Intranasal (IN) Pharmacokinetics (PK) and Bioavailability of ZT-031, a Novel Parathyroid Hormone (PTH) Analog

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ABSTRACT

ZT-031 (formerly cyclic PTH (1-31); Ostabolin-C™), is a novel 31 amino acid cyclic peptide analog of human PTH in late clinical development for osteoporosis and other disorders of bone. Peptide therapeutics are typically administered via subcutaneous (SC) injection, which may limit their utility.

Purpose: If feasible, IN administration would improve compliance and convenience for patients compared to daily SC injection. N-dodecyl-β-maltoside, (DDM; Intravail® A3) is a 12-carbon chain alkylglycoside compound with mucosal permeation enhancing properties. The feasibility of IN administration of ZT-031 combined with DDM was evaluated in rats, rabbits and monkeys.

Methods: Groups of 10 adult female Sprague Dawley rats were administered ZT-031 (10, 75,125, 200 µg/kg in 20 µL) with 0.18% DDM via IN drop instillation into a single nostril or by (SC) injection (75 µg/kg). In New Zealand white rabbits, ZT-031 (2, 4, 8, 20 µg/kg) with 0.18% DDM or without DDM (20 µg/kg only) was instilled by pipette into both nostrils (total dose volume 100µL). In groups of 5 adult female Cynomolgous monkeys ZT-031 (5 and 50 µg/kg in 50 µL) with 0.18%, 0.125% or 0.06% DDM was administered to a single nostril by a hand-actuated spray device; parallel groups received the same doses SC without DDM. In all species, plasma was collected for ZT-031 concentrations via validated ELISA assay pre-dose and at 5, 10, 15, 25, 45, 60, 120 and/or 180 minutes post-dose.

Results: In all three species, good systemic exposures were achieved with dose related increases in maximum plasma concentration (C_{max}) and exposure (AUC). The T_{max} in all species ranged between 5-25 minutes. The relative bioavailability (IN/SC) in the rat was 33% at the 10 µg/kg dose and increased to >100% at higher doses. In rabbits the addition of DDM increased AUC by > 2 fold at 20 µg/kg IN. In monkeys, the relative bioavailability (IN/SC) was ~40%.

Conclusions: When formulated in 0.18% DDM and dosed IN, ZT-031 demonstrated good systemic exposure in rats, rabbits and monkeys, potentially providing a feasible alternate to SC injection for patients.

OBJECTIVE

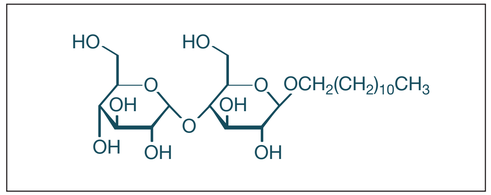
- To investigate the feasibility of intranasal administration of a novel PTH analog, ZT-031, to rats, rabbits and monkeys
- To demonstrate effective absorption enhancement by DDM in the nasal formulation, with the objective of a relative bioavailability of >20%
- If feasible, intranasal administration may improve patient compliance and convenience compared to injection

INTRODUCTION

- ZT-031 [cyclic PTH (1-31), previously known as Ostabolin-C™ is a novel PTH analog that stimulates trabecular and cortical bone growth and has been shown to be an effective anabolic bone formation agent to treat osteoporosis (Hodsman et al, ASMBR 2007)
- Like most peptide and protein drugs, ZT-031 is delivered by subcutaneous (SC) injection in all animal and human investigations. An alternative, needle-free route of administration would be highly desirable for patients and would likely improve compliance.
- Nasal administration is an accepted delivery route for both local and systemic delivery of drugs, including peptides (e.g., calcitonin). The human nasal mucosa is relatively large (~160 cm^2), permeable, highly vascular and provides rapid absorption of peptide and protein drugs and direct access to the systemic circulation, thus eliminating first-pass hepatic metabolism. However, absorption is affected by properties intrinsic to the molecule, such as molecular weight, lipophilicity, water solubility, and by certain intrinsic physiologic barriers, such as the presence of mucous and mucociliary clearance (Costantino 2007). However, peptides and proteins larger than 1000Da cannot readily permeate the nasal epithelium. Therefore, the use of permeation enhancers may be required to obtain permeation and adequate blood levels, and to improve bioavailability.
- Dodecylmaltoside (DDM, Intravail™ A-3) is a synthetic compound intended for use as an absorption enhancement agent for nasal formulations of therapeutic agents. DDM is the glycoside of maltose and the long chain alcohol dodecanol (C12). The alkylglycosides are particularly promising absorption-enhancers because they are nonionic mild surfactants, effective at low concentrations (0.03-0.25%) and metabolized to simple non-toxic metabolites. It is likely that the alkylglycosides enhance drug passage across mucosal membranes by both enhancing transcellular and paracellular mechanisms across the nasal epithelium (Maggio 2006).
- This paper summarizes the feasibility of IN administration of ZT-031 in the presence of 0.18% DDM in preclinical studies supporting the use of DDM as a permeation enhancer for the intranasal delivery of ZT-031.

N-Dodecyl-β-D-Maltoside (DDM)

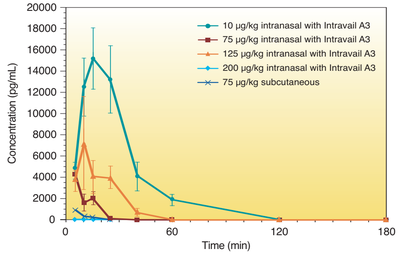
Intranasal Permeation Enhancer



ZT-031 IN PK PARAMETERS IN RATS

Test Article	Route	Dose (µg/kg)	C_{max} (pg/mL)	T_{max} (min)	$AUC_{(0-t)}$ (pg·min/mL)	$t_{1/2}$ (min)	Rel F (%)
ZT-031	SC	75	8548	5	92630	22.3	-
ZT-031 with 0.18% DDM	IN	10	564	5	4188	5.1	33.8
	IN	75	43020	5	477085	25.9	514
	IN	125	72853	10	1522443	35.8	985
	IN	200	152760	15	5178784	15.4	2091

ZT-031 PK Profile in Rats Following IN Administration with 0.18% DDM



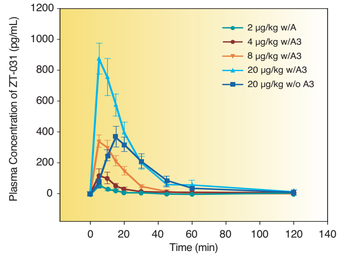
CONCLUSIONS: ZT-031 IN PK IN RATS

- Following IN or SC dosing of ZT-031, T_{max} ranged from 5 to 15 minutes
- The apparent elimination half-live ($t_{1/2}$) increased with increasing IN doses of ZT-031 (ranging from 5.1 to 35.8 minutes) when administered with 0.18% DDM. After SC dose (without DDM), $t_{1/2}$ was 22.3 minutes
- Following IN doses of ZT-031 (10 to 200 µg/kg with 0.18% DDM), the AUC_{0-t} increased in a greater than dose-proportional manner
- The relative bioavailability of ZT-031 (10, 75, 125 and 200 µg/kg), relative to a single SC injection was 33.8, 514, 985 and 2091%, respectively
- In rats, systemic exposures which would be considered therapeutic in humans (C_{max} : ~159 pg/mL; AUC_{0-t} : 9900 pg*min/mL) were achieved

ZT-031 IN PK PARAMETERS IN RABBITS

Dose ZT-031	AUC (0-t) (pg*min/mL)	C_{max} (pg/mL)	$t_{1/2}$ (min)	T_{max} (min)
2 µg/kg w/ 0.18% DDM	475	53	8	5
4 µg/kg w/ 0.18% DDM	2242	117	34	5
8 µg/kg w/ 0.18% DDM	5365	339	8	5
20 µg/kg w/ 0.18% DDM	17702	874	18	5
20 µg/kg w/o DDM	10974	369	19	15

ZT-031 IN PK profile in Rabbits



ZT-031 IN PK PROFILE IN RABBITS

- Dose proportional response seen with C_{max} and AUCs
- IN dosing of 20 µg/kg with 0.18% DDM causes a greater than 2-fold enhanced exposure compared to IN dosing at the same dose without DDM
- The t_{max} in the presence of 0.18% DDM is 5 min and is longer (15 min) in the absence of DDM

DISCUSSION AND CONCLUSIONS

- Intranasal Delivery of ZT-031 is a feasible route of drug administration
- DDM is an effective nasal absorption enhancer at 0.18%
- Rapid onset of plasma levels were achieved in rats, rabbits and monkeys, T_{max}, 5- 15 min with 0.18%
- Good systemic exposures were observed in all species with relative bioavailability in primates between 35-40%
- Human therapeutic levels were achieved in all animal species
- Based on the above, formulations of PTH analogs with DDM at 0.18% seem appropriate for human clinical trials, pending the results of preclinical toxicology studies

REFERENCES

1. A. Hodsman, H. Bone, C. Gallagher, D. Kendler, J. P. Brown, S. Greenspan, C. L. Barclay, P. Morley, R. Anderson, B. R. MacDonald. Ostabolin-C™ Increases Lumbar Spine and Hip BMD after 1 Year of Therapy: Results of a Phase II Clinical Trial. ASBMR presentation #1130. In osteoporosis Treatment (Clinical) III: New Approaches, *JBMR Abstracts* 2007
2. Costantino HR, Illum L, Brandt G et al. Intranasal delivery: physiochemical and therapeutic aspects. *Int J Pharma* 2007;337:1-34.
3. Maggio ET. Intravail™: highly effective intranasal delivery of peptide and protein drugs. *Expert Opin Drug Deliv* 2006; 4:529-39.

ZT-031 IN PK PARAMETERS IN MONKEYS

Route	Dose (µg/kg)	$t_{1/2}$ (min)	C_{max} (pg/mL)	T_{max} (min)	$AUC_{(0-t)}$ (pg.min/mL)	AUC_{0-inf} (pg.min/mL)	Rel F% (AUC_{0-t})
IN	5	nc	145 ± 45	15	8827 ± 4468	nc	39.4
IN	50	33.0 ± 4.8	2718 ± 637	15	114 419 ±54 072	115 509 ±62 324	38.5
SC	5	37.3 ±21.5	664 ±231	10	31 263 ± 12 244	34 946 ± 15 579	N/A
SC	50	29.9 ±8.8	7648 ±4002	5	278 000 ± 149 275	289 521 ± 154 121	N/A

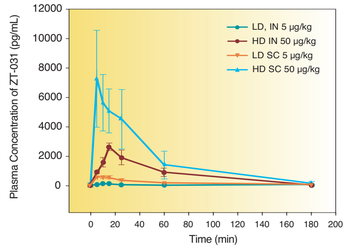
ZT-031 IN PK PARAMETERS IN THE PRESENCE OR ABSENCE OF DDM IN MONKEYS

Route	DDM %	Dose µg/kg	$t_{1/2}$ (min)	T_{max} (min)	C_{max} (pg/mL)	$AUC_{(0-t)}$ (pg.min/mL)	Rel F% (AUC_{0-t})
IN	0	50	nc	15	115 ± 1	14983 ± 2451	5.2
IN	0.06	50	nc	15	287± 277	17885 ± 9901	6.2
IN	0.125	50	45.3	25	590 ± 707	32529 ± 31411	11.2

nc: not calculated as not enough point in the terminal phase or as $AUC_{extrapolated}$ higher than 20%. IN: intranasal; SC: subcutaneous. N/A: Not Applicable

Relative Bioavailability (Rel F%) = (($AUC_{inf\ IN}$ x$Dose_{sc}$) / ($AUC_{inf\ SC}$ x $Dose_{IN}$)) x 100

ZT-031 IN PK Profile in Monkeys



ZT-031 IN PK PROFILE IN MONKEYS

- The relative intranasal bioavailability of ZT-031 compared to subcutaneous injection ranged between 35-40% (AUC) with 0.18% DDM
- Good systemic exposure of ZT-031 was achieved following IN dosing with dose proportionality among the low and high dose groups (AUC)
- T_{max} was rapid, within 15 minutes of dosing
- $t_{1/2}$ (half life) following both IN and SC dosing ranged ~ 1-2 h
- Increasing the concentration of DDM yields enhanced absorption in a non-dose proportional manner

METHODS - RAT STUDIES

- Groups of 10 adult female Sprague Dawley rats were administered ZT-031 (10, 75, 125, 200 µg/kg in 20 µL) with 0.18% DDM via IN drop instillation into a single nostril or 100 uL by (SC) injection (75 µg/kg)
- ZT-031 was administered in 10 mM Sodium Acetate buffer, 0.1% EDTA at pH 5.0 with 0.18% DDM for the IN administration and in a solution of Acetate Buffer at pH 4.0 for the SC injection
- Blood samples (350 µL) were drawn by orbital bleed over a three hour time period at 0, 5, 10, 15, 25, 40, 60, 120, and 180 minutes following either IN or SC administration of ZT-031
- Following blood collection, plasma was immediately prepared from each blood sample using K2EDTA as the anticoagulant. Plasma PTH 1-34 was analyzed using an ELISA method.
- The group mean plasma concentration-time data were used to determine the composite pharmacokinetic parameters. Noncompartmental pharmacokinetic parameters were determined using WinNonlin™, version 4.1 (WinNonlin™ Professional Edition Copyright ©2003, Pharsight Corporation, Mountain View, CA).

METHODS - RABBIT STUDIES

- Groups of 5 male New Zealand white rabbits (Ramona, CA) were administered 100 µL IN instillations to a both nares (50 µL per nare) under mild anesthesia.
- ZT-031 was administered in 10 mM Sodium Acetate buffer, 0.1% EDTA at pH 5.0 with 0.18% DDM
- Blood samples (1 mL each time) were drawn from the ear vein over a two hour time period at 0, 5, 10, 15, 20, 30, 45, 60, and 120 minutes following IN administration and collected in lithium/heparin anticoagulant-treated tubes and centrifuged immediately for plasma preparation. Plasma PTH 1-34 was analyzed using a validated ELISA method.
- The individual plasma concentration-time data at each dose level was used to calculate composite profiles to be used in the calculation of pharmacokinetic parameters of ZT-031 using Pharmacokinetic function component in Excel 2007

METHODS - MONKEY STUDIES

- Two groups of 5 adult female Cynomolgus monkeys per group were administered ZT-031 in a 3-period crossover design.
- In the first period, ZT-031 was dosed by IN administration with 5 or 50 µg/kg ZT-031 with 0.18% DDM in 50 µL using a hand-help spray device into a single nostril followed by a 1-week wash out period
- In the second period, ZT-031 was dosed with 5 or 50 µg/kg in 0.2 mL/kg by SC injection without DDM followed by a 1-week wash out period
- In the third period, 10 animals in 3 groups of 2 or 3 animals received 50 µg/kg ZT-031 with 0, 0.06 or 0.125% DDM
- Blood samples (1 mL) were drawn over a three hour time period at 0, 5, 10, 15, 25, 60 and 180 minutes following either IN or SC administration of ZT-031, collected in K2EDTA as the anticoagulant. Plasma ZT-031 was analyzed using validated ELISA methods
- The plasma concentration-time data were used to determine the pharmacokinetic parameters. Non-compartmental pharmacokinetic parameters were determined using WinNonlin™, version 4.1 (WinNonlin™ Professional Edition Copyright ©2003, Pharsight Corporation, Mountain View, CA).

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