/m^2 beloranib group had a higher baseline bodyweight, this did not confound the overall weight response when an analysis of weight-matched subjects was conducted.

In summary, all doses of IV beloranib, administered twice weekly for 4 weeks to adult females with uncomplicated obesity, appeared to be generally safe and well tolerated. Furthermore, the 0.9 mg/m^2 beloranib dose resulted in significant weight loss and improvements in associated cardiometabolic risk factors. Other methods of beloranib administration, including greater IV doses, once weekly administration, and subcutaneous administration should be evaluated in the context of longer studies with more subjects, which will add to our growing knowledge of beloranib's potential as an anti-obesity therapy.O

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