CENTER FOR DRUG EVALUATION AND RESEARCH

APPLICATION NUMBER:

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NON-CLINICAL REVIEW(S)

DEPARTMENT OF HEALTH AND HUMAN SERVICES
PUBLIC HEALTH SERVICE
FOOD AND DRUG ADMINISTRATION
CENTER FOR DRUG EVALUATION AND RESEARCH

PHARMACOLOGY/TOXICOLOGY NDA/BLA REVIEW AND EVALUATION

Application number:	NDA 209296
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Applicant's letter date:	January 12, 2017
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Product:	Cinvanti (Aprepitant) Injectable Emulsion
Indication:	Prevention of chemotherapy Induced Nausea and Vomiting
Applicant:	Heron Therapeutics, Inc.
Review Division:	Division of Gastroenterology and Inborn Errors Products (DGIEP)
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1 Executive Summary

1.1 Introduction

1.2 Brief Discussion of Nonclinical Findings

For the nonclinical safety of Cinvanti, the applicant relied on the Agency's determination of the safety of the listed drug Emend (aprepitant capsules and fosaprepitant IV). In addition, the sponsor has submitted reports of pharmacokinetic (PK) studies in rats, a repeat dose toxicology study in rats, a local tolerance study in rabbits and an *in vitro* hemolysis study with rat, dog and human blood, conducted with the proposed drug product. Cinvanti is an oil-in-water emulsion of aprepitant for intravenous (IV) use and is indicated for the prevention of acute and delayed chemotherapy-induced nausea and vomiting (CINV).

When groups of male rats were administered single doses of Cinvanti or Emend IV at 2 mg/kg, the exposure to aprepitant in rats dosed with Cinvanti was greater than the animals administered Emend IV. The half-life of aprepitant following administration of Emend IV was longer, while the half-life of aprepitant following administration of Cinvanti was similar to that reported in the other single dose study. Also, a comparison of Cinvanti emulsion or Emend IV to Cinvanti in a low sodium oleate (SO) vehicle emulsion, at a dose of 2 mg/kg in male Sprague-Dawley rats, showed that all the three formulations have similar elimination profiles in rats. The plasma protein binding was similar for samples obtained from animals dosed with the two formulations.

In a 28-day intravenous toxicology study in rats, Cinvanti and Emend IV had similar toxicology profiles. The increases in C_{max} and AUC for aprepitant following administration of Cinvanti were greater than dose proportional. Microscopic findings of mild to minimal periportal vacuolation of the liver was seen mostly in female rats dosed with Emend IV and the mid and high doses of Cinvanti. After the 28-day recovery period, there was a lower incidence and/or severity of infusion site changes and no changes were observed in the liver of the recovery animals.

No genotoxicity studies of Cinvanti have been performed. However, aprepitant and fosaprepitant were not genotoxic in the Ames test, the human lymphoblastoid cell (TK6) mutagenesis test, the rat hepatocyte DNA strand break test, the Chinese hamster ovary (CHO) cell chromosome aberration test and the mouse micronucleus test.

Two year oral carcinogenicity studies were conducted with aprepitant in rats and mice by the innovator. In a 104-week oral carcinogenicity study in Sprague-Dawley rats, aprepitant was administered at 0.05 to 1,000 mg/kg twice daily. Treatment with aprepitant at doses of 5 to 1,000 mg/kg twice daily caused an increase in the incidences of thyroid follicular cell adenomas and carcinomas in male rats. In female rats, it produced hepatocellular adenomas at 5 to 1,000 mg/kg twice daily and hepatocellular carcinomas and thyroid follicular cell adenomas at 125 to 1,000 mg/kg twice daily.

In a 2-year mouse carcinogenicity study, CD-1 mice were treated with oral doses of aprepitant at 2.5 to 2,000 mg/kg/day. Treatment with aprepitant produced skin fibrosarcomas at 125 and 500 mg/kg/day doses in male mice.

Aprepitant did not affect fertility or general reproductive performance of male or female rats at oral doses up to the maximum feasible dose of 1,000 mg/kg twice daily.

In a local tolerance study, Cinvanti was compared to Emend IV after IV administration of a 1 ml/kg dose into the marginal ear vein of New Zealand White rabbits. Injection sites dosed with IV Emend had higher overall scores for endothelial cell loss, edema, hemorrhage, and inflammatory cell infiltrates than sites dosed with IV saline, while the rabbits dosed with Cinvanti emulsion had lesser or equal scores compared to the findings in the saline control rabbits.

Cinvanti or fosaprepitant did not result in hemolysis in rat, dog or human blood in an in vitro hemolysis study.

1.3 Recommendations

1.3.1 Approvability

No nonclinical approvability issue has been identified.

1.3.2 Additional Non Clinical Recommendations

None

1.3.3 Labeling

In this 505(b)(2) NDA submission, nonclinical sections of the labeling are adopted from the innovator's labeling of Emend (fosaprepitant) for injection (NDA 022023). However, the following changes in the proposed labeling are recommended to comply with the PLLR.

8. USE IN SPECIFIC POPULATIONS

8.1 Pregnancy

Risk Summary

Proposed Version:

(b) (4)

The estimated background risk of major birth defects and miscarriage for the indicated populations is unknown. In the U.S. general population, the estimated background risk of major birth defects and miscarriage in clinically recognized pregnancies is 2% to 4% and 15% to 20%, respectively.

Evaluation: The text is not in accordance with 21CFR 201.57(f)(6)(i)(c), and should be modified as proposed (developed in collaboration with the DPMH reviewer) below.

8.1 Pregnancy

Recommended Version:

Risk Summary

There are no available data on CINVANTI use in pregnant women to inform a drug associated risk of adverse developmental outcomes. Avoid use of CINVANTI in pregnant women due to the alcohol content (see Clinical Considerations). In animal reproduction studies, no adverse developmental effects were observed in rats or rabbits exposed during the period of organogenesis to systemic drug concentrations (area under the plasma-concentration time curve [AUC]) of aprepitant approximately equivalent to the exposure at the recommended human dose (RHD) of CINVANTI 130 mg *(see Data)*

The estimated background risk of major birth defects and miscarriage for the indicated populations is unknown. All pregnancies have a background risk of birth defects, loss, or other adverse outcomes. In the U.S. general population, the estimated background risk of major birth defects and miscarriage in clinically recognized pregnancies is 2 to 4% and 15 to 20%, respectively.

8.2 Lactation

Proposed Version:

Risk Summary

(b) (4)

Evaluation: No changes are recommended for this section.

13 NONCLINICAL TOXICOLOGY

13.1. CARCINOGENESIS, MUTAGENESIS, IMPAIRMENT OF FERTILITY

Proposed Version:

(b) (4)

Mutagenesis

(b) (4)

Impairment of Fertility

(b) (4)

Recommended Version:

Carcinogenicity studies were conducted in Sprague-Dawley rats and CD-1 mice for 2 years. In the rat carcinogenicity studies, animals were treated with oral doses ranging from 0.05 to 1000 mg/kg twice daily. The highest dose produced systemic exposures to aprepitant approximately equivalent to (female rats) or less than (male rats) the human exposure at the CINVANTI RHD of 130 mg. Treatment with aprepitant at doses of 5 to 1000 mg/kg twice daily caused an increase in the incidences of thyroid follicular cell

adenomas and carcinomas in male rats. In female rats, it produced hepatocellular adenomas at 5 to 1000 mg/kg twice daily and hepatocellular carcinomas and thyroid follicular cell adenomas at 125 to1000 mg/kg twice daily. In the mouse carcinogenicity studies, the animals were treated with oral doses ranging from 2.5 to 2000 mg/kg/day. The highest dose produced a systemic exposure approximately 2 times the human exposure at the RHD of CINVANTI 130 mg. Treatment with aprepitant produced skin fibrosarcoma at 125 and 500 mg/kg/day doses in male mice.

Mutagenesis

Aprepitant was not genotoxic in the Ames test, the human lymphoblastoid cell (TK6) mutagenesis test, the rat hepatocyte DNA strand break test, the Chinese hamster ovary (CHO) cell chromosome aberration test and the mouse micronucleus test.

Impairment of Fertility

Oral aprepitant did not affect the fertility or general reproductive performance of male or female rats at doses up to the maximum feasible dose of 1000 mg/kg twice daily (providing exposure in male rats lower than the exposure at the RHD of CINVANTI 130 mg and exposure in female rats approximately equivalent to the human exposure).

2 Drug Information

2.1 Drug

Cinvanti

CAS Registry Number
170729-80-3

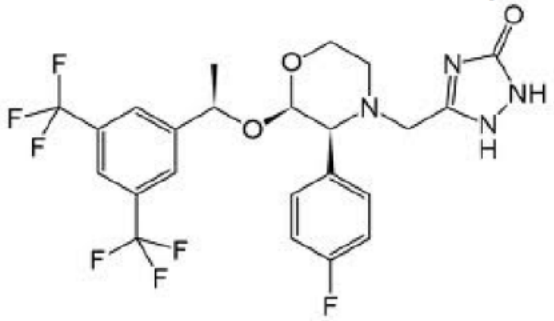
Generic Name
Aprepitant

Code Name
HTX-019

Chemical Name
3-{[(2R,3S)-2-[(1R)-1-[3,5-bis(trifluoromethyl)phenyl]ethoxy]-3-(4-fluorophenyl)morpholin-4-yl]methyl}-4,5-dihydro-1,2,4-triazol-5-one

Molecular Formula/Molecular Weight
$C_{23}H_{21}F_7N_4O_3$ / 534.427

Structure or Biochemical Description



Pharmacologic Class
Substance P/Neurokinin 1 (NK1) receptor antagonist

2.2 Relevant INDs, NDAs, BLAs and DMFs

NDA 22023, Emend (fosaprepitant) IV; NDA 21549, Emend (aprepitant) capsules

2.3 Drug Formulation

Composition of Cinvanti

Component	Concentration (w/w%)	Concentration (mg/mL)	To Deliver Concentration (g/18 mL)	Function	Reference to Quality Standard
Aprepitant	(b) (4)			Drug Substance	Manufacturer
Egg Lecithin, (b) (4)				(b) (4)	NF
Soybean Oil					USP
Ethanol					USP
Sucrose					NF
Sodium Oleate					NF
Water for Injection					USP
Total	100%	(b) (4)	18.2 g	–	–

2.4 Comments on Novel Excipients

There are no novel excipients in the drug formulation.

2.5 Comments on Impurities/Degradants of Concern

There was no degradation product in Cinvanti (aprepitant) injectable emulsion at levels (b) (4) %, and therefore, no identification work were performed. The following (b) (4)

(b) (4) in the manufacturing process were controlled at not more than (NMT) the indicated levels: (b) (4)

2.6 Proposed Clinical Population and Dosing Regimen

Cinvanti injectable emulsion is proposed to be used in adults (≥ 18 years of age), in combination with other antiemetic agents, for the prevention of acute and delayed nausea and vomiting associated with initial and repeat courses of highly emetogenic cancer chemotherapy (HEC), including high-dose cisplatin. Cinvanti is also proposed for use in the prevention of (b) (4) nausea and vomiting associated with initial and repeat courses of moderately emetogenic cancer chemotherapy (MEC). The proposed dosage of Cinvanti injectable emulsion is 130 mg for HEC (single dose regimen) or 100 mg for MEC (3-day dosing regimen), administered on Day 1 as intravenous infusion over 30 minutes, approximately 30 minutes prior to chemotherapy.

2.7 Regulatory Background

The sponsor was advised by the FDA in a type-B teleconference pre-IND meeting on May 13, 2015 to conduct an *in vitro* hemolysis study with the drug product, and also to justify the safety of the excipients in the drug product.

3 Studies Submitted

3.1 Studies Reviewed

Pharmacokinetic Studies (ADME)

1. Pharmacokinetics and Ex Vivo Protein Binding Comparison of Emend Fosaprepitant and HTX-019 Emulsion in Male Rats (Study No. 33-21)

2. Single Dose Pharmacokinetic of HTX-019 and HTX-019 + Dexamethasone in Sprague-Dawley Rats (**Study No. PHD-53**)
3. Single Dose Pharmacokinetics of HTX-019 and Aprepitant in a Low Sodium Oleate Vehicle Emulsion (**Study No. PHD-84**)

Toxicology

Repeat Dose Toxicology Study

1. HTX-019: A 28-Day Repeat Dose Intravenous Toxicity Study with 28-Day recovery period in Sprague-Dawley Rats (**Study No. 33-26**)

Local Tolerance Study

1. Effects of HTX-019 and Emend IV in the Rabbit Ear Phlebitis Model after a Single Intravenous or Perivascular Administration. (**Study No. 33-001**)

Other Toxicity Study

1. Hemolysis Screening of Test Article HTX-019 (**Study No. 1176-R1**)

3.2 Studies Not Reviewed

N/A

3.3 Previous Reviews Referenced

The nonclinical studies listed above were previously reviewed under IND 125926 for HTX-019 (Aprepitant) injection, and were copied from the IND review.

4 Pharmacology

No pharmacology studies were submitted

5 Pharmacokinetics/ADME/Toxicokinetics

5.1 PK/ADME

Pharmacokinetics and Ex Vivo Protein Binding Comparison of Emend Fosaprepitant and HTX-019 Aprepitant Emulsion in Male Rats (Study No. 33-21)

Two groups of male Sprague-Dawley rats, 6/group (average weight 300 g) were each administered a single IV bolus dose of Emend fosaprepitant or HTX-019 at 0.5 mg per rat using a dose volume of 0.5 ml/rat and a concentration of 1 mg/ml for each formulation. The test articles were diluted in saline. Blood samples were collected from 3 rats /group at 2, 10, 20, 30, 40 minutes, 1, 1.5, 2, 3, 5, 12, and 18 hours post-dose. Plasma from the collected blood was analyzed for total aprepitant concentration, using LC-MS/MS. The remaining plasma was used to evaluate protein binding of aprepitant by equilibrium dialysis.
Results: Plasma concentrations and PK parameters were similar for rats administered single IV doses of fosaprepitant (group 1) or HTX-019 emulsion (group 2). In each group, plasma concentrations were highest at the time of the first sample collection, and

declined through 18 hours post-dose, with a mean C_{max} of 851 ± 15.8 for group 1 and a C_{max} of 1053 ± 234 for group 2. The AUC was 1733 h•ng/ml for group 1 and 1953 h•ng/ml for group 2. Clearance was 289 ml/h for group 1 and 256 ml/h for group 2. Vd_{ss} averaged 1426 ml for group 1 and 1294 ml for group 2. The terminal half-life was 3.07 h for group 1 and 3.59 h for group 2.

Plasma protein binding appeared to be concentration independent at concentrations of aprepitant ranging from 4.498 to 864.248 ng/ml after IV dosing of fosaprepitant, and from 6.123 to 1212.693 ng/ml after IV dosing of HTX-019. Protein binding was similar for samples in animals dosed with the two formulations. The overall mean percent unbound was 0.67 ± 0.12 % after IV dosing of Emend and 0.70 ± 0.19 % after IV dosing of HTX-019. A summary of the PK results are shown in the sponsor's table below.

Mean Plasma Concentrations and PK Parameters in Rats Dosed with Fosaprepitant and HTX-019

	Plasma Aprepitant (ng/mL)			
	Emend		HTX-019	
Time (min)	Mean	SD	Mean	SD
2	851.063	15.863	1052.630	234.366
10	389.534	42.877	594.096	32.356
20	408.955	56.369	458.936	45.772
30	287.768	70.610	352.110	39.448
40	273.557	3.576	306.180	5.968
60	233.915	32.193	270.978	27.391
90	176.208	16.483	205.804	31.007
120	201.636	37.564	215.812	5.062
180	149.767	27.125	164.137	7.344
300	124.650	27.585	131.627	16.461
480	76.317	0.602	80.019	21.527
720	42.565	1.689	45.140	10.165
1080	6.522	1.878	10.483	5.051
AUC_{last} (h•ng/mL)	1704	92[a]	1899	103[a]
AUC_{inf} (h•ng/mL)	1733		1953	
$t_{1/2}$ (h)	3.07		3.59	
CL (mL/h)	289		256	
Vd_{ss} (mL)	1426		1294	

[a] SE, determined in Phoenix WinNonlin using sparse sampling analysis

In conclusion, the administration of Emend IV (fosaprepitant) or HTX -019 at 0.5 mg dose to rats resulted in PK parameters that were not significantly different in rats dosed with the two formulations. The assessment of plasma protein binding in rats showed that the overall mean percent unbound was 0.67 ± 0.12 % after IV dosing of Emend and 0.70 ± 0.19 % after IV dosing of HTX-019, indicating similarity in protein binding by the two formulations.

Single Dose Pharmacokinetic of HTX-019 and HTX-019 + Dexamethasone in Sprague-Dawley Rats (**Study No. PHD-53**)

Four groups of male Sprague-Dawley rats (6/group), 6-9 weeks of age and weighing approximately 250 g were administered a single IV dose of 0.22 mg/kg Dexamethasone phosphate (DP) alone (group 1), 2 mg/kg HTX-019/0.22 DP, or 2 mg/kg HTX-019 (groups 2 and 3), or Emend IV (group 4) via the tail vein. HTX-019, aprepitant or DP were all dosed at a volume of 0.5 ml. The test articles were diluted in saline. Blood samples were collected via jugular vein from 3 rats/group/time point from rats (1-3) at 2, 20, 40, 60 minutes, 1.5, and 18 hours; and from rats (4-6) dosed with each test article at 10, 30, minutes, 2, 3, 5, 8, and 12 hours. Plasma collected from groups 1 and 2 rats were analyzed for DP, while plasma from groups 3 and 4 rats was analyzed for aprepitant (and group 4 also for fosaprepitant) by LC/MS/MS.

Results: The exposure to aprepitant in rats dosed HTX-019 was greater than the exposure from Emend IV because the PK of aprepitant in Emend IV is dependent on the conversion of the pro-drug fosaprepitant to aprepitant. The delayed conversion to aprepitant and the rapid systemic elimination of fosaprepitant accounts for the reduced exposure to aprepitant in the Emend IV formulation. However, the PK of DP was not affected when administered in combination with HTX-019. The half-life for aprepitant in Emend IV was longer, while the half-life for aprepitant in HTX-019 was similar to the half-life reported in the single-dose study. A summary of the PK results are shown in the sponsor's table below.

PK Parameters for HTX and Dexamethasone in Male SD Rats

Group	Cmax (ng/mL)	Tmax (hrs)	Half-life (hrs)	AUC_{last} (ng8hr/mL)	AUC_{0-6} (ng*hr/mL)
Group 1 Dexamethasone	144.67	0.50	3.70	511.10	403.64
Group 2 HTX-019 + Dexamethasone	145.67	0.03	3.56	496.69	376.65

PK Parameters for HTX-019 with Dexamethasone and Emend IV in Male SD Rats

Group	C_{max} (ng/mL)	T_{max} (hrs)	Half-life (hrs	AUC_{last} (ng*hr/ml)	AUC_{0-6hrs} (ng*hr/ml)
Group3 HTX-019 + Dexamethasone	1333.33	0.03	6.57	3909.45	2188.90
Group 4 EMEND® IV	985.72*	0.03	7.48	2952.16	1808.59

*adjusted for fosaprepitant contribution

Single Dose Pharmacokinetics of HTX-019 and Aprepitant in a Low Sodium Oleate Vehicle Emulsion (Study No. PHD-84)

The purpose of the study was to compare the PK of aprepitant in HTX-019 and in a low sodium oleate (SO) vehicle emulsion in SD rats.

Three groups of male Sprague-Dawley rats (6/group), 6-9 weeks of age and weighing approximately 250 g were administered single IV dose of 2 mg/kg Emend IV or fosaprepitant dimeglumine (group 1), 2 mg/kg HTX-019 (group 2), or 2 mg/kg aprepitant in a low SO vehicle emulsion (group 3) via the tail vein. All the test articles were dosed at a volume of 1 mg/ml. Blood samples were collected via jugular vein from 3 rats/group/time point from rats (1-3) dosed with each test article at 2, 20, 40, minutes, 1.5, 3, 8 and 18 hours; and from rats (4-6) dosed with each test article at 10, 30, 60 minutes, 2, 5, and 12 hours. Plasma collected from the treated rats was analyzed for the concentration of the test articles by LC/MS/MS.

Results: The C_{max} for aprepitant was higher for HTX-019 than for the low SO aprepitant formulation, whereas the AUC and half-life were similar for the two formulations. The PK parameters measured for the Emend IV dosed rats were out of the expected range due to a dosing error and were discarded. A summary of the PK are shown in the sponsor's table below.

PK Parameters for HTX-019 vs. the low SO vehicle Emulsion in Male SD Rats

Group	Half-Life (hrs)	T_{max} (hrs)	C_{max} (ng/mL)	AUC_{last} (ng*hr/mL)	AUC_{INF} (ng*hr/mL)	MRT_{last} (hrs)
HTX-019	3.66	0.03	3116.67	2336.36	2548.23	3.07
Low SO emulsion	4.07	0.03	1870.00	2149.44	2425.92	3.51

In conclusion, the administration of 2 mg/kg aprepitant in HTX-019, or in the low SO vehicle emulsion in male SD rats results in a similar elimination profile.

6 General Toxicology

6.2 Repeat-Dose Toxicity

Study title: HTX-019: A 28-Day Repeat Dose Intravenous Toxicity Study with 28-Day recovery period in Sprague-Dawley Rats.

Study no.:	33-26
Study report location:	Pages 1 to 1323 of the electronic submission
Conducting laboratory and location:	(b) (4)
Date of study initiation:	July 6, 2015
GLP compliance:	OECD Principles of Good Laboratory Practice.
QA statement:	Yes
Drug, lot #, and % purity:	HTX-019, lot # ENG 1505 (purity not given); Emend IV, lot # LO11518, LO10591 (purity not given); Saline, lot # WSD15AO; HTX-019 placebo, lot # 062415.

Key Study Findings

Administration of Emend IV (7.2 mg) and HTX-019 (7.2 mg) by repeated IV injections to Sprague-Dawley rats for 28 days followed by 28 day recovery period was associated with random death or morbidity at all doses. The cause of death or morbidity was the systemic spread of procedure-related (catheter infusion protocol) inflammatory and/or thrombotic lesions at the infusion site. Minimal to mild periportal vacuolation was observed in the liver of female animals that were treated with Emend IV and mid (2.4 mg) and high (7.2) dose HTX-019 and survived till termination. After a 28 day recovery period, the liver periportal vacuolation finding was not seen in the recovery animals, suggesting reversibility of the effect. Also, a lower incidence and/or severity of the procedure-related macroscopic/microscopic findings were noted at the infusion site as well as in other organs and tissues of the recovery animals, when compared to those observed in the main study animals, indicating the likelihood of progressive ongoing reversal of effect with a longer recovery period.

Methods

Doses:	Saline control (group 1), vehicle control (group 2), aprepitant 7.2 mg (comparator Emend IV, group 3), 0.72 mg (HTX-019 low-dose, group 4), 2.40 mg (HTX-019 mid-dose, group 5) and 7.2 mg (HTX-019 high-dose, group 6).
Frequency of dosing:	Daily for 28 days
Route of administration:	Intravenous injection (slow bolus)
Dose volume:	1 ml
Formulation/Vehicle:	HTX-019/ (b) (4), soybean oil, ethanol, sucrose and sodium oleate
Species/Strain:	Crl:CD (SD) Sprague-Dawley rats
Number/Sex/Group:	10/sex/group
Age:	9-10 weeks old
Weight:	282-390 g (males), 182-253 g (females)
Satellite groups:	3 rats/sex from group 1 (saline control), and 6 rats/sex from groups 2 to 6 were designated as TK animals.
Unique study design:	None
Deviation from study protocol:	On several occasions, the dose formulation was outside of the acceptance criteria ± 10 %. On several occasions, observations for clinical signs on some animals were either performed in the afternoon rather than in the morning, or were performed twice on the same day for one animal, or were not performed at all on the scheduled day. The terminal blood was collected from the main study animal's abdominal aorta instead of from cardiac puncture. The deviations did not affect the study results.

Observations and Results

Mortality

Mortality checks were performed at least once a day

Two Emend IV dosed animals (1 main study, and 1 recovery), and two low dose TK animals were found dead between treatment days 13 and 26. There were no adverse clinical observations in the animals prior to death. Two high dose TK animals and four recovery (1 vehicle control, 1 comparator, 1 mid dose and 1 high dose) animals were euthanized prematurely due to poor condition between treatment days 13 and 26. The animals euthanized prematurely exhibited decreased activity, whole body cold to touch, pale limbs and pinna, slightly increased respiration, moderately thin body and skin turgor, extreme swelling at the right hind limb, slight lump and crust present at the lumbar area, labored respiration and moderate swelling at the surgical catheter implant

site, moderately distended abdomen, wet fur at urogenital area, loss of hind limb function, red material in dry cage and hunched posture.

Clinical Signs

All animals were examined once daily during the pretreatment period, and at least once daily during the treatment and recovery periods.

There were no test article-related clinical signs observed in the surviving animals during the treatment and recovery periods. Clinical signs of skin discoloration (left/right axillary, or ventral/dorsal thorax), swelling at the surgical site (dorsal or neck area), and hair loss were observed during pre-treatment, and/or recovery periods, and were scattered throughout the study animals.

Body Weights

Individual animal body weights were recorded once prior to treatment initiation and weekly during the treatment and recovery periods.

There were no test article (HTX-019) or comparator (Emend IV) related effects noted on group mean body weight values of animals, when compared to the saline or vehicle control values during the treatment or recovery period. The occasional body weight increases noted had no significance since they were observed in the vehicle control comparator and test groups, when compared to the saline controls.

Feed Consumption

Food consumption by each animal was recorded weekly during the treatment and recovery periods.

There were no test article (HTX-019) or comparator (Emend IV) related effects seen on Group mean food consumption values, since all the values were comparable to the vehicle and saline control values over the treatment and recovery periods.

Ophthalmoscopy

Ophthalmic examination of all the animals were conducted pre-dose and during week 4 of the treatment period.

There were no test article (HTX-019) or comparator (Emend IV) related ophthalmic changes as most of the end of dosing findings were also observed during the pre-treatment period in the animals or were considered as background changes.

ECG

No ECG evaluations of the animals were performed.

Hematology

Blood samples for hematology were collected from surviving animals by cardiac puncture at the end of the treatment and recovery periods.

There was no test article (HTX-019) related change in hematology parameters during the treatment and recovery periods.
The occasional significant changes observed (on day 29) in mean values such as, hemoglobin (HBG) levels, mean corpuscular volume (MCV), mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) in the female group 3 (comparator Emend IV), female group 6 (High dose HTX-019), and platelet counts for group 3 females (comparator), group 4 (HTX-019 low dose) and group 6 (high dose HTX-019), were of low magnitude, and were not dose related.
There were no test article (HTX-019) or comparator (Emend IV) related effects on the measured coagulation parameters.

Clinical Chemistry

Blood samples for clinical chemistry were collected from surviving animals by cardiac puncture at the end of the treatment and recovery periods.

There were only normal variations in serum chemistry parameters measured in animals during the treatment and recovery periods. Occasional significant changes such as TP levels seen in group 6 animals, globulin levels and A/G ratios seen in groups 3, 5 and 6 females, albumin level seen in group 3 females and cholesterol level changes seen at the end of treatment day 29 were generally of low magnitude, and remained within historical reference ranges. The changes seen in albumin and A/G ratio at the end of the recovery period was also of low magnitude and within historical ranges.

Urinalysis

Urine samples were collected from animals at the end of treatment and recovery period from 12 to 18 hours.

There were no test article (HTX-019) related effects on urinalysis parameters when compared to the comparator (Emend IV) or the saline controls during the treatment and recovery periods.

Gross Pathology

All the surviving animals were examined, euthanized, and necropsied at the end of the treatment and recovery periods.

There was no macroscopic observation considered related to treatment with the comparator or test item, HTX-019 (low, mid or high dose) in the main study or recovery animals.

The macroscopic findings of mass, pale thickening, and/or dark material observed at the infusion site were due to the indwelling infusion catheter, were correlated with microscopic changes in the animals, and were observed in all the study animals. However, there was an increase in the incidence of mass in the animals treated with the comparator (Emend IV) and HTX-019 high dose, indicating exacerbation by these test items, in comparison with the controls.

Other macroscopic finding considered to be associated with the infusion site findings include: adrenal gland dark area; liver mass, dark/pale discoloration, adhesion; lymph nodes (bronchial, iliac, mandibular, mediastinal, pancreatic) enlargement; lungs, dark focus/pale area/firm; prostate, firm/pale area and small; spleen enlargement; testes, small; thymus, small. These macroscopic findings correlated with the microscopic findings.

The infusion site macroscopic findings showed lower incidence after the 28 day recovery period, when compared to the main animal findings at the end of dosing. Infusion site related macroscopic observations were seen in other organs and tissues (adipose, prostate, testes, epididymis and urinary bladder) of a recovery animal (#3012D).

Organ Weights

The following organs were dissected and weighed prior to fixation:

Adrenals	Pituitary
Brain	Prostate
Heart	Spleen
Kidneys	Testes
Liver (2 lobes)	Thymus
Lungs (2 lobes)	Thyroid gland/parathyroids
Ovaries	Uterus

All the paired organs were weighed together.
The mean relative and absolute organ weights were found to be comparable to that of the saline control group ranges at treatment day 29 and recovery day 57 for all the treatment groups, with the exception of the kidneys and liver.

At the end of dosing on day 29, the mean absolute liver weight of group 3 (comparator) and group 6 (high dose) animals, and for group 5 (mid dose) females increased significantly when compared to saline controls. Similarly, the relative liver weight for group 3 (comparator), group 5 (mid dose) and group 6 (high dose) animals were also significantly increased in comparison to that of the saline controls. In addition, the relative kidney weights for group 3 (comparator) animals and male animals of group 5 (mid dose) were significantly increased.

At the end of the recovery period (day 57), the mean absolute liver and kidney weights of the treated animals were comparable to the saline control group. However, the mean relative liver and kidney weight of groups 3, 5 and 6 animals were still significantly increased.

A summary of the organ weight data is presented in the tables below.

Mean Absolute Organ Weights (G) in Male Rats at the end of Dosing on Day 29

Organs	Group 1 Saline Control	Group 2 Vehicle Control	Group 3 Aprepitant 7.2 mg	Group 4 HTX-019 0.72 mg	Group 5 HTX-019 2.40 mg	Group 6 HTX-019 7.2 mg
Liver	10.2013 ± 1.05799	10.3941 ± 1.12691	12.2343 ± 1.58993	10.5704 ± 0.75649	11.1981 ± 1.19855	12.6969 ± 0.82554
Kidneys	2.5207 ± 0.12325	2.7408 ± 0.26064	2.7039 ± 0.21782	2.6189 ± 0.20971	2.8092 ± 0.28345	2.6981 ± 0.21461

Mean Absolute Organ Weights (G) in Female Rats at the end of Dosing on Day 29

Organs	Group 1 Saline Control	Group 2 Vehicle Control	Group 3 Aprepitant 7.2 mg	Group 4 HTX-019 0.72 mg	Group 5 HTX-019 2.40 mg	Group 6 HTX-019 7.2 mg
Liver	6.9115 ± 0.70505	6.8621 ± 0.87815	9.6987 ± 1.24879	7.8868 ± 0.68084	9.1030 ± 1.32967	10.3092 ± 1.12878
Kidneys	1.6420 ± 0.11791	1.7730 ± 0.16623	1.8448 ± 0.10881	1.7615 ± 0.08799	1.8432 ± 0.21658	1.8472 ± 0.13256

Mean Absolute Organ Weights (G) in Male Rats at the end of Recovery Day 57

Organs	Group 1 Saline Control	Group 2 Vehicle Control	Group 3 Aprepitant 7.2 mg	Group 4 HTX-019 0.72 mg	Group 5 HTX-019 2.40 mg	Group 6 HTX-019 7.2 mg
Liver	11.3164 ± 0.66574	12.5048 ± 0.60776	12.9486 ± 1.78050	12.3137 ± 1.28742	11.5990 ± 1.14648	11.8490 ± 1.75034
Kidneys	2.7986 ± 0.43310	2.9326 ± 0.20289	2.9372 ± 0.21777	2.9254 ± 0.33434	2.8941 ± 0.28724	2.7811 ± 0.30752

Mean Absolute Organ Weights (G) in Female Rats at the end of Recovery Day 57

Organs	Group 1 Saline Control	Group 2 Vehicle Control	Group 3 Aprepitant 7.2 mg	Group 4 HTX-019 0.72 mg	Group 5 HTX-019 2.40 mg	Group 6 HTX-019 7.2 mg
Liver	7.7672 ± 0.64445	7.0625 ± 0.83655	7.2951 ± 1.53232	7.6381 ± 0.48535	8.0809 ± 0.41355	7.7040 ± 0.95388
Kidneys	1.7488 ± 0.13641	1.7569 ± 0.27392	1.6116 ± 0.30192	1.8360 ± 0.16463	1.8236 ± 0.17822	1.7609 ± 0.20258

Results are mean values ± standard deviation

Mean Relative Organ Weights (G) in Male Rats at the end of Dosing on Day 29

Organs	Group 1 Saline Control	Group 2 Vehicle Control	Group 3 Aprepitant 7.2 mg	Group 4 HTX-019 0.72 mg	Group 5 HTX-019 2.40 mg	Group 6 HTX-019 7.2 mg
Liver	2.5542 ± 0.17833	2.5478 ± 0.12731	3.0945 ± 0.25465	2.7081 ± 0.16718	2.8057 ± 0.26740	3.1354 ± 0.15046
Kidneys	0.6325 ± 0.02332	0.6739 ± 0.05313	0.6862 ± 0.04299	0.6713 ± 0.05310	0.7062 ± 0.08776	0.6671 ± 0.05765

Mean Relative Organ Weights (G) in Female Rats at the end of Dosing on Day 29

Organs	Group 1 Saline Control	Group 2 Vehicle Control	Group 3 Aprepitant 7.2 mg	Group 4 HTX-019 0.72 mg	Group 5 HTX-019 2.40 mg	Group 6 HTX-019 7.2 mg
Liver	2.9335 ± 0.23134	2.9052 ± 0.29927	4.0998 ± 0.40913	3.2123 ± 0.19572	3.6493 ± 0.45139	4.0910 ± 0.41728
Kidneys	0.6985 ± 0.05133	0.7514 ± 0.04576	0.7832 ± 0.06269	0.7201 ± 0.06147	0.7371 ± 0.04518	0.7339 ± 0.05709

Mean Relative Organ Weights (G) in Male Rats at the end of Recovery Day 57

Organs	Group 1 Saline Control	Group 2 Vehicle Control	Group 3 Aprepitant 7.2 mg	Group 4 HTX-019 0.72 mg	Group 5 HTX-019 2.40 mg	Group 6 HTX-019 7.2 mg
Liver	0.3728 ± 0.01059	2.5855 ± 0.07003	2.5486 ± 0.17608	2.5229 ± 0.20466	2.5532 ± 0.15859	2.5067 ± 0.16337
Kidneys	0.6309 ± 0.07759	0.6058 ± 0.01204	2.9372 ± 0.21777	0.5985 ± 0.04431	0.6371 ± 0.03854	0.5922 ± 0.06202

Mean Relative Organ Weights (G) in Female Rats at the end of Recovery Day 57

Organs	Group 1 Saline Control	Group 2 Vehicle Control	Group 3 Aprepitant 7.2 mg	Group 4 HTX-019 0.72 mg	Group 5 HTX-019 2.40 mg	Group 6 HTX-019 7.2 mg
Liver	2.9343 ± 0.19197	2.7662 ± 0.16764	3.0337 ± 0.39360	2.7460 ± 0.17769	2.8929 ± 0.10687	2.8801 ± 0.12888
Kidneys	0.6599 ± 0.02420	0.6862 ± 0.06019	0.6718 ± 0.07819	0.6600 ± 0.05784	0. 6510 ± 0.02116	0.6590 ± 0.02637

Results are mean values ± standard deviation

Histopathology

Tissue samples for histopathology were fixed in 10 % neutral buffered formalin, and were prepared for microscopic examination by embedding in paraffin wax, sectioning

and staining with hematoxylin and eosin. Gram's stain was used for the liver, kidney, lungs, infusion site and/or heart for some animals. The following organs were examined:

ORGANS/TISSUES	Retain (•)	Weigh (√)	Examine (€)	ORGANS/TISSUES	Retain (•)	Weigh (√)	Examine (€)
Adrenals	•	√	€	Duodenum	•		€
Animal identification	•			Jejunum	•		€
Aorta (thoracic)	•		€	Ileum	•		€
Blood	•			SC, cervical	•		€
Bone marrow smears (3)	•			Spleen	•	√	€
Brain	•	√	€	Sternum & marrow	•		€
Cecum	•		€	Stomach	•		€
Colon	•		€	Testes	•d	√	€
Epididymides	•d		€	Thymus	•	√	€
Esophagus	•		€	Thyroid gland/parathyroids	•	√	€
Eyes	•a		€	Tongue	•		€
Femur & marrow	•		€	Trachea	•c		€
Heart	•	√	€	Urinary bladder	•		€
Kidneys	•	√	€	Uterus	•	√	€
Liver (2 lobes)	•	√	€	Vagina	•		€
Lungs (2 lobes)	•bc	√	€				
LN, mandibular	•		€	Abnormal findings	•		€
LN, mesenteric	•		€				
Mammary gland (inguinal)	•		€				
Optic nerves	•a		€	**Additional Tissues presented below**			
Ovaries	•	√	€	Infusion site	•		€
Pancreas	•		€				
Pituitary	•	√	€				
Prostate	•	√	€				
Rectum	•		€				
SG, mandibular	•		€				
Sciatic nerve	•		€				
Seminal vesicles	•		€				
Skeletal muscle	•		€				
Skin & subcutis (inguinal)	•		€				

a	Davidson's fluid (euthanized animal only)
b	Lungs were infused with 10% neutral buffered formalin (euthanized animal only)
c	Lungs were weighed with trachea and recorded in Provantis as lungs in organ weight record
d	Bouin's fluid (euthanized animal only)
LN	Lymph node
SG	Salivary gland
SC	Spinal cord
€	Examine microscopically

Notes:
Paired organs weighed together
Parathyroids, mammary gland and optic nerves were only examined histologically if present in routine sections

Adequate Battery
Yes
Peer Review
Yes
Histological Findings

There was no microscopic finding that was considered related to treatment with Emend IV or HTX-019 (high dose) in the main study animals.
There were minimal to mild periportal vacuolation observed in the liver of 3 of 9 female animals treated with Emend IV, 3 of 10 female animals treated with high dose HTX-019, and 1 female animal treated with HTX-019 mid dose. This effect was also seen in the liver of two female animals (dosed with the comparator Emend IV), one found dead (#3504A) and one that was pre-terminally euthanized (3515F) during the study.

There were several microscopic findings observed at the infusion site of the animals treated with the comparator (Emend IV) or the test article, HTX-019 as well as the controls (saline and vehicle). The microscopic findings at the infusion site included: minimal to severe vascular wall necrosis/chronic-active inflammation/abscess and/or mural thrombosis (with/without mineral deposits or cholesterol clefts). Some of these findings correlated with the macroscopic observations already noted at the infusion site from catheter placement in the animals. A higher incidence and/or severity of the infusion site findings was observed in the animals treated with Emend or the high dose test article, when compared the controls (saline and vehicle), suggesting exacerbation by these test items.

Other microscopic findings randomly observed in a variety of organs and tissues, and considered related to the inflammatory and/or thrombotic changes at the infusion site are: infarct of the adrenal gland; necrosis/chronic-active inflammation/abscess/mural thrombosis of the liver; increased cellularity of the lymph node; alveolar/interstitial inflammation/vasculitis of the lungs; prostatitis; extramedullary hematopoiesis of the spleen; increased granulopoiesis of the femoral and sternal bone marrow; seminiferous tubular atrophy; and/or decreased cellularity of the thymus. These microscopic findings often correlate with the observed macroscopic findings and were considered to be embolic, reactive and/or degenerative changes secondary to the infusion site changes.

After the 28-day recovery period, the periportal vacuolation seen in the liver of the female rats treated with Emend or HTX-019 was reversed. There was a lower incidence of the injection site related microscopic findings at the end of the 28 day recovery period, suggesting the possibility of progressive ongoing reversal of the injection site adverse effect with a longer recovery period. Injection site related microscopic findings were noted in other organs and tissues such as adipose tissue, prostate, testes, epididymis and urinary bladder of one recovery animal (3012D).

In conclusion, the administration of the HTX-019 vehicle control, Emend IV (7.2 mg), HTX-019 (0.72, 2.40 and 7.2 mg) by IV injection for 28 days to Sprague-Dawley rats followed by 28 day recovery period resulted in random death or morbidity at all doses due to the systemic spread of procedure-related (catheter infusion) inflammatory and/or thrombotic lesions, at the infusion site. A higher incidence of dead or preterminally euthanized animals was noted in the females treated with Emend IV. Minimal to mild periportal vacuolation was observed in the liver of female animals that were treated with Emend IV and mid (2.4 mg) and high (7.2) dose HTX-019 and survived till termination. This finding was also observed in the liver of two female animals (dosed with Emend IV), one found dead (3504A) and one euthanized (3515F) during the study. After the 28 day recovery period, the liver periportal vacuolation finding was not seen in the recovery animals, suggesting reversibility of the effect. Also, a lower incidence and/or severity of the procedure-related macroscopic/microscopic findings were noted at the infusion sites as well as in other organs and tissues of the recovery animals, when compared to those observed in the main study animals, indicating the likelihood of progressive ongoing reversal of the effects with a longer recovery period.

Summaries of the gross pathology and histopathology findings are presented in the sponsor's tables below.

Table 1 Incidence of Macroscopic Observations noted at the Infusion Site of the Terminal (Main), Found Dead and Preterminally Euthanized Animals

Sex	M	F	M	F	M	F	M	F	M	F	M	F
Group Numbers	1		2		3		4		5		6	
Group Designation	Saline Control		Vehicle Control		Comparator (EMEND® IV)		Low Dose HTX-019		Mid Dose HTX-019		High Dose HTX-019	
AP1 (mg) Aprepitant	NA		NA		7.2		.72		2.40		7.2	
Dose Volume (mL)	1.0											
Number of Animals Examined	10	10	11*	10	10	12*	10	10	10	11*	11*	10
Infusion Site												
Mass	1	0	2*	0	2	4*	1	0	0	2	2	3
Thickening	2	1	2	1	1	2*	1	0	1	0	2	0
Pale material	0	0	0	0	1	0	0	0	0	0	2*	0
Gelatinous material*	0	0	0	0	0	2*	0	0	0	0	0	0

*Found Dead and Preterminally Euthanized Animals included

Table 3 Incidence of Macroscopic Observations noted at the Infusion Site of the Recovery Animals

Sex	M	F	M	F	M	F	M	F	M	F	M	F
Group Numbers	1		2		3		4		5		6	
Group Designation	Saline Control		Vehicle Control		Comparator (EMEND® IV)		Low Dose HTX-019		Mid Dose HTX-019		High Dose HTX-019	
AP1 (mg) Aprepitant	NA		NA		7.2		.72		2.40		7.2	
Dose Volume (mL)	1.0											
Number of Animals Examined	5	5	4	5	5	3	5	5	5	4	4	5
Infusion Site												
Thickening	0	0	0	0	0	0	0	0	1	0	0	0

Table 2 Incidence of Microscopic Findings noted at the Infusion Site in the Terminal (Main), Found Dead and Preterminally Euthanized Animals

Sex		M	F	M	F	M	F	M	F	M	F	M	F
Group Numbers		1		2		3		4**		5**		6	
Group Designation		Saline Control		Vehicle Control		Comparator (EMEND® IV)		Low Dose HTX-019		Mid Dose HTX-019		High Dose HTX-019	
API (mg) Aprepitant		NA		NA		7.2		.72		2.40		7.2	
Dose Volume (mL)		1.0											
Number of Animals Examined		10	10	11*	10	10	12*	3	0	1	3*	11*	10
Infusion Site													
Necrosis vascular wall	Total	0	1	0	1	1	7*	0	0	0	2	2	2
	minimal	0	0	0	0	0	1	0	0	0	0	0	0
	mild	0	1	0	0	0	2	0	0	0	0	1	0
	moderate	0	0	0	1	0	3*	0	0	0	1	0	0
	severe	0	0	0	0	1	1	0	0	0	1	1	2
Abscess	Total	3	0	1	0	4	2	0	0	0	0	2	2
	moderate	2	0	1	0	0	0	0	0	0	0	1	0
	severe	1	0	0	0	4	2	0	0	0	0	1	2
Inflammation, vascular wall	Total	9	5	6*	2	6	11*	1	0	0	2	8*	4
	minimal	1	3	2	1	0	0	0	0	0	0	2	2
	mild	5	1	2	0	2	1	0	0	0	0	2	0
	moderate	2	1	1	0	0	4	1	0	0	0	2*	0
	severe	1	0	1*	1	4	6*	0	0	0	2	2	2
Granuloma, foreign body	Total	0	0	3	2	1	0	0	0	0	0	1	1
	minimal	0	0	3	2	1	0	0	0	0	0	1	1
Thrombosis, mural	Total	6	3	3	5	2	2*	0	0	0	0	5	1
	minimal	0	0	0	1	1	1	0	0	0	0	2	0
	mild	6	3	3	4	1	0	0	0	0	0	3	1
	moderate	0	0	0	0	0	1*	0	0	0	0	0	0
Thickening, intimal	Total	0	5	4	5	2	1	2	0	1	0	1	1
	minimal	0	4	3	4	1	0	0	0	1	0	0	1
	mild	0	1	1	1	1	1	1	0	0	0	1	0
	moderate	0	0	0	0	0	0	1	0	0	0	0	0
Catheter tract	Total	1	3	2	4	6	5	1	0	0	0	3	3
	present	1	3	2	4	6	5	1	0	0	0	3	3

* Found Dead and Preterminally Euthanized Animals included

** Infusion site tissue was evaluated for only animals (Groups 4 and/or 5) with gross pathology finding

Table 4 Incidence of Microscopic Findings noted at the Infusion Site of the Recovery Animals

Sex		M	F	M	F	M	F	M	F	M	F	M	F
Group Numbers		1		2		3		4**		5**		6	
Group Designation		Saline Control		Vehicle Control		Comparator (EMEND® IV)		Low Dose HTX-019		Mid Dose HTX-019		High Dose HTX-019	
API (mg) Aprepitant		NA		NA		7.2		.72		2.40		7.2	
Dose Volume (mL)		1.0											
Number of Animals Examined		5	5	4	5	5	3	0	0	1	0	4	5
Infusion Site													
Inflammation, vascular wall	Total	4	1	1	3	4	3	0	0	1	0	1	3
	minimal	1	0	0	0	1	0	0	0	1	0	0	1
	mild	3	1	1	3	2	3	0	0	0	0	0	2
	moderate	0	0	0	0	1	0	0	0	0	0	1	0
Granuloma, foreign body	Total	0	1	0	0	0	0	0	0	0	0	0	0
	minimal	0	1	0	0	0	0	0	0	0	0	0	0
Thrombosis, mural	Total	1	1	0	1	2	0	0	0	1	0	1	0
	minimal	0	1	0	1	0	0	0	0	0	0	0	0
	mild	0	0	0	0	1	0	0	0	0	0	1	0
	moderate	1	0	0	0	1	0	0	0	1	0	0	0
Thickening, intimal	Total	0	0	0	0	0	0	0	0	1	0	0	0
	minimal	0	0	0	0	0	0	0	0	1	0	0	0
Catheter tract	Total	1	0	0	0	2	1	0	0	0	0	0	0
	present	1	0	0	0	2	1	0	0	0	0	0	0

**Infusion site tissue was evaluated for only animals (Groups 4 and/or 5) with gross pathology finding

Toxicokinetics

Blood samples (0.4 ml) were collected from TK animals groups 3 to 6 (7.2 mg Emend, 0.72, 2.4 and 7.2 mg, HTX-019), respectively, on treatment days 1 and 28 pre-dose, at 10, 30 min, 1, 2, 4, 8, 12 and 24 hours after dosing. Additional blood samples were collected pre-dose and 10 min post dose on days 8, 15 and 21 for groups 3, 4, 5, and 6 animals.

After the IV bolus administration of HTX-019 (aprepitant) once daily for 28 days, the systemic exposure to aprepitant increased generally in a more than dose-proportional manner in male and female animals on Day 1. However, on Day 28, systemic exposure to aprepitant increased in a slightly more than dose-proportional manner for the females dosed at (0.72 - 2.4 mg/day); for male animals dosed at (2.4 – 7.2 mg/day); and generally in a dose proportional manner for females dosed at (2.4 – 7.2 mg/day), and for males dosed at (0.72 – 2.4 mg/day).

Exposure to Aprepitant (C_{max}, $AUC_{0\text{-}Tlast}$ and AUC_{INF}) was higher in both sexes dosed with 7.2 mg HTX-019, than the group 3 animals dosed with 7.2 mg Emend IV on Day 1 and Day 28. Over the 28-day treatment period, C_{max}, $AUC_{0\text{-}Tlast}$ and AUC_{INF} values obtained from male animals dosed with 0.72 mg HTX-019 were comparable on Day 1 and Day 28. However, the TK parameters obtained on Day 28 from group 3, 5, and 6 male and female animals, and from group 4 female animals were lower than the values obtained on Day 1. A summary of the TK parameters is shown in the sponsor's Table below.

Table 1: Mean Day 1 Toxicokinetic Parameters of Aprepitant in Rat Plasma for the IV bolus Administration

Day	Group	Sex	R^2	$T_{1/2}$ (hr)	T_{max} (hr)	C_{max} (ng/mL)	$AUC_{0\text{-}Tlast}$ (hr*ng/mL)	AUC_{INF} (hr*ng/mL)	Vz (mL)	Cl (mL/hr)
1	3	Female	0.96	3.57	0.17	8727	84769	85913	432	83.81
1	3	Male	1.00	3.06	0.17	5833	47320	47662	667	151.06
1	4	Female	1.00	4.09	0.17	1680	8208	8383	507	85.89
1	4	Male	0.97	4.15	0.17	688	2775	2825	1525	254.87
1	5	Female	1.00	2.72	0.17	13253	43500	43628	216	55.01
1	5	Male	0.99	2.85	0.17	6763	18255	18293	539	131.20
1	6	Female	0.99	4.15	0.17	121000	279206	283156	152	25.43
1	6	Male	0.99	2.55	0.17	38387	99280	99384	267	72.45

Table 2: Mean Day 28 Toxicokinetic Parameters of Aprepitant in Rat Plasma for the IV bolus Administration

Day	Group	Sex	R^2	$T_{1/2}$ (hr)	T_{max} (hr)	C_{max} (ng/mL)	$AUC_{0\text{-}Tlast}$ (hr*ng/mL)	AUC_{INF} (hr*ng/mL)	Vz (mL)	Cl (mL/hr)
28	3	Female	0.99	2.34	0.50	5170	22794	22830	1067	315.37
28	3	Male	0.94	3.70	0.17	2583	11653	11847	3243	607.76
28	4	Female	0.96	4.64	0.50	825	4068	4149	1162	173.54
28	4	Male	0.98	3.78	0.17	482	2126	2148	1827	335.22
28	5	Female	0.99	2.75	0.17	9830	17299	17340	549	138.41
28	5	Male	1.00	3.09	0.17	3860	6402	6418	1666	373.97
28	6	Female	0.99	2.04	0.50	30400	53380	53397	398	134.84
28	6	Male	0.88*	2.54	0.17	26800	26102	26168	1006	275.14

* Out of specification ≥ 0.9

Dosing Solution Analysis

The concentrations of aprepitant (0.72, 2.4 and 7.2 mg) at the initial testing and end of study re-testing for the stock solution were within the 90.0 to 111.0% of the target concentrations. No aprepitant was detected in the saline (control) group.

10 Special Toxicology Studies

Effects of HTX-019 and Emend IV in the Rabbit Ear Phlebitis Model after a Single Intravenous or Perivascular Administration. (**Study No. 33-001**)

The objective of this study was to determine the local tolerance of New Zealand White Rabbits to HTX-019, Emend IV (Fosaprepitant) in different vehicle formulations after a single intravenous (IV) and paravenous (PV) injection.

Methods:

Doses: HTX-019 (lot # 110514), Emend IV (lot # K006624), Vehicle control emulsion (lot # 112514), Aprepitant in a low Sodium Oleate vehicle control emulsion (lot # 120314), Low Sodium Oleate vehicle control emulsion (lot # 120414) and 0.9 % Sodium Chloride NaCl) for Injection (lot # C903161) were administered by single IV injection (1

ml/kg for 20 mins) to rabbits through the marginal right ear vein, and by subcutaneous bolus perivascular injection (0.5 ml) through the marginal left ear vein. The saline (0.9 % NaCl) was used as the vehicle control. 7.2 mg/kg of the test article or Emend was injected IV into the right ear vein, and 3.6 mg was injected subcutaneously into the left ear vein.

Study design: Twenty eight (28) male New Zealand White Rabbits, 12 weeks old, and weighing 2.3 to 2.8 kg were randomly divided into 6 groups (5/group for 1 to 5, and 3 animals in group 6) and were administered the test article, vehicle formulations or negative control IV (at 1 ml/kg) via the right marginal vein and by subcutaneous perivascular injection (at 0.5 ml) via the left ear. The areas (approximately 1 cm) beyond the point of entry of the needle were evaluated macroscopically and microscopically. Clinical signs were recorded at 1, 4 and 24 hours post-dose, and irritation at the dosing sites was graded (by Draize method) at 1 and 24 hours post-dose. Two samples of tissues were excised from the injection sites (3-10 mm proximally and 20-30 mm distally) and were fixed in 10 % neutral buffered formalin (NBF) and evaluated microscopically. All the surviving animals were weighed on day 2 prior to euthanasia. Tissues for evaluation were embedded in paraffin blocks, thin sectioned, and stained with hematoxylin and eosin for microscopic evaluation. Histopathological evaluations were done on all collected tissues and the findings were scored with respect to loss of venous endothelial cells, inflammatory cell infiltration, edema, epidermal degeneration and thrombus formation.

Results:

There were no animal mortalities observed in any of the treatment groups. There were no significant changes observed in the mean body weight of the animals, and there were no adverse clinical signs observed at all the time points (1, 4 and 24 hours after dosing).

Macroscopic results/Injection site scoring

Group 1; comparator Emend IV (Fosaprepitant dimeglumine):

Animals receiving the comparator Emend IV did not show any signs of erythema/edema at the intravenous (IV) site, 1 hour post-dose. However, at the perivenous (PV) bolus injection site, all the animals show a well-defined erythema but no edema at 1 hour post-dose. At 24 hours after dosing, 2 of 5 animals showed very slight erythema and 1 of 5 animals showed well-defined erythema at the intravenous site. At 24 hours after dosing at the PV site, all the animals exhibited well-defined erythema but no edema.

Group 2; HTX-019 vehicle formulation 1:

In animals receiving HTX-019 vehicle formulation, at 1 hour after dosing, 2 of 5 animals showed very slight erythema, and 1 of 5 animals showed well-defined erythema and very slight edema at the IV site. At the PV site, 2 of 5 animals showed well-defined

erythema and 1 of 5 animals showed slight edema at 1 hour post-dose. At 24 hours after dosing, 1 of 5 animals showed very slight erythema and 1 of 5 animals showed well-defined erythema at the IV site. There was no edema at the IV site. At 24 hour after injection at the PV site, 2 of 5 animals showed very slight erythema, but no edema.

Group 3; Vehicle control emulsion formulation 2:

In animals receiving the vehicle control emulsion 1 hour after dosing, 2 of 5 animals showed very slight erythema, and 1 of 5 animals exhibited well-defined erythema at the IV site. However, there was no edema seen in any of the animals at the IV or PV sites at 1 hour post-dose. At the PV site, 4 of 5 animals showed very slight erythema and 1 of 5 animals showed well-defined erythema at 1 hour post-dose. At 24 hours post-dose, there was no edema at the IV or PV sites. One of 5 animals showed very slight erythema at the IV site. At the PV site, 2 of 5 animals showed very slight erythema, and 3 of 5 animals showed well-defined erythema.

Group 4; Aprepitant in a low SO vehicle emulsion formulation 3:

In animals receiving Aprepitant in a low sodium oleate (SO) vehicle emulsion, at 1 hour post-dose, 1 of 5 animals exhibited well defined erythema at the IV site, whereas, at the PV site, 3 of 5 animals showed very slight erythema and 3 of 5 animals showed well-defined erythema. There was no edema at the IV or PV sites in any animal at 1 hour post-dose. At 24 hours post-dose, 1 of 5 animals showed very slight erythema, whereas, at the PV site, 3 of 5 animals showed very slight erythema and 1 of 5 animals showed well-defined erythema. There was no edema at the IV or PV sites in any animal at 24 hours post-dose.

Group 5; Low SO vehicle control emulsion formulation 4:

In animals receiving the low SO vehicle control emulsion, at 1 hour post-dose, there were no signs of erythema or edema at the IV sites, whereas, at the PV site, 2 of 5 animals showed very slight erythema, and 1 of 5 animals showed well defined erythema. There was also no edema at the PV site at 1 hour post-dose. At 24 hours post-dose there were no signs of erythema or edema at the IV site, whereas, at the PV site, 4 of 5 animals exhibited very slight erythema and edema. There was also no edema at the PV site at 24 hours post-dose.

Group 6; 0.9 % Sodium Chloride negative control:

In animals receiving the saline negative control, at 1 hour post-dose, there were no signs of edema at the PV or IV sites, and only 1 of 3 animals exhibited well-defined erythema at the IV site. At the PV site, 2 of 3 animals showed very slight erythema, and 1 of 3 animals showed well defined erythema. At 24 hours after dosing, 3 of 3 animals showed very slight erythema at the IV site, whereas, there were no signs of edema at the IV or PV sites. At the PV site (24 hours post-dose), 1 of 3 animals showed very slight erythema and 2 of 3 animals showed well-defined erythema.

Microscopic results/Histopathology

Histopathology results such as endothelial cell loss, edema, inflammatory cell infiltrates, and epidermal degeneration were scored per protocol using the criteria presented in the sponsor's table below.

Animals dosed intravenously (IV) with fosaprepitant had higher overall scores for endothelial cell loss, edema, hemorrhage, and inflammatory cell infiltrates than the animals that were dosed with saline control. Also, injection sites dosed perivenously (PV) with fosaprepitant had higher overall scores than the PV saline control sites, which were all normal. In addition, injection sites dosed PV with fosaprepitant had higher scores than the sites that were dosed IV with fosaprepitant.
In summary, the findings in the animals dosed with fosaprepitant were consistent with minor vascular irritation, inflammation, and were more severe when the test article was administered PV.
In general, the animals that were dosed with the four different vehicle emulsion formulations had lesser or equal scores to the saline control animal scores, and the findings were considered inconsequential both IV and PV.

B. Table 2 – Criteria for Histopathological Examination

Histopathological Findings	Grade
Loss of venous endothelial cells	
None	0
Less than 1/3 of the vein (in cross-section)	1
1/3 to 2/3 of the vein	2
More than 2/3 of the vein	3
Inflammatory Cell Infiltration	
None	0
Few inflammatory cells in venous wall or perivascular tissue	1
Many inflammatory cells in venous wall or perivascular tissue	2
More diffuse and dense inflammatory cell infiltrates in perivascular tissue	3
Edema	
None	0
Localized to perivascular tissue	1
More diffuse edema	2
Edema of the entire region	3
Epidermal degeneration	
None	0
Some degenerated epidermal cells near the vein	1
Numerous degenerated epidermal cells near the vein (more diffuse and severe epidermal degeneration)	2

Kohno, E., Murase, S. ,Nishikata, M., Okamura, N., Matzno, S., Kuwahara, T., & Matsuyama, K. (2008). Methods of preventing vinorelbine-induced phlebitis; an experimental study in rabbits. *International Journal of Medical Sciences., 5*: 218-223.

Ulcers were seen on the right ear of 3 animals (#422 and #423 from group1; and #435 from group 3). The ulcers were observed away from the injection sites in two animals dosed with IV fosaprepitant in group 1 (#422 and #423), and over the ear vein in one animal dosed with vehicle control emulsion in group 3 (#435). Since there was no other animal with significant epidermal degeneration associated with the injection site, it is likely that the ulcers were caused by trauma (from clipping, scratching, injecting or taping of the catheter), rather than a direct effect of the test article. However, because of the location of the ulcer in animal #435, the finding was listed under injection site.

The minimal granulomatous inflammation seen surrounding hair fragments in the ear section of several animals were considered incidental background findings since they were present before the test agent was administered. Animals dosed with HTX-019 (group 2), vehicle control emulsion (group 3) aprepitant in a low sodium oleate vehicle emulsion (group 4), or the low sodium oleate vehicle control emulsion (group 5) generally had lesser or equal histopathologic grade scores than the findings in the saline control animals, and were considered inconsequential, both intravenously and perivenously.

In conclusion, injection sites dosed IV or PV with fosaprepitant had higher overall scores for endothelial cell loss, edema, hemorrhage, and inflammatory cell infiltrates than sites dosed with the four different vehicle emulsion formulations, which also had lesser or equal scores to the saline control animal. The injection sites dosed PV with fosaprepitant had higher scores than the sites that were dosed IV with fosaprepitant. A summary of the study results in presented in the sponsor's table below. The findings in the animals dosed with fosaprepitant, although more severe than that of the animals dosed with the other four different vehicle emulsion formulation, were consistent with minor vascular irritation, inflammation, and were more severe when the test article was administered PV.

Macroscopic Results/Injection Site Scoring (Draize)

Group 1			1 hour				24 hour			
			Erythema		Edema		Erythema		Edema	
Animal #	Sex	Group	R (TA at iv 1 mL/kg)	L (TA pv at 0.1 mL)	R (TA at iv 1 mL/kg)	L (TA pv at 0.1 mL)	R (TA at iv 1 mL/kg)	L (TA pv at 0.1 mL)	R (TA at iv 1 mL/kg)	L (TA pv at 0.1 mL)
446	M	1	0	2	0	0	2	2	0	0
442	M	1	0	2	0	0	1	2	0	0
423	M	1	0	2	0	0	0	2	0	0
424	M	1	0	2	0	0	0	2	0	0
425	M	1	0	2	0	0	1	2	0	0

TA = Emend IV (Fosaprepitant dimeglumine) for Injection

Group 2			1 hour				24 hour			
			Erythema		Edema		Erythema		Edema	
Animal #	Sex	Group	R (V1 at iv 1 mL/kg)	L (V1 pv at 0.1 mL)	R (V1 at iv 1 mL/kg)	L (V1 pv at 0.1 mL)	R (V1 at iv 1 mL/kg)	L (V1 pv at 0.1 mL)	R (V1 at iv 1 mL/kg)	L (V1 pv at 0.1 mL)
426	M	2	0	0	0	0	0	0	0	0
427	M	2	0	0	0	0	0	0	0	0
428	M	2	2	0	1	0	2	0	0	0
429	M	2	1	2	0	0	1	1	0	0
430	M	2	1	2	0	1	0	1	0	0

V1 = HTX-019

Group 3			1 hour				24 hour			
			Erythema		Edema		Erythema		Edema	
Animal #	Sex	Group	R (V2 at iv 1 mL/kg)	L (V2 pv at 0.1 mL)	R (V2 at iv 1 mL/kg)	L (V2 pv at 0.1 mL)	R (V2 at iv 1 mL/kg)	L (V2 pv at 0.1 mL)	R (V2 at iv 1 mL/kg)	L (V2 pv at 0.1 mL)
431	M	3	1	1	0	0	1	1	0	0
432	M	3	0	1	0	0	0	2	0	0
433	M	3	1	1	0	0	0	1	0	0
434	M	3	0	1	0	0	0	2	0	0
435	M	3	0	2	0	0	0	2	0	0

V2 = Vehicle Control Emulsion

Macroscopic Results/Injection Site Scoring (Draize)

Group 4			1 hour				24 hour			
			Erythema		Edema		Erythema		Edema	
Animal #	Sex	Group	R (V3 at iv 1 mL/kg)	L (V3 pv at 0.1 mL)	R (V3 at iv 1 mL/kg)	L (V3 pv at 0.1 mL)	R (V3 at iv 1 mL/kg)	L (V3 pv at 0.1 mL)	R (V3 at iv 1 mL/kg)	L (V3 pv at 0.1 mL)
436	M	4	0	1	0	0	0	0	0	0
437	M	4	0	1	0	0	0	1	0	0
438	M	4	2	1	0	0	1	1	0	0
439	M	4	0	2	0	0	0	2	0	0
440	M	4	0	2		0	0	1	0	0

V3 = Aprepitant in a Low SO Vehicle Emulsion)

Group 5			1 hour				24 hour			
			Erythema		Edema		Erythema		Edema	
Animal #	Sex	Group	R (V4 at iv 1 mL/kg)	L (V4 pv at 0.1 mL)	R (V4 at iv 1 mL/kg)	L (V4 pv at 0.1 mL)	R (V4 at iv 1 mL/kg)	L (V4 pv at 0.1 mL)	R (V4 at iv 1 mL/kg)	L (V4 pv at 0.1 mL)
441	M	5	0	0	0	0	0	1	0	0
442	M	5	0	0	0	0	0	1	0	0
443	M	5	0	2	0	0	0	2	0	0
444	M	5	0	1	0	0	0	1	0	0
445	M	5	0	1	0	0	0	1	0	0

V5 = Low SO Vehicle Control Emulsion

Group 6			1 hour				24 hour			
			Erythema		Edema		Erythema		Edema	
Animal #	Sex	Group	R (V5 at iv 1 mL/kg)	L (V5 pv at 0.1 mL)	R (V5 at iv 1 mL/kg)	L (V5 pv at 0.1 mL)	R (V5 at iv 1 mL/kg)	L (V5 pv at 0.1 mL)	R (V5 at iv 1 mL/kg)	L (V5 pv at 0.1 mL)
447	M	6	0	1	0	0	1	2	0	0
448	M	6	2	2	0	0	1	1	0	0
449	M	6	0	1	0	0	1	2	0	0

V6 = 0.9% Sodium Chloride for Injection

Other Toxicity Studies

Hemolysis Screening of Test Article HTX-019 (**Study No. 1176-R1**)

The study objective was to evaluate the hemolytic potential of HTX-019 in dog, rat and human red blood cells *in vitro*.

Methods:
Heparinized blood from dog, rat and human stored at room temperature was used within 8 hours of collection. Two-fold dilutions (0.08-10 % of the test article were prepared in saline or D5W and then diluted into aliquots of blood, which were then incubated at 37^{o}C for 45 min and then centrifuged to separate the cells from plasma. An aliquot of plasma was diluted with a (Drabkin's) reagent and the OD540 was measured. A calibration curve prepared by dilution of non-centrifuged blood was used to determine the degree of hemolysis, and triton X-100 was used as the reference compound.

Results:

There was no significant hemolysis (>10 %) observed at any concentration in any of the species (rat, dog or human) in either saline or D5W diluent.

The test article at the highest concentration (especially for dog) prevented good separation of the plasma from the RBC pellet and this could have resulted in the slight hemolysis seen at the highest concentration. In addition, the test article (neat) caused the plasma to become slightly opaque which also may have affected the absorbance reading at the highest concentration. The triton-X-100 positive control resulted in hemolysis in this assay samples.

In conclusion, the incubation of HTX-019 at concentrations of 0.08 % to10 % with rat, dog or human blood, in either saline or D5W diluent or with the undiluted 100 % test article did not result in any hemolysis.

Safety Evaluation of the Container Closure System for Leachables and Extractables

Cinvanti injectable emulsion is filled in 20 ml (USP (b) (4) clear glass vials with 20 mm opening. The vials are closed with 20 mm (b) (4) rubber stopper and capped with (b) (4) aluminum overseal including matte red plastic flip-off cap. The interior surface of the Cinvanti stopper (b) (4) which provides a barrier to leachables and extractables. To evaluate the safety of the container closure system, extraction studies with the stopper was performed. The extraction studies with the whole stopper with organic solvents were not found to be representative and gave a large number of peaks. Therefore, a study was designed to evaluate extractable compounds only from the (b) (4) portion of the stopper.

Component of the Cinvanti Injectable Emulsion Container Closure Packaging System

Component Description	Manufacturer/Supplier	DMF
20 mL USP (b) (4) lass vial, clear, with 20 mm opening	(b) (4)	(b) (4)
(b) (4)		
(b) (4) Aluminum overseal with Matte Red plastic flip-off cap		

Controlled Extraction Study (Sealed Vials)

In this study, vials were filled with 18 ml of isopropanol and sealed with (b) (4) stoppers and (b) (4) aluminum seals. The sealed vials were placed in a beaker in an inverted orientation (exposing the maximum stopper surface to the solvent) (b) (4) were no extractables greater than the analytical evaluation

threshold [redacted] (b) (4). Thus, the [redacted] (b) (4) provides a nearly complete barrier for extraction of compounds in organic solvents.

Leachables Evaluation on Storage

Leachable compounds assessment was performed on the drug product stored [redacted] (b) (4)

11 Integrated Summary and Safety Evaluation

In this 505(b)(2) NDA submission, the sponsor is seeking approval of cinvanti [redacted] (b) (4) in combination with other antiemetics, for the prevention of acute and delayed nausea and vomiting associated with highly emetogenic cancer chemotherapy (HEC), and [redacted] (b) (4) nausea and vomiting associated with initial and repeat courses of moderately emetogenic cancer chemotherapy (MEC).

Aprepitant is a selective, potent Substance P/Neurokinin 1 (NK1) receptor antagonist approved for chemotherapy-induced nausea and vomiting (CINV). Aprepitant acts by inhibiting emesis induced by cytotoxic agents, such as cisplatin, via the central nervous system.

The Applicant is relying on the nonclinical studies performed by the innovator for the nonclinical safety of cinvanti. In addition, the Applicant submitted nonclinical pharmacokinetic (PK) studies in rats, a repeat dose toxicology study in rats, a local tolerance study in rabbits and a hemolysis study in rat, dog and human blood.

In single dose PK studies in male rats dosed with Emend IV or Cinvanti, the PK parameters were not significantly different for the two test articles. In vitro plasma protein binding was similar for blood samples obtained from animals dosed with the two formulations.
In groups of male rats administered a single dose of Cinvanti or Emend IV at 2 mg/kg, the exposure to aprepitant in rats given Cinvanti was greater than the exposure from Emend IV, because the PK of aprepitant in Emend IV is dependent on the conversion of the pro-drug fosaprepitant to aprepitant. In addition, a comparison of Cinvanti emulsion or Emend IV to Cinvanti in a low sodium oleate (SO) vehicle emulsion, at a dose of 2

mg/kg in male Sprague-Dawley rats, showed that all the three formulations have similar elimination profiles in rats.
Toxicology studies conducted by the innovator in different animal species, in addition to the toxicology study submitted by the sponsor have established the nonclinical safety of cinvanti injectable emulsion.

No genotoxicity studies of cinvanti have been performed. However, aprepitant was not genotoxic in the Ames test, the human lymphoblastoid cell (TK6) mutagenesis test, the rat hepatocyte DNA strand break test, the Chinese hamster ovary (CHO) cell chromosome aberration test and the mouse micronucleus test.

Carcinogenicity studies were conducted with aprepitant in Sprague-Dawley rats and in CD-1 mice for 2 years. In the rat carcinogenicity studies, animals were treated with oral doses ranging from 0.05 to 1000 mg/kg twice daily. The highest dose produced systemic exposures to aprepitant approximately equivalent to (female rats) or less than (male rats) the human exposure at the cinvanti RHD of 130 mg. Treatment with aprepitant at doses of 5 to 1000 mg/kg twice daily caused an increase in the incidence of thyroid follicular cell adenomas and carcinomas in male rats. In female rats, it produced hepatocellular adenomas at 5 to 1000 mg/kg twice daily and hepatocellular carcinomas and thyroid follicular cell adenomas at 125 to 1000 mg/kg twice daily. In the mouse carcinogenicity studies, the animals were treated with oral doses ranging from 2.5 to 2000 mg/kg/day. The highest dose produced a systemic exposure approximately 2 times the human exposure at the RHD of cinvanti 130 mg. Treatment with aprepitant produced skin fibrosarcomas at 125 and 500 mg/kg/day doses in male mice.

Aprepitant did not affect the fertility or general reproductive performance of male or female rats at doses up to the maximum feasible dose of 1000 mg/kg twice daily.

In a local tolerance study to evaluate the potential for venous irritation, cinvanti was compared to Emend IV after IV administration of a 1 ml/kg dose into the marginal ear vein of New Zealand White rabbits. Injection sites dosed IV with fosaprepitant had higher overall scores for endothelial cell loss, edema, hemorrhage, and inflammatory cell infiltrates than sites dosed with cinvanti emulsion, which had lesser or equal scores than the saline control rabbits.

Cinvanti or fosaprepitant did not result in any hemolysis when incubated with rat, dog or human blood.

The safety assessment of potential migrants (leachables and extractables) from the glass vial (container closure system) sealed with (b) (4) stopper for the cinvanti injectable emulsion was performed. The data from extractables in the solvents (b) (4) tested and toxicological evaluation of potential leachables in (b) (4) indicates that the container closure system is compatible with Cinvanti injectable emulsion drug product, and is assessed to be safe.

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/s/

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